Soil Biology in Agriculture

Proceedings of a workshop on current research into soil biology in agriculture

Tamworth Sustainable Farming Training Centre

11-12 August 2004

Editor: Rebecca Lines-Kelly
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Foreword

The idea for this workshop germinated late last year after a query from an agricultural advisory officer about the use of soil biological products on farms. A few phone calls later, several NSWDPI scientists with an interest in soils met to discuss ways to build soil biology knowledge within the department. As GRDC had initiated investment in soil biology research, we contacted GRDC's Soil Biology Initiative coordinator Greg Bender to see how we might combine forces. Greg was enthusiastic and joined our committee to develop a workshop to showcase current research and knowledge about soil biology, build links between research and extension and obtain feedback from advisors and farmers about future research directions. GRDC provided some funding, both directly and through the Soil Biology Initiative, and NSWDPI organised the workshop for 150 farmers, advisory officers and scientists. However, demand has proved much greater than spaces available, alerting us to the deep fascination that soil biology holds for landholders, advisors and researchers alike. We hope this workshop is the first of many such workshops around NSW and Australia to help us understand the life in our soil and how it contributes to productivity and environmental health.

Organising committee
Greg Bender, leader, GRDC Soil Biology Initiative
Justine Cox, soil scientist, NSWDPI Alstonville
Trevor Gibson, program leader, soils and waste management, NSWDPI Richmond
David Herridge, research scientist, NSWDPI Tamworth
Kaara Klepper, soil scientist, NSWDPI Cowra
Rebecca Lines-Kelly, extension specialist, NSWDPI Wollongbar
Bob Martin, director, Tamworth Agricultural Institute NSWDPI
Greg Reid, soils advisory officer, NSWDPI Wollongbar
Peter Slavich, director, Wollongbar Agricultural Institute NSWDPI

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Our thanks to the team who organised the workshop:

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Kaara Klepper, soil scientist, NSWDPI Cowra
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Bob Martin, director, Tamworth Agricultural Institute, NSWDPI Tamworth
Greg Reid, soils advisory officer, NSWDPI Wollongbar
Peter Slavich, director, Wollongbar Agricultural Institute, NSWDPI Wollongbar

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Understanding soil biota and biological functions: Management of soil biota for improved benefits to crop production and environmental health

VVSR Gupta, David K. Roget
CSIRO Land and Water, Glen Osmond SA

Diversity and populations of soil biota

Soil is one of our most precious non-renewable resources and the soil biota represents a large portion of the earth's biodiversity. Soil organisms (biota) carry out a wide range of ecosystem processes that are essential for crop production, soil resource quality and environmental health in both natural and managed agricultural soils. Production by both crops and pastures is supported and enhanced by soil biological processes. There is a two way relationship between the soil biota and agricultural production. For example, as soil biota play a key role in a number of nutrient transformation processes, crop residues form the essential supply of carbon (energy source) and nutrients for microbial activity.

A diverse, balanced and active soil biota could help provide soil conditions necessary for sustainable crop production, with very little negative environmental effect, through

- increasing microbial activity, carbon turnover and nutrient-supplying potential of soils
- preventing aggressive plant pathogens taking hold and improving plants' ability to withstand disease effects
- supporting populations of plant beneficial microorganisms (eg plant growth promoting rhizobacteria)
- reducing the loss of inorganic fertilisers from erosion and leaching by short-term immobilisation
- stabilising soil structure
- reducing the reliance for agrochemicals and reduced persistence of pesticides in soil and thus lessening off-site impacts.

Soil organisms can be grouped according to their size, morphological characteristics, function and trophic (food) preference. Soil microorganisms are also combined into groups based on their role in specific soil functions (functional groups), irrespective of their taxonomic classification, in order to relate their activities to soil processes. For example nitrifying microorganisms are those that convert ammonia nitrogen into nitrate nitrogen. Soil organisms range in size from microscopic, eg bacteria (two thousandths of a millimetre) to centimetres (earthworms). The four major groups of soil biota, based on their body size, include

- microflora (bacteria, fungi, algae and actinomycetes)
- microfauna (protozoa, nematodes)
- mesofauna (collembola, mites)
- macrofauna (earthworms, beetles, termites).

In addition, soil animals are also classified into various groups based on their principal food source and feeding mode, eg bacterial-feeding, fungal-feeding, plant parasitic or predatory fauna.

Table 1. Some examples of key microbial functions in soil

TYPE OF MICROORGANISM	FUNCTION IN SOIL
Organisms that add nutrients to soil	
Nitrogen-fixing microorganisms Symbiotic N_2-fixing bacteria eg *Rhizobium* and *Bradyrhizobium* species	Fix atmospheric nitrogen in symbiosis with legume plants
Non-symbiotic N_2-fixing bacteria eg *Azospirillum*, *Azotobacter* species	Fix atmospheric nitrogen in bulk soil, near crop residues and in rhizosphere
Organisms that transfer nutrients into plant available forms or facilitate their uptake by plants	
Nitrifying microorganisms eg *Nitrosomonas* and *Nitrobacter* species	Convert ammonia nitrogen into plant available nitrate form
Sulfur-oxidizing microorganisms eg *Thiobacillus thioxidans*, most heterotrophic bacteria and fungi	Convert elemental sulfur and organic sulfur into plant-available sulfates
Mycorrhizae eg Vesicular Arbuscular Mycorrhizae (VAM) (except for crops such as canola)	Facilitate the uptake of phosphorus and zinc by most agricultural crops
Phophorus-solubilising microorganisms (eg *Penicillium* species)	Solubilise plant-unavailable inorganic and organic phosphorus into available forms
Organisms whose action results in the loss of nutrients from soil	
Denitrifying microorganisms eg *Thiobacillus denitrificans*	Convert nitrate nitrogen into nitrogen and nitrous oxide gasses
Sulfur-reducing bacteria eg *Desulfovibrio* species	Reduce sulfate sulfur into hydrogen sulfide gas
Organisms involved in the decomposition of crop residues	
Cellulolytic bacteria and fungi eg *Cellulomonas* species	Decompose cellulose and like compounds in crop residues
Organisms that promote above-ground and/or below-ground plant growth	
Plant growth promoting rhizobacteria eg *Pseudomonas* spp, *Bacillus* spp *Streptomyces* spp	Promote above-ground and/or below-ground plant growth through hormone production or other mechanisms
Organisms involved causing plant diseases	
Rhizoctonia solani, *Pythium ultimum*, *Fusarium* spp, *Verticillium* spp, *Ggt*)	Rhizoctonia barepatch, take-all, damping-off diseases.
Organisms involved in the control of plant diseases*	
Bacteria *Pseudomonas fluorescens, Bacillus subtilis* Fungi *Trichoderma koningi, Fusarium oxysporum* Actinomyces *Streptomyces rimosus*	Control soil-borne plant diseases

* Additional details on commercially available biocontrol products are available at the website http://www.oardc.ohio-state.edu/apsbcc/productlist.htm

Microbial functions

Some examples of the key microbial functions in soil systems are given in Table 1. Soil biota mediated processes can be grouped into two broad categories, single species mediated functions and multiple species/organism mediated functions. For example, symbiotic nitrogen fixation in legumes is carried out by specific partnerships of legume and *Rhizobium* species, whereas a diverse group of bacteria have been shown to perform non-symbiotic nitrogen fixation. Similarly, mineralisation of nutrients (eg nitrogen, sulfur) and decomposition of soil organic matter or crop residues involves the activity of organisms across different trophic groups. Another important point is that the factors (soil, environmental, and food and nutrient) that regulate the activity differ for various functional and trophic groups of biota, thus presence of organisms does not necessarily equate to their effective contribution to ecosystem function.

The successful functioning of many soil biological processes requires a balance of biota interactions in a complex soil biota community (detritus food web). In a detritus food web, organisms across trophic levels are linked, based on the flow of energy and food preference. This approach has been used successfully to understand and model changes in organic matter decomposition (a multiple organism mediated function) following changes in management practices such as tillage and pesticide use. For example, tillage-induced shifts in the fungi:bacteria ratio influences the rate of organic matter decomposition and nutrient availability. Reduced tillage systems support a fungal-based food web (accumulator organisms) whereas conventional tillage systems support a bacterial-based food web. Such an understanding of the importance of microbial community composition does help in the management of soil organic matter and nutrient availability both in agroecosystems and natural environments. A similar approach may be needed to understand the mechanisms behind suppression of plant pathogens in agricultural soils, a major constraint in Australian agriculture and carbon sequestration by agricultural soils.

Habitat and environment

One of the fundamental characteristics of the soil environment is its heterogeneity. Both the population level and composition of soil microbiota and the physico-chemical variables will change considerably over very small distances (eg millimetres). In addition these gradients in space along with the quality of microsites also change over time due to seasonal changes. Our understanding of the seasonal dynamics of various functional groups of soil biota and the ways to regulate their activities to suit a sustainable agricultural system is limited.

Effective populations

In the majority of dryland cropping regions in southern Australia, moisture availability plays a critical role in determining the activity of both microflora and soil fauna. Even though measurable populations of larger soil fauna such as macrofauna are present in most cropping soils, their role in ecosystem functions is determined by the availability of optimum soil and environmental conditions. For example, in the dryland Mallee soils macrofauna rarely play a significant role in ecosystem functions due to lack of optimum soil moisture conditions.

Examples of microbial functions

Carbon and nutrient cycling and nutrient availability

Organic matter in soil is the most important fraction that supports microbial populations, especially the biologically available portion of soil organic matter. Microbial biomass (MB), the living component of soil organic matter, constitutes 2-7% of the organic carbon in soils. Microbial biomass acts as the engine for organic matter turnover and nutrient release. The size of microbial biomass in the surface soil may range from 250 mg C/kg in a sandy soil to 1100 mg C/kg in a clay soil rich in organic matter. Microbial biomass carbon may only represent a small portion of soil organic matter (2-7%), but it is dynamic and living and thus is more sensitive to management practices than total soil organic matter. Microbial biomass is a storehouse of plant-essential nutrients. For example, nitrogen levels in microbial biomass range from 15 kg to 150 kg N/ha. Microbial biomass also holds 5-15 kg of sulfur and 10-45 kg phosphorus per ha. Nutrients held in microbial biomass are not prone to leaching, are tied up only temporarily, and are released for plant uptake as a result of predation by microfauna and the death of microbes during soil drying. It is the interactions between microorganisms and organic matter in the soil that largely determine the fertility and overall quality of the soil. Therefore it is extremely important to use farm management practices that maintain organic matter levels, especially biologically available organic matter, in our soils.

Soil biological function in most Australian cropping soils, particularly southern and western Australian dryland cropping soils, is regulated mainly by the amount of available carbon. Soil microbes require this carbon as a source of energy, in particular the heterotrophic organisms which constitute the majority of soil microbiota. Australian soils are inherently low in biologically available carbon, so carbon inputs have a major influence on soil biological activity. In high-yielding eastern Australian agricultural soils, where the level of carbon inputs is not a constraint, it is the composition of specific functional groups of microorganisms that affect plant growth and production. Plants are the major source of available carbon for biological activity, so soil biodiversity and biological activity depend on the quality and quantity of carbon inputs from plants, through root exudation and above- and below-ground plant residues, and plant-induced changes in soil physical and chemical properties. Pastures are composed of mixtures of plant types (legumes, grasses, C3, C4) so are considered to have a greater potential to influence diverse biological processes. However, the availability of carbon in grazed systems is mediated strongly by grazing management, due to above- and below-ground plant growth in response to grazing.

When considering the nutrient transformations and availability in field situations it is important to account for the implications of spatial scales of biota distribution, seasonal patterns of biological activity, and landscape position for any ecosystem function. These interactions not only influence our perception of biological functions in an ecosystem but also affect our ability to manage them for maximum benefit. In undisturbed natural ecosystems there is generally a high degree of synchronisation between availability of nutrients and plant nutrient need and uptake, which results in low levels of nutrients in soil solution at any one time. However, in most agricultural systems, greater amounts of nutrients flow in and out, and synchronisation of supply with plant requirement is generally not optimum.

Plant growth promotion

Rhizosphere microorganisms have one or more specific associations with plants that influence plant growth. The rhizosphere microbial community is generally metabolically highly active and associated with specific plant types. Among the many mechanisms associated with plant-rhizosphere microorganism interactions, the production of biologically active metabolites is one of the most important ways that rhizosphere microbiota influence plant growth.

Suppression / biocontrol

Disease suppressiveness of soil is the ability of a soil to suppress disease severity even in the presence of a pathogen, host plant and favourable climatic conditions. The different types of disease suppression mechanisms are related to the establishment of the pathogen, reduced parasitic activity of the pathogen and the level of disease incidence or severity. For the disease suppression known as 'general suppression', the inhibition of pathogenic populations is related to either the activity of the total microflora or diverse microbial-faunal interactions. The 'specific suppression phenomenon' has been attributed to the activity of specific microbial groups (antagonists). Some abiotic factors of soil such as pH and clay content have also been attributed to certain types of disease suppressiveness such as fusarium wilts. The increase in disease suppression against a range of cereal root diseases, documented at Avon SA, was related to increased soil carbon inputs from stubble and roots derived from higher yielding crops and greater cropping frequency. Increased carbon inputs result in changes to the composition and activity of the soil microbial community over time. These changes result in greater competition for soil resources that, along with predation and inhibition of pathogens, lead to increased suppression of many soil-borne fungal diseases. All soils have the ability to suppress soil-borne root diseases to some extent through the activity of soil microbes, so disease suppression is not an absolute characteristic but a continuum from highly suppressive soils to poorly suppressive (disease conducive) soils. However, the opportunity to improve suppressive activity to an effective level may be limited by the time required for changes in microbial diversity to occur. These changes depend on the management practices that affect microbial growth, composition and metabolic status specific to different soils, and environmental conditions.

While a variety of bacteria and fungi have been found to be effective biocontrol agents against different crop diseases based on controlled environment experiments, no single microbial inoculant has yet been successful under Australian broadacre agriculture systems. Briefly, for an introduced biocontrol agent to be effective it must first survive, establish, acclimatise and grow in field soils and interact with the pathogenic organisms alone or in the presence of plant. Soil and environmental conditions are generally harsh and not optimal for introduced microbes in Australian cropping systems which have carbon-poor soils, short-periods of warm, moist soil, and long dry, hot periods. Unlike the rhizosphere-based inoculants, endophytes escape problems associated with survival in the harsh and carbon poor soil. Research on the field-based ecological aspects of inoculants has been very limited.

Management impacts

The various management practices involved in grain and pasture crop production have an impact on either the populations of different groups of beneficial and deleterious biota and/or their activities in all soil types and cropping regions. Management practices affect soil biota by

- direct effects on the populations
- changes in the soil habitat (eg physical and chemical properties), microclimate and carbon (energy) sources
- influences on above ground productivity and community composition.

Crop rotation influences soil biota directly by choice of plant types and indirectly by associated agronomic practices. Some of the agronomic practices that impact on populations of biota include

- stubble retention
- tillage
- application of herbicides
- insecticide and fungicide application
- application of manures or fertilisers
- application of chemical amendments or waste products.

A number of papers have been published describing the effects of management practices on soil biota even for Australian conditions.

Agrochemicals

Herbicide use is a vital component of modern agriculture, in particular under reduced till systems. With increased adoption of stubble retention and reduced till practices and the introduction of new herbicides, herbicide use will remain an essential practice in the near future. Non-target effects of herbicides on soil biological activities may cause undesirable effects on essential transformation processes such as reduced nitrification and nitrogen mineralisation, or result in unexpected damage to crops through increased diseased incidence. Non-target effects of herbicides could be either positive or negative. If herbicide application is to remain a viable practice in sustainable farming systems, evaluation of herbicide effects, especially from repeated and long-term use, is essential to ensure optimum nutrient availability and plant growth. Results from laboratory, glasshouse and field experiments conducted over three years in South Australia and Victoria indicate that not all herbicides have negative effects on soil biota and biological processes.

Some of the implications and recommendations for using herbicides based on our results and the related information available in literature are listed below.

- It is essential to use an integrated approach of testing key groups of biota and associated activities in order to evaluate and predict unforeseen non-target effects and effects from repeated and long-term use of herbicides. Effects of each herbicide need to be considered separately.
- The short-term impacts of most of the herbicides we tested are reversible, so it may be possible to develop management options to reduce non-target negative impacts.
- Landholders need to modify management practices to avoid application of specific herbicides.
- An appropriate recovery period for soil biota should be allowed between herbicide applications.
- Soils with a healthy biota could recover from short-term negative effects of herbicide application. Appropriate use of herbicides could be less destructive to soil biota if management practices that improve biological activity are promoted.

Methods to measure/study biota and biological functions in soils

The study of biological processes and population dynamics involve the use of one or more of microscopic, culture, biochemical, isotopic or molecular techniques. Modern biochemical and molecular techniques allow a more accurate identification of biota species and also offer the potential to integrate the roles of different species or groups of organisms based on their involvement in specific functions. No single technique can provide enough information for prediction and management of biological processes. Developing methods for the measurement of microbiological processes *in situ* is one of the key requirements to identify regulating factors.

References

Abbott LK, Murphy DV 2003. Soil biological fertility: A key to sustainable land use in agriculture. Kluwer Academic Publishers, Dordrecht.

Alef K, Nannipieri P Eds 1995. Methods in applied soil microbiology and biochemistry. Academic Press, New York.

Coleman DC, Crossley Jr DA 1996. Fundamentals of soil ecology. Academic Press, New York.

Gupta VVSR, Roper MM, Roget DK 2002. Potential for non-symbiotic N_2-fixation in different agroecological zones of southern Australia. Symposium papers of the 17th World Congress of Soil Science, Bangkok, Thailand. Symposium 12, Paper 1553, pp 1-10.

Gupta VVSR, Sivasithamparam K 2002. Relevance of plant pathogens to soil biological fertility. In Soil biological fertility: A key to sustainable land use in agriculture Eds Abbot LK, Murphy DV pp 163-186. Kluwer Academic Publishers, New York.

Gupta VVSR, Yeates GW 1997. Soil microfauna as indicators of soil health. In Biological indicators of soil health Eds Pankhurst CE, Doube BM, Gupta VVSR pp 419-435. CAB International, Wallingford UK.

Roget DK 1995. Decline of root rot (*Rhizoctonia solani* AG-8) in wheat in a tillage and rotation experiment at Avon, South Australia. Australian Journal of Experimental Agriculture 35 1009-13.

Roget DK, Coppi JA, Herdina, Gupta VVSR 1999. Assessment of suppression to *Rhizoctonia solani* in a range of soils across SE Australia. In Proceedings 1st Australasian soil-borne disease symposium Gold Coast Qld.

Contact details

Dr VVSR Gupta
Senior Research Scientist
CSIRO Land & Water
PMB 2 Glen Osmond SA 5064
Phone 08 8303 8579
Fax 08 8303 8550
Email Gupta.Vadakattu@csiro.au

Overview of 'soil biology' tests

Lyn Abbott and Dan Murphy
University of Western Australia

Introduction

Soil tests have been routinely conducted by farmers and researchers to determine constraints to productivity in agricultural systems and to refine recommendations for nutrient inputs and other soil amendments such as lime. Traditionally, these tests have concerned soil chemical constraints, with a lesser emphasis on evaluation of soil physical constraints (Walker, Reuter 1996). However, there is increasing interest in understanding changes in soil biological processes to assess the impact of land management practices on soil conditions and to implement soil testing programs that focus on soil biological fertility.

The use of the term 'soil fertility' without reference to its components has lead to a primary focus on soil chemical fertility (Abbott, Murphy 2003). It is more difficult to interpret soil tests in relation to soil biological fertility. However, when soil biological fertility is considered in addition to soil physical and chemical fertility, a completely different dimension is opened up for interpretation. For example, some soil biological processes alter aspects of soil chemical fertility, but is this taken into account when soil chemical tests are interpreted? How can they be taken into account in practical terms? Previously, this has been shown to be difficult (Walker, Reuter 1996) because the great diversity, inter-relationship and dynamism of living components of soil lead to difficulties in knowing what and when to measure and knowing how to interpret what is measured. 'Soil biology' tests are also relatively expensive.

Tests associated with soil biological characteristics can involve quantification of whole organisms (eg the total microbial biomass or the abundance of specific groups of soil organisms - fungi, bacteria and fauna) or quantification of specific activities of organisms (eg an enzyme activity, respiration of organisms (measured as CO_2) or the 'potential activity' of soil organisms (as can be assessed after adding known carbon sources to soil). Other tests involve detection of specific genes using molecular probes or identifying specific components of organisms (eg fatty acids or elemental composition such as nitrogen, phosphorus and sulfur). Tests for various carbon substrates (eg different organic matter fractions) are also relevant because they are a source of carbon and energy for many groups of organisms in soil so their presence, accessibility and relative abundance partly controls microbial activity. Numerous tests relevant to soil biological fertility have been summarised comprehensively by Pankhurst et al (1997) (see their Table 17.2). In most cases, these tests are not routinely used by farmers to build an understanding of the biological state of soil in parallel with their knowledge of the soil's chemical and physical status for identifying constraints to plant growth.

Biological fertility of soil depends closely on the soil type and how it is managed (Abbott, Murphy 2003). However, it is not as easy to define the biological state of soil as it is to define the chemical and physical state of soil, even though each depends on soil type and land use history. Of course, some agricultural soils have a higher level of chemical fertility due to nutrient inputs than the soil under its original vegetation. The reverse may apply to the physical state of soil due to disturbance and loss of an environment suitable for soil fauna (Abbott, Parker 1980). There is no 'ideal' biological

state of soil because what is required depends on the planned land use as well as the soil type, climate and demands of the management system. Definitions need to be placed in this context. In addition, the balance between chemical and biological contributions to soil fertility needs to be taken into account because increasing reliance on chemical inputs decreases contributions from biological processes (Abbott, Murphy 2003).

Soils have a high diversity of organisms irrespective of their land use. Agricultural practices may increase biological activity associated with degradation of large quantities of plant residue so greater abundance of organisms may be present periodically in agricultural soils than under the original vegetation. In contrast, agricultural practices generally decrease the residual level of carbon in soil and this may be reflected in a decrease in 'biological fertility'. Overall, it is not possible to say that a certain amount or type of organisms within a community leads to a 'biologically fertile' soil. Benchmarks are possible for some components of soil biological fertility but, as indicated above, they will change with climatic conditions, soil type (eg clay content) and farming system. Therefore, tests related to soil biological fertility need to be interpreted in the context of specific questions such as 'If a wheat crop is grown in this soil this year, is it likely to suffer from a fungal disease?' This question can be investigated. Similarly, other questions might be 'If a legume is grown in this soil, will it nodulate?, or 'If a rotation of wheat/lupin/lupin/wheat is included in this farming system, will there be a high level of mycorrhizal fungi in the following wheat crop?' or 'If a crop is managed by minimum tillage at this site, will microbial biomass be sufficient to supply 50% of its nitrogen?'. Questions like this have both specific and general aspects. Knowledge of soil biological characteristics can be combined with knowledge of the physical, chemical, climatic and economic constraints to derive some answers. Long-term questions about the sustainability of a farming system can also be addressed through understanding of soil biological fertility. This is because biological characteristics change more rapidly than chemical characteristics (such as %C) and can therefore be used to predict impacts more effectively than can chemical soil tests.

The heterogeneity of soil is well known, and as this is the habitat of soil organisms, it is understandable that there is a great variation in distribution and abundance of soil organisms both in space (vertical and horizontal) and time. Therefore, appropriate sampling procedures are of great importance when testing soil for biological activity. In addition, the treatment of soil after sampling and prior to analysis is particularly important for soil biological characteristics because they can be affected greatly by temperature, aeration and moisture. Sampling for some biological assays is best done when soil is collected in a dry state, but this is not always practical. For any soil biological analysis, the impact of topographical, spatial, seasonal and management practice effect needs to be considered carefully. Figure 1 illustrates the heterogeneity of microbial biomass across a whole farm measured on a 150m grid. It shows a relationship with landscape parameters, in which microbial biomass changed more with elevation than with land use (Figure 1, Djuuna et al, personal communication).

Due to the heterogeneity of the soil environment, the number of soil samples taken within an area can affect the mean value of soil biological characteristics. For example, on the farm at Wickapin, Western Australia (Figure 1), the number of samples required to estimate the microbial biomass and mycorrhizal infectivity at a paddock scale was not the same in any of the 14 paddocks. The number of samples required differed between paddocks for both of these biological assays (Djuuna, pers. comm). Bulking of samples is possible, but soil mixing can affect some biological attributes, therefore,

caution is required. Soil drying after sampling is also an issue to be considered, especially in terms of the relationship between the biological soil tests and what they represent on the farm. Thus, the 'actual' biological state of soil may not be very well represented by one or two soil samples. The dynamic nature of soil biological processes also affects how a soil test conducted at one point in time is interpreted. Soil biological tests have to be interpreted in the broader context of how they change with time.

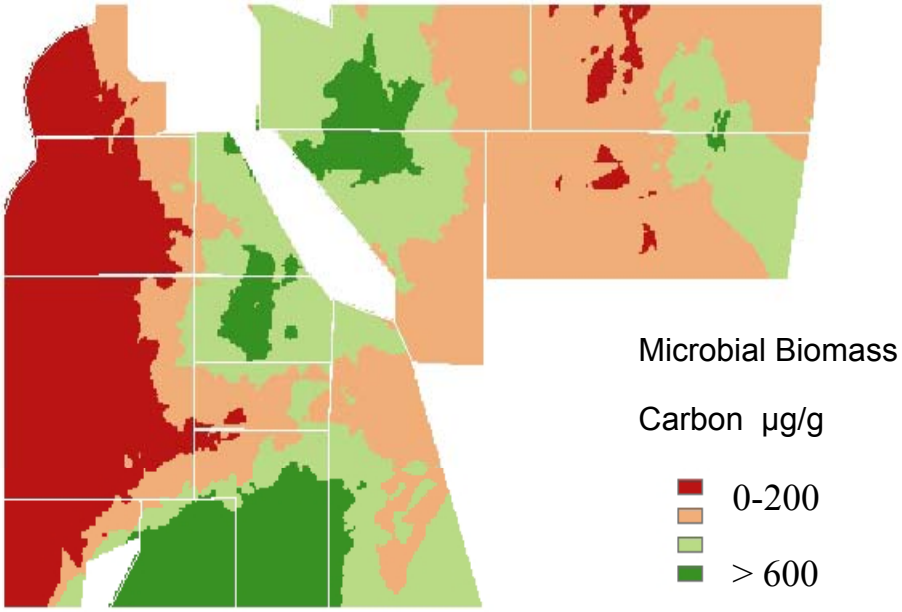


Figure 1. Heterogeneity of microbial biomass carbon for a farm at Wickepin, Western Australia assessed on a 150m grid. Soil was sampled in summer during the dry season of the prevailing Mediterranean climate in this region (Djuuna et al, unpublished data).

What can be measured?

There are many types of soil tests suitable for quantifying soil organisms or soil biological activity. Some of the analyses are relatively straight forward, whereas others are highly complex. Therefore, greater caution is required when interpreting soil biological tests than is necessary for most soil chemical and physical characteristics. A summary of some soil biological tests and comments about them is included in Table 1.

Interpreting 'soil biology' tests

Critical values for 'soil biology' tests need to be developed in relation to climate, soil type and land use. Broad generalisations can be made (eg for some disease-causing organisms and for rhizobia). However, generalisations for other assays remain to be assessed over a wide range of soils, climates and farming systems within Australia. An alternative approach to the use of critical values is the role of soil biological tests in tracking changes (monitoring) in selected characteristics in relation to management practice. Consideration of sampling strategy, time of sampling as well as depth and position is important. New developments with mid infra-red technology (MIR) may be useful in overcoming the tedious nature of soil testing (see Murphy, Milton, this proceedings).

Table 1. Examples of tests for biological components of soil and comments about the methodology (see also Pankhurst et al 1997). Methods can be by observation (ie direct) or by inference (indirect) based on assessment of products of reactions or other functional attributes.

MICROBIAL BIOMASS MEASUREMENTS	**Organisms can be assessed without first separating them into specific groups, but the identity of individuals making up the microbial biomass is not determined by these methods. Relevance: Indication of the amount of organisms in soil Ease of measurement: Relatively straight forward**
Total soil microbial biomass (or microbial carbon (C), nitrogen (N), phosphorus (P), sulfur (S) etc)	Microbial biomass in soil can be measured by fumigation-incubation, fumigation-extraction and substrate-induced respiration methods. Fumigation methods involve killing the microbial biomass then extracting released nutrients such as nitrogen. Estimates of microbial biomass are determined according to known proportions of elements in soil organisms. Methodological problems associated with applying these methods to different soil types and at different times of the year have been extensively researched and the practical aspects are well understood, but tests need to be conducted with caution. If roots and larger animals are removed from the soil prior to assessment, microbial biomass will include mainly microorganisms and smaller soil fauna (eg mites and springtails). This methodology allows estimation of the amount of C, N, P and S in living soil organic matter.
Fungal counts	Direct: Measurement of length of hyphae (km/g soil) is possible but it is not usually possible to identify the fungi present. Indirect: Some fungi can be grown on artificial nutrient media but this represents only 1-5% of the total organisms present. Quantification of some important fungal pathogens.
Bacterial counts	Direct: It is possible to estimate the number of bacteria in soil, but this is a very rough estimate. As bacteria occur in small pores within the soil, many are not easily extracted. Bottomley and Maggard (1990) showed that if bacteria were 'washed' from soil, then the soil was again, a similar number of bacteria appeared in the second wash. Therefore, caution is required in interpreting these numbers. Another problem with direct counts of bacteria is that it is not possible to distinguish between living and dead bacteria on the microscope slides (but techniques are available to address this question (Bottomley, Maggard 1990)). Indirect: Although many soil bacteria will grow on agar or in nutrient broth, only a small proportion can do so, therefore indirect counts of bacteria based on this type of methodology are of little relevance to the number of bacteria in soil.
ASSESSMENT OF GROUPS OF ORGANISMS	**Organisms in soil can be assessed in groups (eg mites or earthworms can be counted) or as number per group (eg as genera or species). For bacteria and fungi, special techniques can be used for particular groups: eg serological tests or molecular tests are available for some bacteria (eg rhizobia). Relevance: Some are potential indicators of the soil food chain Ease of measurement: Variable**
Protozoa	Indirect: This is a tedious method. The total number is deceiving because it reflects multiplication (which depends on the availability of food such as bacteria) and predation (eaten by larger organisms).

Nematodes	Direct: Important for assessing presence of excessive numbers of plant pathogenic nematodes. Important for identification of balance between beneficial and detrimental nematodes. Indirect: DNA probes are available for some nematodes.
Termites	Direct: Easily quantified and could be an indicator of soil health in some agricultural environments if calibrated.
Enchytraeids	Direct: Could be an indicator of soil health in some agricultural environments if calibrated.
Earthworms	Direct: Could be an indicator of soil health, but this is disputed because species differ between soils. Can be calibrated locally.
Microarthropods	Direct: Counts can be included in diversity indices.
Rhizobia	Direct: Isolation and identification is possible (eg see Bottomley, Maggard 1990) or from nodules on field plants. Indirect: Isolation and identification from plant bioassays.
Specific 'beneficial' fungi or bacteria (including rhizobacteria)	Indirect: Molecular markers can be used. Important for research purposes.
Abruscular mycorrhizal fungi	Direct: Arbuscular mycorrhizal fungi can be assessed by directly scoring colonisation of roots using a microscope. Indirect: Bioassays using a standard bait plant can detect infective hyphae present in the soil at a particular point in time.
Plant pathogens	Direct: Root or leaf disease assessments. Indirect: Molecular markers can be applied directly to soil or plants for some pathogens. Indirect: Plant bioassays are easy to establish for some pathogens. Tests can be calibrated as indicators of potential for plant disease (eg DNA tests, bioassays, root scores, disease rating).
SOIL BIODIVERSITY	**Expanding opportunities are being made available for measurement of soil biodiversity, especially with the development of molecular tools. Caution is still required in interpreting the data from these methods.** **Relevance: Potential indicators of biological state of soil.** **Ease of measurement: Difficult, especially interpretation.**
Species richness indices	Indirect: Various forms of indices are available, but caution is required in their use because they are not all statistically sound.
Biolog™ Tests	Indirect: This assesses a component of the microbial community.
Fatty acids	Indirect: Used to compare a fraction of cell components.
DGGE and related tests	Indirect: Diversity in the DNA banding pattern of the microbial population in soil may reflect genetic diversity.
Microarray	Indirect: Rapidly developing molecular technology for simultaneously detecting large numbers of organisms in soil.
CARBON-BASED FRACTIONS (includes soil microbial biomass – see above)	**Fractions of organic matter in soil represent pools of nutrients with different relative availabilities.** **Relevance: Potential indicators of biological state of soil.** **Ease of measurement: Relatively easy.**
Mineralisable nitrogen	Indirect: Readily available pool of nitrogen

Carbon fractions (including relative availability)	Indirect: Carbon fractions affect the physical habitat of soil organisms. Some fractions may be readily degradable by soil organisms but they may be located inside soil aggregates and are inaccessible/protected from breakdown by soil organisms.
Carbon Management Index	Indirect: Calculates the change in a treatment soil compared with a benchmark soil for labile organic matter (Blair et al 1995). Result is dependent on defining an adequate reference soil.
MICROBIAL PROCESSES	**Quantification of biological processes can give an indication of the activity of soil organisms. This may be more relevant than the abundance of organisms for some purposes, therefore both abundance and activity measurements of soil organisms are required.** **Relevance: Potential indicators of biological state of soil and/or soil chemical fertility.** **Ease of measurement: Relatively easy.**
Enzyme activity	Indirect: eg cellulase activity can be assessed indirectly using the cotton strip assay or by biochemical means.
Mineralisation (including respiration)	Indirect: eg ammonium released during mineralisation of organic matter can be measured but the amount of ammonium converted to nitrate must also be taken into account.
Nitrification	Indirect: eg nitrate can be measured as an indication of the activity of nitrifying bacteria, but loss of nitrate by leaching needs to be taken into account or the activity of the nitrifying organisms will be underestimated.
Methane production	Indirect: Methane production by soil organisms can be measured.
Denitrification	Indirect: Loss of nitrogen through denitrification is measured.
Substrate Induced Respiration (SIR)	Indirect: The 'potential activity' of soil organisms can be assessed by adding a relatively easily used carbon source (a sugar) and the amount of carbon dioxide released is measured.
Catabolic activity	Indirect: Various carbon substrates can be added to soil and respiration assessed to indicate the catabolic diversity of the organisms present (Degens, Harris 1997).
FUNGAL/BACTERIAL RATIOS	**Some management practices can change the relative abundance of fungi and bacteria in soil, so there is potential to use this as an indication of the impact of management practice on soil biological activity.** **Relevance: Potential indicators of biological state of soil.** **Ease of measurement: Difficult to interpret.**
Fungal bacterial ratio (direct count method)	Direct: Fungi and bacteria can be directly assessed (see above) and the ratio of their abundance calculated.
Fungal bacterial ratio (PLFA method)	Indirect: This method uses biochemical tests of fungi and bacteria (fatty acid analysis) as a basis for estimating the proportion of fungi and bacteria in soil (see above for fatty acids).
Fungal bacterial ratio (SIR method)	Indirect: This method assesses the ratio of fungi and bacteria in soil based on response to addition of carbon substrates (see SIR method above). It is based on inhibition of fungi and bacteria in separate assays and inhibition of all biological activity as a control which is difficult to achieve across different soils.

References

Abbott IJ, Parker CA 1980. Agriculture and the abundance of soil animals in the Western Australian wheatbelt. Soil Biology & Biochemistry 12 455-459.

Abbott LK, Murphy DV 2003. What is soil biological fertility? In Soil biological fertility: A key to sustainable land use in agriculture. Eds Abbott LK, Murphy DV pp 1-15. Kluwer Academic Publishers, Dordrecht.

Blair GJ, Lefroy RDB, Lisle L 1995. Soil carbon fractions based on their degree of oxidation and the development of a carbon management index for agricultural systems. Australian Journal of Soil Research 46 1459-1466.

Bottomley J, Maggard SP 1990. Determination of viability within serotypes of a soil population of *Rhizobium leguminosarum bv. trifolii*. Applied & Environmental Microbiology 56 533-540.

Degens BP, Harris JA 1997. Development of a physiological approach to measuring the metabolic diversity of soil microbial communities. Soil Biology & Biochemistry 29 1309-1320.

Pankhurst CE, Doube BM, Gupta VVSR 1997. Biological indicators of soil health: synthesis. In Biological indicators of soil health. Eds Pankhurst CE, Doube BM, Gupta VVSR pp 419-435. CAB International, Wallingford UK.

Walker J, Reuter DJ 1996. Indicators of catchment health: A technical perspective. CSIRO Publishing, Collingwood.

Contact details

Professor Lyn Abbott
Head, School of Earth and Geographical Sciences
The University of Western Australia
35 Stirling Highway Crawley WA 6009
Phone 08 6488 2499
Fax 08 6488 1050
Email labbott@cyllene.uwa.edu.au

The GRDC Soil Biology Initiative

Greg Bender
GRDC Canberra

Introduction

The establishment of a national Soil Biology Initiative for carefully targeted and coordinated research is a crucial opportunity for GRDC investment to support grain producers across all regions. Soil-borne constraints to crop performance represent a major impediment to the continued health and strength of the grains industries in Australia. These are not likely to be understood and overcome quickly without significant investment in research and development. To address this gap in research, the GRDC has invested $10 million in a five year Soil Biology Initiative from 2002 to 2007.

Purpose of the Initiative

Improvements in 'above-ground' grain cropping technologies, such as time of sowing, equipment efficiency, seed and fertiliser placement, weed management, and improved varieties, have all contributed to increasing productivity. As the constraints to production associated with these factors have been progressively tackled and improvements made, attention has increasingly focused on the serious below-ground limitations to crop performance. Poor nutrient availability and uptake, limited use of soil water, and sub-soils hostile to root growth due to acidity, salt or sodicity, have all been identified as important factors limiting yields and profitability. It has become clear that the yield potential of existing varieties is not being achieved under field conditions. Many of the primary limiting factors are related to biological interactions between crop roots, soil microorganisms and the physical and chemical characteristics of the soil medium.

In this situation, where the interaction between varietal genotype and the soil environment is a primary limitation on crop performance, there are two clear research opportunities:

- re-engineer varietal genotypes so that they are better suited to Australian environments, including the particular characteristics of Australian soils
- through agronomic practice and other means, amend the soil environment to better suit the genotypes available.

GRDC has a significant investment addressing different aspects of genotype improvement. Current investment in R&D to improve and better manage the soil environment for genotypic expression is much lower. However, there is already evidence that better management of the soil environment and of crop-soil interaction can provide substantial increases in yield and profitability. The purpose of the Soil Biology Initiative is to directly address the opportunity to bring about a substantial increase in the economic performance of grain farms and of producers.

The stated goal of the Initiative is to develop a suite of practical methods and cost-effective products, based on a sound scientific understanding of crop root-soil interactions, which will assist in overcoming soil-based limits to crop performance and significantly improve profit margins in grain cropping systems.

Primary research outputs

The primary research outputs from the Initiative will be:

- identification and evaluation of soil-based constraints to crop production, and assessment and ranking of commercial opportunities for new agronomic practices and market products
- significantly improved understanding of critical processes, pathways and controlling factors governing the interactions between the soil matrix, microorganisms and crop roots
- increased capacity within Australia to undertake soil biology R&D in support of the grains industries, with a focus on multidisciplinary teams and effective collaboration between public sector and commercial organisations
- new agronomic practices and commercial products developed, tested and validated for practical and economic performance under field conditions, and provided to the industry in forms that can be readily adapted by growers to suit their particular situations.

Structure of the Soil Biology Initiative

The Soil Biology Initiative has been structured around five major research themes:

- Theme 1: Development of prospective microbial actives to improve crop nutrition and to control pests and diseases
- Theme 2: Control of soil-borne plant pathogens
- Theme 3: Improving nutrient availability and balance
- Theme 4: Interaction between soil physics, chemistry and biology
- Theme 5: Analysis of available data to relate agronomic practice, soil biota and crop performance.

For each theme, the starting point is the analysis of known constraints to crop production and an evaluation of market opportunities, including improved agronomic practice and development of specific products. These analyses have been undertaken, in collaboration with industry and researchers, to set priority projects for each theme. Each theme has four stages of development:

- Stage 1: Identification, assessment and ranking of investment opportunities
- Stage 2: Research to identify and quantify critical processes, pathways and controlling factors that affect crop performance through soil biological, physical and chemical interactions
- Stage 3: Conceptual development of new practices and products to overcome soil-borne limitations to crop performance, and proof of concept through laboratory, glasshouse and field trials
- Stage 4: Completion of agreements for communication, extension and commercialisation of theme outputs, leading to efficient product delivery to industry.

Work in the different themes, and projects within a single theme, move through these four stages at different speeds. For example, a number of potential products already at a pre-commercial stage will be further developed and tested in Theme 1, and should be ready for commercial release by the completion of the Soil Biology Initiative. In contrast, the focus in Theme 4 will initially be on strategic research, and it is likely to be

some time before research in this theme moves into proof of concept or demonstration stages.

Initiative strategies run across the five themes:

- A specific strategy is to expand the research capability within Australia to undertake high quality soil biology R&D in support of the grains industries. The Soil Biology Initiative supports a number of postgraduate and postdoctoral positions, with the specific objective of attracting and retaining in this area of research high quality R&D staff. This requires negotiation of commitment by research organisations to support soil biology R&D for the grains industries.
- An essential strategy is to incorporate commercial expertise at an early stage of project development wherever appropriate. Market and commercial skills are used to assist the initial identification and assessment of research opportunities and ranking of project priorities. This strategy also includes developing a mix of public sector research and commercial skills in all themes and most projects.
- Participation by all sectors of the grains industry in the Initiative is also a crucial strategy. Growers, farmer groups, district agronomists, and GRDC Panel members are involved in the initial assessment and ranking of priorities within each theme. The program coordinator is responsible for establishing early on, and maintaining, channels for two-way flow of ideas and information between the reearchers, producers and other industry participants. The aim of this strategy is to ensure that research priorities are closely attuned to industry needs, and that the industry is fully aware of, and actively participates in, the Soil Biology Initiative.

Conclusion

Soil biology and soil health are popular subjects with farmers. However, farmers are receiving mixed messages on products and practices in the market and there is very little in the way of solid field data relevant to the needs of their particular regions. In an effort to build a credible scientific foundation for soil biology, along with some practical products and farm management practices, the GRDC and its research partners have been trying to put the pieces together in what is a very challenging science. With the GRDC Soil Biology Initiative established and approaching its third year, we look forward to the development of this branch of science and its application for the benefit of farmers and broadacre agriculture in Australia.

Contact details

Greg Bender
Program Coordinator
Soil Biology Initiative
Phone 02 6248 0165
Email greg.bender@netspeed.com.au

Impact of management practices on activity of soil biota and productivity constraints in Vertosols of the northern grains region

Mike Bell[1], Nikki Seymour[2], Graham Stirling[3], Lukas Van Zwieten[4], Graeme Sutton[5] and Phil Moody[6]

[1]Queensland DPIF Kingaroy [2]Queensland DPIF Toowoomba

[3] Biological Crop Protection Pty Ltd, Moggill Qld

[4] NSWDPI Wollongbar [5] Landmark Pty Ltd, Dalby Qld

[6] Queensland DNR Indooroopilly

Introduction

The Vertosols represent the predominant soil type supporting the broadacre grain and cotton industries in the northern grains region. They are characterised by relatively high plant available water holding capacity (PAWC: >20mm plant available water/10cm profile depth–Webb et al 1997) and, in their virgin state, moderate to high levels of chemical fertility. However sustained cropping, initially using conventional tillage but in recent years employing reduced or zero tillage practices, has resulted in a decline in soil chemical fertility so that chemical fertiliser applications (especially nitrogen and phosphorus) are necessary to sustain productivity (Dalal, Probert, 1997).

Farming systems vary significantly across the region. In the higher/more reliable rainfall areas of the east, cropping is the dominant land use. Further west, mixed cropping and grazing enterprises are more common, with cropland rotated with grazed pasture phases. The dominant crops in the region are sorghum and cotton (summer) and wheat and chickpeas (winter), with barley, mungbeans and maize also important in some areas. This variety of crop options leads to many varied crop rotations across the region.

The predominant factor limiting crop productivity is water, with extremely variable annual rainfall contributing to a cropping sequence that becomes increasingly opportunistic in more western and northern areas (Freebairn et al 1997). Growing seasons are often characterised by long periods without rain, with crops solely reliant on subsoil moisture reserves. Fallowing is used to replenish soil water reserves between crops, with 12-18 month bare fallows common in the region. Long (12-15 month) fallows are also commonly used during the transition from summer to winter crops in the rotation (eg sorghum harvested in autumn, with wheat sown in winter the following year). The widespread adoption of conservation tillage and direct-drill methods have increased both the crop frequency and system productivity due to improved efficiency of water capture and use by crops.

Research has been conducted in the northern region on specific functional components of the soil biota, particularly on soil-borne diseases, plant-parasitic nematodes and on the importance of mycorrhizae in the cropping system (Wildermuth et al 1997). Less work has been done on various components of the detritus food web that can potentially moderate the impact of these functional components on crops in the farming system. This paper reports preliminary findings from a current research project that aims to quantify the impact of various management practices on soil biota in these land use systems, and to relate these impacts to crop productivity and system performance.

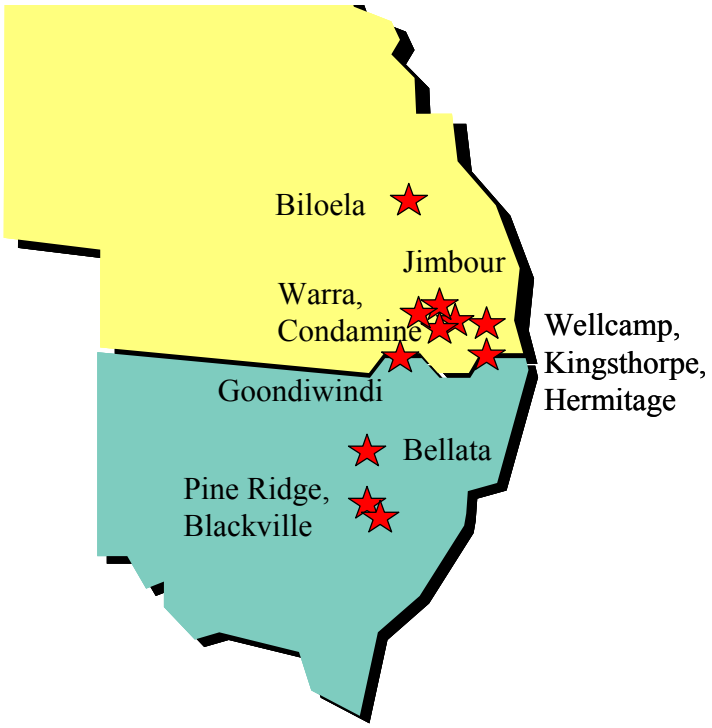
Materials and methods

Experimental sites

Experimental sites have been chosen for a variety of measurements of both soil biota and crop performance across the region, from Biloela in the north to Pine Ridge in the southern Liverpool Plains, and west to Goondiwindi (Figure 1). Given the predominance of chickpea, wheat and sorghum in the cropping system, sites that were growing these crops as part of the crop rotation were preferentially selected, while these species were also used in subsequent field and glasshouse studies.

The sites were chosen to represent long-term contrasts in management (ie paired sites in adjoining fields, or in controlled experiments), to represent contrasting farming systems (eg ley pasture phases versus continuous cropping) or as representative sites on which to benchmark system performance in a district or region. In addition to sampling existing sites and experiments, the project has also established a more controlled experiment to examine the impact of fallow length and crop rotation on soil biota and any biotic constraints to crop performance.

Figure 1. Locations at which soils have been collected or research sites established in the northern soil biology project.



Assessment of microbial diversity and chemical fertility in response to management and organic amendments

Stratified soil samples were collected from the 0-5 cm, 5-15 cm and, in some cases, the 15-30 cm layers of the soil profile, with a number of representative samples collected from each plot (in the case of replicated field experiments) or at multiple sites (generally five) within a commercial field. While important differences in populations of soil biota can occur at greater depths in the soil profile (eg lesion nematodes, Peck et al 1993), it was assumed that the most significant impacts of management on soil biota would be detected in the upper layers.

Samples were homogenised and assessed either 'as is', or after amendment with a range of contrasting organic materials (feedlot manure, grass or legume plant material). In the case of the organic amendments, samples were moistened periodically for a period of six months to allow equilibration of the microbial communities before subsequent assessment. All samples were then separated into sub-samples for microbial or chemical analysis, or used in glasshouse bioassays.

Microbial analyses included determination of microbial biomass carbon, microbial activity using FDA, total DNA, nematode community analysis and mycorrhizal spore levels. A smaller subset of samples was assessed for root pathogens by the SARDI Root Disease Testing Service and for the composition of the microbial community using PL-FAME. Soil chemical properties included total and labile fractions of soil organic carbon, in addition to more traditional measurements of chemical fertility.

Crop responses to soil pasteurisation and biocide applications

Initial glasshouse bioassays were conducted using soil samples with or without pre-treatment of steam pasteurisation (60^{o}C for 60 minutes), to assess potential biotic constraints in different regions and cropping systems. After pre-treatment, soils were potted with levels of nutrients designed to provide luxury levels of chemical fertility. Pots were placed on benches equipped with self-regulated watering systems and bioassay plants (sorghum cv MR Buster, wheat cv Hartog and chickpea cv Jimbour) were grown for eight weeks. Dry weights of tops and roots, root health, root length and mycorrhizal colonisation levels have been recorded.

In order to more closely examine the key components of soil biota associated with pasteurisation responses and suppression of lesion nematodes, a second series of experiments with treatments comprising a range of specific biocides (fungicides, nematicides and antibiotics, in addition to more general biocides like irradiation and pasteurisation) are being conducted.

To confirm the relevance of the glasshouse bioassays as predictors of biotic constraints in field-grown crops, field fumigation assays using a commercial fumigation rig to apply methyl bromide at rates of 1000 kg/ha are being undertaken on sites in the Jimbour-McAllister and Pine Ridge districts. Soil from these sites is simultaneously being used in additional glasshouse bioassays using both disturbed and undisturbed soil samples to assess response to pasteurisation, irradiation or more specific biocides.

Assays for suppression of lesion nematode

Soil from a subset of sites with contrasting managements (eg continuous crop v ley pastures) has been used to assess the impact of management on suppression of lesion nematodes in glasshouse experiments with wheat. Again, a range of biocides is also being used to help identify key components of soil biota responsible for reductions in pathogen reproduction and/or pathogenicity.

Benchmarking the performance of field crops

A number of crops, primarily in commercial fields, have been benchmarked against soil fertility and seasonal conditions using APSIM. Soils have been extensively characterised for PAWC, soil fertility at planting and seasonal rainfall and water use, with differences between simulated (potential) and actual yields being assessed as a potential measure of biotic constraints. Soil from these sites is also used in other assessments of biotic constraints (eg glasshouse bioassays).

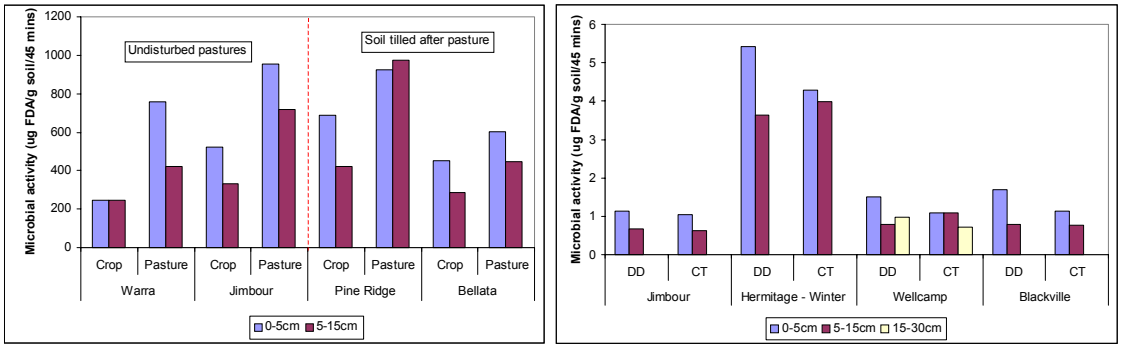
Results and discussion

Impact of management and soil amendments on soil biota

There have been some clear and consistent effects of management on soil biota (both amount and diversity) across the region.

- There were clear indications of a reduction in overall microbial activity resulting from continuous cropping compared with periodic pasture leys (Figure 2a), and with increasing length of bare fallow. The differences between crop and pasture ley soils were less pronounced once the pastures had been removed using conventional tillage.

Figure 2a. Effect of crops and pasture leys on soil microbial activity at planting.
Figure 2b. Effect of tillage on soil microbial activity at planting



- Changing from conventional tillage to direct-drill (Figure 2b) had only relatively small effects on microbial activity, with these primarily restricted to the top five cm of the soil profile, a layer that is dry for long periods in most years. These changes were minor compared with the increase arising from stubble retention (data not shown).
- Nematode faunal profiles were used to broadly characterise the biological status of these soils. The soil food web components identified in this analysis did not differ greatly in response to management. There was evidence of increasing food web structure being maintained under minimum/zero tillage, but stresses due to erratic rainfall and carbon inputs combined to minimise these differences. The predominance of fungal-feeding nematodes and the lack of bacterial-feeding enrichment opportunist species suggested slow residue decomposition and nutrient cycling dominated by fungi. Confirmation of this is being sought by other methods.
- PL-FAME analyses also suggested little change in dominance of the various functional groups within the microbial biomass in response to management, with the main impact on the overall level of activity.
- Adding organic amendments to soils increased soil microbial activity, but had only small effects on microbial diversity and crop performance.

Plant response to elimination of biotic constraints

Glasshouse bioassays comparing growth in pasteurised and unamended soil also showed consistent treatment responses, some of which are illustrated in Figure 3.

- Strong positive growth responses were recorded in response to soil pasteurisation in wheat and sorghum, but effects were much smaller in chickpea.
- Pasteurisation responses tended to be less when crops were grown in soil from the 0-5cm layer than from deeper layers of the profile.

- Pasteurisation responses were generally greater in continuously cropped soils compared with soils in pasture leys (data not shown).

Collectively, data suggest there may be widespread biotic constraints in the northern Vertosols that predominantly affect grain crops rather than grain legumes. There is also a fairly consistent trend for these constraints to be minimised in situations with greater levels of microbial activity.

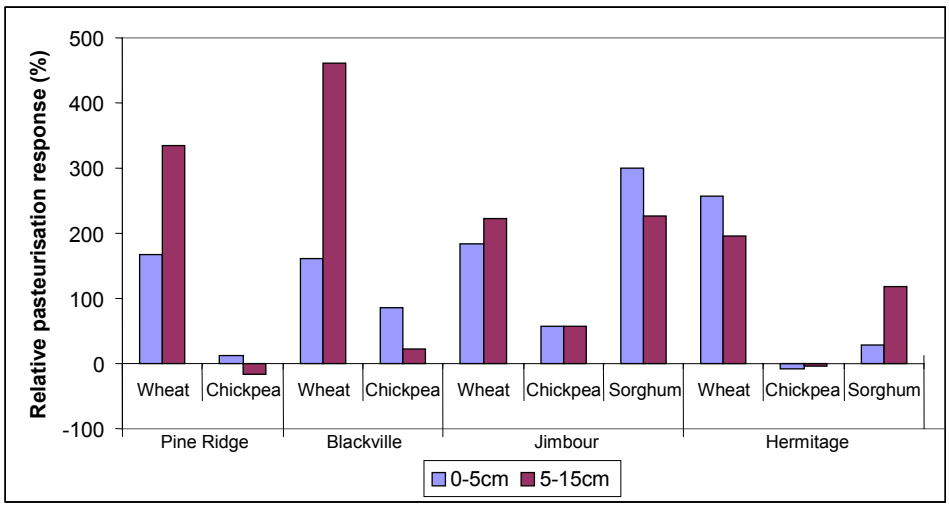


Figure 3. Effects of soil pasteurisation on the relative growth response of sorghum, wheat and chickpea crops in soil from sites at Jimbour. Relative pasteurisation response was calculated as (growth in pasteurised soil/growth in unpasteurised soil) * 100.

Field fumigation studies are underway in an effort to confirm these results, and to determine the importance of these constraints under differing climatic conditions. However, benchmarking of a number of commercial and experimental crops using APSIM has so far suggested that a number of grain crops, especially wheat, have yielded poorly relative to the availability of water and nitrogen for the respective growing seasons (Figure 4). These results are consistent with the suggestion that there may be some significant biotic factors constraining yields of grain crops in this region. The ability of APSIM to predict yield in field-fumigated sites (analogous to potential yield in the absence of biotic constraints) is currently being assessed.

Impacts of management on suppression of lesion nematode

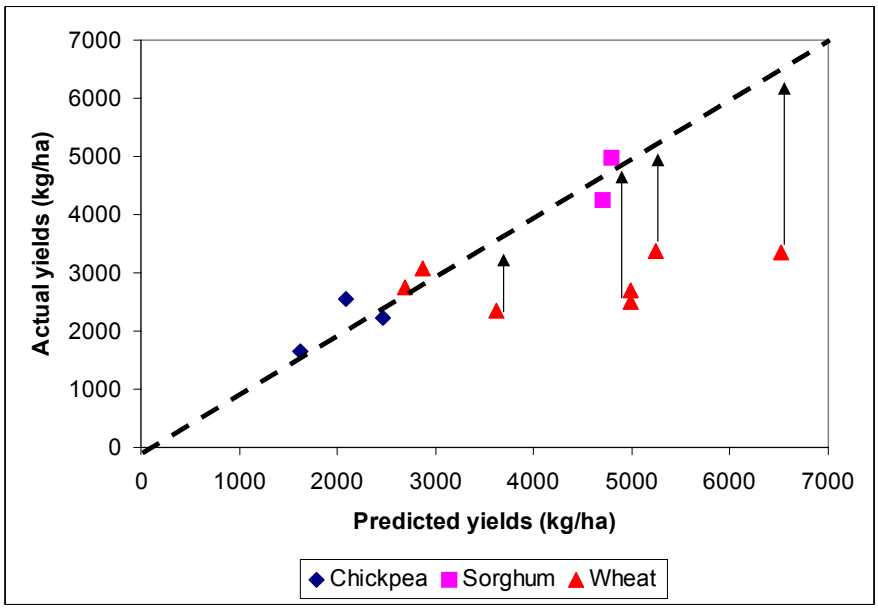
There has been clear evidence of suppression of lesion nematode multiplication in wheat grown in soils under a long-term pasture ley, relative to those with a long-term cropping history (Table 1).

Table 1. Effect of site (crop v pasture) and heat (heat v no heat) on multiplication of *Pratylenchus thornei* on wheat. All pots were inoculated so that the initial inoculum density was about 2.8 *P. thornei*/g soil.

Site	*Final no. of Pratylenchus/g soil*	
	Heated	Non-heated
Crop	23.0	5.4
Pasture	9.5	1.9
LSD (P= 0.05)	3.01	

This suppression seemed to consist of both a biotic component that could be eliminated by heating, and an abiotic component that was still evident after heating. The exact nature of this suppression is being investigated in further glasshouse studies, and a similar technique will be used to investigate potential for suppression of other pathogens such as crown rot in wheat.

Figure 4. Actual commercial yields versus simulated yields predicted using APSIM for a number of commercial sites on the Darling Downs in 2002/03 and 2003 seasons. The arrows represent potential yield lost to unknown factors that could include soil biota.



Management implications, and where to from here

Data collected in the early stages of this project have suggested that detrimental soil biota may be having significant negative impacts on productivity of broadacre grain crops in the Vertosols of the northern region. There are some key components of the cropping system that are impacting on microbial activity and diversity (fallowing, crop rotation, use of pasture leys and tillage), and others are being investigated (eg use of inorganic fertilisers). However, the relationship between these management impacts and the incidence of particular soil-borne diseases is unclear at this time.

There is some concern at the obvious stratification of the majority of microbial activity in shallow surface layers that are dry for long periods of the growing season in this environment, especially under direct-drill systems. Similarly, management strategies that have been devised to conserve soil water (long fallows, skip row cropping) may be having negative impacts on soil biota due to a reduced frequency and non-uniform spatial distribution of organic matter inputs. The low frequency of pasture leys in the more reliable cropping areas is also contributing to the low levels of microbial activity. The implications of some of these management strategies are being investigated in more detail in the continuing experimental program.

References

Dalal RC, Probert ME 1997. Soil nutrient depletion. In Sustainable crop production in the sub-tropics: An Australian perspective. Eds Clarke AL, Wylie PB pp 42-63. Queensland Department of Primary Industries, Toowoomba.

Freebairn DM, Connolly RD, Dimes J, Wylie PB 1997. Crop sequencing. In Sustainable crop production in the sub-tropics: An Australian perspective. Eds Clarke AL, Wylie PB pp 289-305. Queensland Department of Primary Industries, Toowoomba.

Peck DM, Thompson JP, Clewett TG, Haak MI 1993. The root lesion nematode *Pratylenchus thornei* survives extended clean fallows and builds up quickly with wheat cropping. Proceedings of the *Pratylenchus* workshop, 9th biennial conference Australian Plant Pathology Society, Hobart.

Webb AA, Grundy MJ, Powell B, Littleboy M 1997. The Australian sub-tropical cereal belt: soils, climate and agriculture. In Sustainable crop production in the sub-tropics: An Australian perspective. Eds Clarke AL, Wylie PB pp 8-23. Queensland Department of Primary Industries, Toowoomba.

Wildermuth GB, Thompson JP, Robertson LN 1997. Biological change: diseases insects and beneficial organisms. In Sustainable crop production in the sub-tropics: An Australian perspective. Eds Clarke AL, Wylie PB pp 112-130. Queensland Department of Primary Industries, Toowoomba.

Contact details

Dr Mike Bell
Principal Agronomist, Plant Science
QDPI PO Box 23 Kingaroy Qld 4610
Phone 07 4160 0730
Mobile 0427 600 730 or 0429 600 730
Fax 07 4162 3238
Email mike.bell@dpi.qld.gov.au

Soil organic matter, biological activity, and productivity: myths and realities

Graeme Schwenke
NSW Department of Primary Industries, Tamworth

What is soil organic matter?

Any discussion of soil organic matter (SOM) must first define just what is being talked about. This is because different people tend to have slightly different ideas of what SOM is and what it includes. Table 1 shows an example of these differences in the concept of SOM among 23 NSW Department of Primary Industries (NSWDPI) district agronomists in response to a recent survey. For research, extension and discussion of the roles, benefits, and properties of SOM to be coherent, there needs to be a clearer consensus on what is meant by SOM. Baldock and Nelson (2000) derived the following definition from several eminent sources: 'Soil organic matter is the sum of all natural and thermally altered biologically derived organic material found in the soil or on the soil surface irrespective of its source, whether it is living or dead, or stage of decomposition, but excluding the above-ground portion of living plants.' More simply put, soil organic matter is everything in the soil of biological origin, whether living or non-living.

Table 1. SOM components, along with the percentage of 23 NSWDPI district agronomists who would include each component in a description of SOM to a client.

SOM component	% NSWDPI	OC?	SOM component	% NSWDPI	OC?
Humus	100	Y*	Earthworms	52	N
Live plant roots	57	Y/N	Dead animals	70	N
Dead plant roots	100	Y/N	Animal manure	87	Y/N
Viruses	35	Y	Leaf litter	96	Y/N
Bacteria	70	Y	Charcoal	52	Y/N
Fungi (including VAM)	70	Y	Standing stubble	43	N
Protozoa (eg amoebae, flagellates)	52	Y	Party decomposed plant residues	100	Y
Nematodes	61	Y	Sugars	52	Y
Micro arthropods (eg springtails, mites)	26	Y/N	Amino Acids	57	Y
Macro arthropods (eg insects, spiders, centipedes)	22	Y/N	Organic acids (eg citrate, malate)	57	Y

*The Y or N in the OC? column refers to whether the component is measured in an organic carbon soil test. Those marked Y/N are included either partially or occasionally, depending on the thoroughness of the pre-analysis processing, or the analysis method used.

A major reason for the differences in people's understanding of SOM is that soil sampling, sample processing, and laboratory analysis for SOM excludes many of the components listed in Table 1. Nevertheless, SOM is an diverse mixture of components (Figure 1) with proportions in any given soil sample differing enormously depending on climate, parent material, soil texture, vegetation, animals, microorganisms, topography

and land management. Because there is such a range of components encompassed in SOM, components are often grouped on the basis of their typical breakdown rates in soil and their biochemical makeup. The main groups are stable SOM and active SOM (Figure 1).

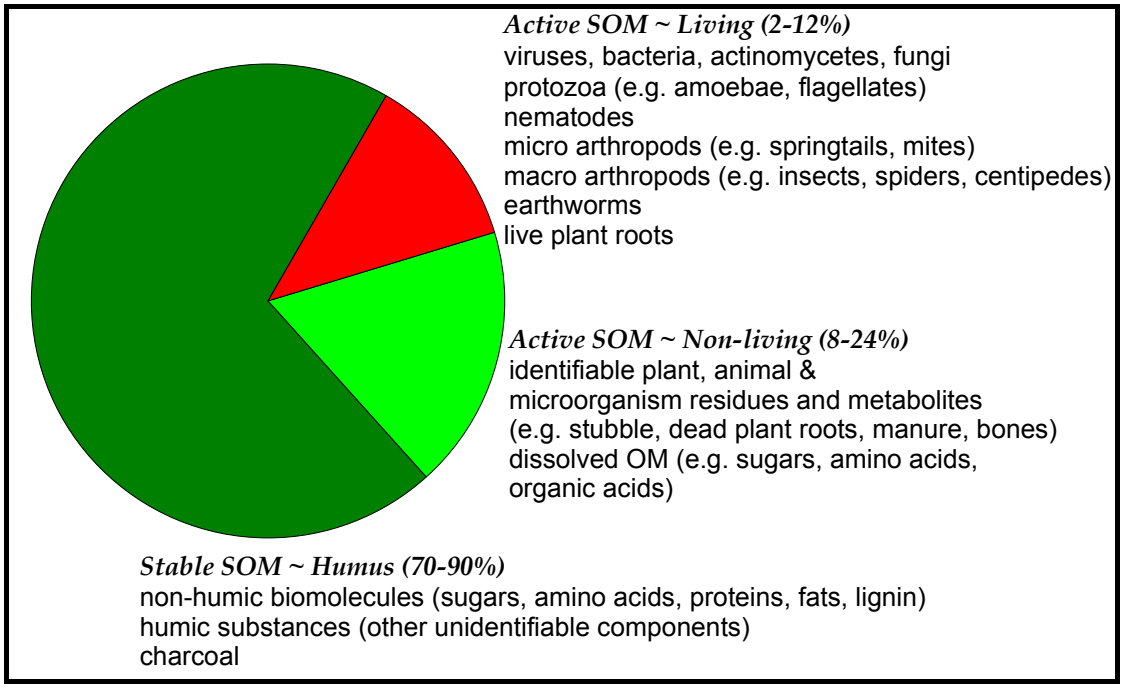


Figure 1. Soil organic matter components and proportions (after Gregorich et al 1997).

Stable components of SOM, known collectively as humus, are either chemically or physically stabilised. Chemically stabilised compounds are highly decomposed compounds of high molecular weight that few microbes can degrade. Physically stabilised compounds are those bound inside soil aggregates where microbes cannot reach. Carbon dating and isotope abundance techniques have shown that the residence time of humus in soils ranges from decades to centuries. Some compounds such as charcoal are practically inert. Chemical compounds within humus are a mixture of identifiable (non-humic substances) and more complex organic molecules (humic substances).

Modern spectroscopic techniques such as nuclear magnetic resonance spectroscopy (NMR), pyrolysis-gas chromatography-mass spectrometry (PyGCMS), and infrared spectroscopy (IR) characterise the chemical structure of SOM compounds non-destructively. These techniques have largely superseded the chemical extractions used until recently to subdivide humus into humic acid, fulvic acid and humin based on grouping humic substances according to their relative solubility in alkali and acid extractants. These terms can be misleading because humic and fulvic acids do not have a specific chemical formula, like sulfuric acid for example, but are instead groupings of many different chemical forms. The modern techniques mentioned above have shown these older subdivisions are flawed because new compounds are created during the extraction procedure, the subdivisions do not relate well to biological functions of organic matter in soils, and the properties of specific extracted compounds are altered compared with the same compounds in their native state in the soil.

Active or labile SOM (Figure 1) is so named because its components break down over periods ranging from days to years. A further subdivision is often used to discriminate between living and non-living components. Living components can be further subdivided using normal taxonomic classifications. Non-living components are typically subdivided based on their physical characteristics. The first subdivision typically occurs at the point of soil sampling, when choosing areas to be sampled (dead animals, animal manures, tree stumps, ash beds, etc. are avoided), when sampling (standing stubble and large leaf litter are physically removed from the surface), and when processing the samples (large leaf litter and plant roots are usually removed). Further subdivision can then be made by first dispersing the soil sample in water or another solution, then either floating off the 'light fraction' or washing the soil through a sieve retaining the 'macro organic fraction'. Organic compounds dissolved in soil water can also be separated from the bulk soil and further characterised.

What are the roles and properties of soil organic matter?

Owing to the tremendous diversity of SOM components, its roles and properties in soil can be biological, chemical, physical or environmental. Table 2 lists these roles, along with their perceived importance to 23 surveyed NSW DPI district agronomists, with responses grouped according to the dominant agricultural industry in their district.

Table 2. Properties of soil organic matter (from Baldock, Nelson 2000), as ranked by NSWDPI district agronomists in order of importance to agricultural industries.

SOM property	Ranked according to dominant industry in district		
	>67% Cropping	Mixed	>67% Grazing
Water-holding capacity (direct – absorbs water)	1	2	3
Soil aggregate 'glue' (stabilises soil structure)	2	1	4
Water-holding capacity (indirect – enhances structure and pore geometry)	3	2	1
Nitrogen storage and supply	3	3	2
Nutrient cycling (from plant and animal residues to plant nutrients)	3	4	8
Cation exchange capacity	4	6	7
Phosphorus storage and supply	4	9	6
Food for microbes (reservoir of metabolic energy)	5	8	8
Sulfur storage and supply	6	10	5
pH-buffering capacity	7	5	10
Ecosystem resilience (eg resists loss of soil fertility induced by disturbance)	8	7	9
Low solubility (organic materials are not leached from soil)	9	11	14
Chelation helps reduce losses of micronutrient	10	14	13
Chelation reduces potential toxicity of metals	11	12	15
Degrades activity and persistence of pesticides	11	13	11
Dark colour (affects soil thermal properties)	12	15	12

Although all district agronomists rated soil structural and nitrogen supply properties highly, responses differed in their rankings according to what the principal agricultural industries of the district were. Interestingly, the group whose clients are mostly graziers felt nutrient cycling from animal and plant residues was less important than cropping-dominant or mixed groups.

The list given in Table 2 is noticeably biased towards the positive aspects or benefits of SOM. Some negative aspects include pathogenic organisms, allelopathic chemicals formed when some residues decay, hydrophobicity (water repellence), and the nitrogen tie-up by plant litter and stubble with high carbon:nitrogen ratio. There are also newly recognised roles and properties of SOM including carbon sequestration (mitigates greenhouse effect), and suppression of weeds (eg Kremer, Li 2003), nematodes (Dunn 1990) and pathogenic microbial organisms. Suppression may be either general suppression, derived from the overall diversity and activity of the soil biota, or specific suppression against single pathogens (Alabouvette et al 2004).

How important is soil organic matter to Australian agriculture?

Nearly all agriculture, apart from soil-less hydroponics, requires some or all of the components of SOM to function. Since SOM is not a nutrient per se but a diverse collection of components, it is nearly impossible to set 'critical levels' below which the system will be affected. Estimates of the nutrient-supplying capacity have sometimes been used to indicate SOM levels required to produce sufficient nitrogen for a crop, but this source on its own is seldom sufficient to produce optimum yields. Other studies have found levels of SOM components below which deterioration in some physical properties has been linked to crop production (eg Bell et al 1998).

Almost all of the 23 district agronomists surveyed felt that SOM was important to very important to the main agricultural industries of their district, with some commenting that SOM was 'critical' or 'vital'. Feelings tended to be strongest in the group whose districts were dominated (>67%) by cropping. It was also this group where the farmers thought SOM important to the main agricultural industries of the district, according to their agronomists. District agronomists whose client base was dominated by grazing, or where cropping and grazing were of similar proportions, found SOM of lesser importance to their industries. Several agronomists commented that while many of their farmers had good knowledge of SOM and felt it was very important, they were still more focussed on other, more immediate, factors such as pH, soil depth, nitrogen or phosphorus fertiliser applications. While many had adopted minimal tillage operations and stubble retention, some industries were still intent on selling off plant residues, even when it had been demonstrated that the fertiliser value of the residues was greater the price received for the residues. The reason was purely economical – they needed a cash flow when produce price dropped below cost.

How can soil organic matter be managed?

SOM is a dynamic, not a static resource. Many of the properties listed in Table 2 occur only as a result of SOM components being constantly broken down by biota to yield simpler compounds for other organisms, 'glues' for aggregate structure, and nutrients for biota and plants. In natural systems, an equilibrium has developed between the supply of raw materials for breakdown and the breakdown rates dictated by the environment. It is important to realise that once an area is converted from a natural system to a managed cropping and grazing system this equilibrium cannot be the same and should not be expected to be. Numerous studies have found that native SOM levels

rapidly decline by up to 60% within a few years of clearing and cultivation (Dalal, Chan 2001). The decline occurs because

- erosion removes SOM-rich topsoil
- cultivation aerates and breaks down aggregates exposing previously protected SOM to microbial activity
- cultivation dilutes SOM-rich topsoil with SOM-poor subsoil
- bare soils have increased periods of wet soil and increased temperature that benefit decomposition
- there are decreased levels and frequency of organic inputs to the soil
- increased quality of organic inputs allows faster breakdown (eg more nitrogen for microbes and less hard-to-digest lignins).

For land managers to manage their SOM requires continued efforts to counter these processes so that SOM decline may be halted and possibly even reversed. To build up SOM to its potential under a managed cropping or grazing system you need to add organic material, monitor SOM levels and reduce SOM losses.

Add organic material

- Grow healthy, productive crops and pastures (better use of fertilisers and crop rotations to increase plant biomass).
- Retain as much residue as possible through stubble, roots or even cover (green manure) crops or ley pastures.
- Apply animal manures, biosolids, etc.
- Locate and use off-farm sources of organic matter, such as food-processing wastes and composted products.

Reduce losses

- Reduce tillage operations by following minimum or no-tillage management systems.
- Retain crop residues by not burning or baling for fodder.
- Reduce periods of bare soil or fallowing by opportunity or response cropping.
- Control erosion.
- Grow plants more resistant to microbial breakdown.

Monitor SOM levels

- Keep in mind the likely effects of various operations on SOM and check periodically by soil testing.

The simplest check on SOM levels is a soil test for organic carbon (OC). While SOM can be measured directly, the method (called weight loss on ignition) can give errors in some soils (where some minerals also lose weight). OC is more often used as the method is simple and carbon is a relatively constant proportion of SOM, although OC to SOM conversion factors quoted in the literature vary from 1.724 (Van Bemmelen factor) to 2.0. Further conversions are sometimes used if the OC method used is ‘Walkley-Black’ as this method may not measure all carbon present. In both instances the conversion factors, if used at all, should really be specifically measured for that particular soil type and soil depth. The tediousness of doing this means that people usually report OC without conversion to SOM.

More component- or fraction-specific methods for monitoring SOM are available, but these tests are often not commercially available. One reason is that there are usually no universal interpretation criteria for the tests, and many are highly specific to recent, often transient conditions occurring in the soil. Doran (2000) found that while measurements of soil organisms were sensitive to management and correlated with beneficial soil functions, meaningful tests of soil organisms for use by land managers still required development.

What are some practicalities of SOM amendments?

Soils depleted of SOM can be 'restocked' by adding more and losing less. While options for losing less are becoming more commonplace, particularly reduced tillage, large amounts of external organic amendments are required to make any short-term impact on soil SOM levels. For example, a hectare of soil to a depth of 10 cm weighs approximately 1000 tonnes. If OC is 1% (10 t/ha), then SOM is 1.7% (17 t/ha). To increase SOM to 3.5% (35 t/ha) (OC to 2.0%) would require an extra 18 t/ha of OM (10 t/ha OC). However, simply adding 18 t/ha of, say, animal manure would not give you the 1.8% increase in SOM because 80-90% of the original material is lost in the decomposition process over several years. Carbon is lost as CO_2 to the atmosphere. An alternative is to add material that is already largely decomposed, such as well-matured composts, considered to be equivalent to the more stable fractions of SOM. A review by Gibson et al (2002) found many cases of improvements in SOM, various soil properties and plant productivity as a result of adding such composted material, termed recycled organics. Where the balance of OM inputs exceeds losses over time, SOM should increase. While large additions of recycled organics or animal manures should increase SOM rapidly, improvements in cropping and pasture systems may take five years or more to register an increase in an OC soil test. Increases occur firstly in the smaller active SOM fraction with benefits to soil structure and microbial diversity, then later in the stable SOM.

Building SOM levels is unlikely to be the major aim of broadacre farm management, but should be an important consideration when planning management decisions that affect inputs and losses. For example, the use of organic amendments such as animal manures or recycled organics should be considered not just in a SOM-building sense but primarily as a means of supplying plant nutrients. Other important considerations of manures include cost, carbon:nitrogen ratio, weeds, diseases, contaminants, allelopathic chemicals, and application issues. Applying manures in excess of current plant requirements increases potential for environmental damage from runoff or leaching. Long-term trials (20-120 years) comparing manuring and inorganic fertiliser application (Edmeades 2003) have shown that manured soils had higher contents of SOM and numbers of microfauna than fertilised soils, and were more enriched in several plant nutrients. Manured soils also had lower bulk density and higher porosity, hydraulic conductivity and aggregate stability, relative to fertilised soils. However, on average, there was no difference in long-term effects on crop production between fertilised and manured plots. Why? High input agriculture often 'masks' the relationship between yield or crop health and soil quality parameters such as SOM (Gregorich et al 1997). But fertilisers and pesticides are significant farming costs. The benefits of extra SOM for nutrient supply or pest suppression should be valued as cost-saving or risk-reducing. A case study by Ringrose-Voase et al (1997) was able to link OC levels in several land units of the Wagga Wagga region to land values based on the relationships between OC and economical and productive potential.

What about applying microbial products to the soil?

Our hypothetical soil of 1000 t/ha weight would conceivably contain about 1 t/ha of microbes. Commercially available microbial products claim not to increase this soil fraction directly, but rather to inoculate the SOM with specific organisms or groups of organisms that will rapidly build up in the soil to a point where they will influence soil properties. Favourable environmental conditions of moisture, aeration, temperature, pH, and energy source are required for inoculants to build up in soil; conditions under which native soil biota are also likely to be abundant and strongly competitive against the added organisms (Sullivan 2001). Until more widespread scientific testing of such products in the field is conducted, farmers should be equipped with robust methods of making their own comparisons between current and new technologies, rather than the ad hoc testing that predominates.

What about applying humic acids to the soil?

Humic acid or humate products are generally extracts from leonardite or lignite, a mineral similar to brown coal, although the extracted compounds differ from those extracted as humic acid from SOM (Ayuso et al 1997). As with microbial products, increasing SOM measurably with humic acid products is unlikely given the scale of addition advocated versus the background levels in SOM. However, there are many claims and some reports in the scientific literature that adding humic acid products to soils may stimulate plant growth and increase yield, possibly due to mechanisms such as delaying precipitation of phosphorus from mineral fertilisers in certain soil types (Delgado et al 2002). Whether such applications will work and, if so, are economical, will be affected by your particular farming system with its unique combination of soil type, climate, landscape, paddock history and economic situation. Determining this requires unbiased, scientifically rigorous, but not necessarily complex, testing and evaluation.

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References

Alabouvette C, Backhouse D, Steinberg C, Donovan NJ, Edel-Hermann V, Burgess LW 2004. Microbial diversity in soil: Effects on crop health. In Managing soil quality: Challenges in modern agriculture. Eds Schjønning P, Elmholt S and Christensen BT pp 121-138. CABI Publishing, Wallingford UK.

Ayuso M, Moreno JL, Hernandez T, Garcia C 1997. Characterisation and evaluation of humic acids extracted from urban waste as liquid fertilisers. Journal of the Science of Food and Agriculture 75 481-488.

Baldock JA, Nelson PN 2000. Soil organic matter. In Handbook of soil science Ed Sumner ME pp B25-B84. CRC Press, Boca Raton.

Bell MJ, Moody PW, Connolly RD, Bridge BJ 1998. The role of active fractions of soil organic matter in physical and chemical fertility of Ferrosols. Australian Journal of Soil Research 36 809-819.

Dalal RC, Chan KY 2001. Soil organic matter in rainfed cropping systems of the Australian cereal belt. Australian Journal of Soil Research 39 435–464.

Delgado A, Madrid A, Kassem S, Andreu L, del Carmen del Campillo M 2002. Phosphorus fertiliser recovery from calcareous soils amended with humic and fulvic acids. Plant and Soil 245 277-286.

Doran JW, Zeiss MR 2000. Soil health and sustainability: managing the biotic component of soil quality. Applied Soil Ecology 15 3–11.

Dunn RA 1990. Soil organic matter, green manures and cover crops for nematode management. Nematology Plant Protection Pointer 38 2.

Edmeades DC 2003. The long-term effects of manures and fertilisers on soil productivity and quality: A review. Nutrient Cycling in Agroecosystems. 66 165-180.

Gibson TS, Chan KY, Sharma G, Shearman R 2002. Soil carbon sequestration utilising recycled organics: A review of the scientific literature. Project 00/01R-3.2.6A prepared for Resource NSW by The Organic Waste Recycling Unit, NSW Agriculture August 2002. http://www.resource.nsw.gov.au/data/Soil_Carbon_Sequestration.pdf.

Gregorich EG, Carter MR, Doran JW, Pankhurst CE, Dwyer LM 1997. Biological attributes of soil quality. In Soil quality for crop production and ecosystem health Eds Gregorich EG and Carter MR pp 81-113. Elsevier, The Netherlands.

Kremer RJ, Li J 2003. Developing weed-suppressive soils through improved soil quality management. Soil & Tillage Research 72 193–202.

Rinrose-Voase AJ, Geeves GW, Merry RH, Wood JT 1997. Soil indicators of changing land quality and capital value. CSIRO Land and Water technical report 17/97. http://www.clw.csiro.au/publications/technical/tr17-97.pdf.

Sullivan P 2001. Alternative soil amendments. National Sustainable Agriculture Information Service: Horticulture technical note. http://attra.ncat.org/attra-pub/altsoilamend.html.

Contact details

Dr Graeme Schwenke
Soil Scientist
Tamworth Agricultural Institute
NSWDPI RMB 944 Calala Lane Tamworth NSW 2340
Phone 02 6763 1137
Fax 02 6763 1222
Email graeme.schwenke@dpi.nsw.gov.au

Impact of management practices on soil microbial functions in alkaline Mallee soils.

David Roget and VVSR Gupta
CSIRO Land and Water, Glen Osmond SA

Introduction

The Mallee region covers an area of some 5 million ha of SA, Vic and NSW. It is characterised by dune/swale landforms that contain soils that are generally coarse in texture (5-15% clay). The soils are inherently of low fertility and low in organic matter (total carbon = 0.5%). Rainfall is low (250-350mm/year), variable and winter-dominant.

The marginal production characteristics of the Mallee have led to the development of low risk farming systems based on cereal/pasture or cereal/fallow rotations with low fertiliser inputs which have consisted largely of additions of phosphorus. This has resulted in low productivity both in terms of crops and pastures with cereal yields achieving around 50% of the water limited potential (average wheat yield 1.2 t/ha). Productivity can be limited by numerous factors including cereal root diseases and chemical subsoil constraints but overall it is limited in dry years by lack of rainfall and in better rainfall years by lack of fertility. The low productivity combined with other factors including heavy grazing, fallowing and endemic wind erosion have resulted in low returns of organic matter to the soils with subsequent limitations to microbial activity and microbial functions.

A project was initiated in 1997 (Mallee Sustainable Farming Project) to investigate the potential to significantly improve Mallee farming systems. The project was based on the hypothesis that productivity gains of up to 100% could be made by more efficient utilisation of the available rainfall with more intensive cropping and improved tillage and fertiliser strategies. This increase in cropping intensity, with increased productivity in association with limited grazing and zero tillage, would result in a substantial increase in the return of organic matter to the soil with an associated increase in microbial activity and function, which are essential for the long-term sustainability of any farming system. In these coarse-textured Mallee soils with low levels of organic carbon and limited opportunity for protection of organic matter it was hypothesised that an increased flow of microbially available carbon would quickly provide a significant improvement in microbial biomass, microbial activity and populations of microbial functional groups involved in nutrient cycling and other microbial functions.

This paper reports on results on the impact of agronomic management on soil microbial function from both the Mallee Sustainable Farming Project and an earlier experimental program at Avon SA.

Methods and Materials

Avon field site

A long-term field trial has been maintained at Avon, South Australia (34° 14’ S, 138° 18’ E) since 1979. The climate is Mediterranean, characterised by hot dry summers and a winter-dominant, average annual rainfall of 350 mm. The soil is an alkaline calcareous sandy loam, classification Gc1.12 (Northcote et al 1975), or solonised brown soil (Stace et al 1968), or calcic xerosol (Dudal 1968). Soil chemical properties at the start of the

trial were pH(water) 8.2, organic carbon 1.6%, total nitrogen 0.14%, Colwell bicarbonate-extractable phosphorus 65mg/kg, and calcium carbonate 8%. Particle size distribution (%) is clay 12, silt 3, fine sand 34 and coarse sand 42.

Waikerie field site

A field trial was established at Waikerie, South Australia (34° 17' S, 140° 02' E) in 1998. The climate is Mediterranean, characterised by hot dry summers and a winter-dominant average annual rainfall of 260 mm. The soil is an alkaline calcareous loamy sand, classification Um5 (Northcote et al 1975), or gray-brown or red calcareous soil (Stace et al 1968), or alfisol (Dudal 1968). Soil chemical properties at the start of the trial were pH(water) 8.6, organic carbon 0.68%, total nitrogen 0.05%, Colwell bicarbonate-extractable phosphorus 12 mg/kg, and calcium carbonate 0.4%. Particle size distribution (%) is clay 6, silt 1, fine sand 43 and coarse sand 47.

Analytical details

Microbial biomass carbon, nitrogen and phosphorus levels were determined using chloroform-fumigation extraction methods (Joergensen 1995). *In situ* microbial activity measurements were performed using a portable infrared gas analyser (EGM-1 Environmental gas monitor, PP Systems, Hertfordshire, UK). Carbon and nitrogen mineralisation potentials were estimated using a 21-day laboratory incubation method (Gupta et al 1994) and the off-season nitrogen mineralisation values were calculated from the deep nitrogen measurements (mineral nitrogen measurements in one metre profile) at the harvest of a crop and seeding of next season's crop. Mineral nitrogen levels were measured on 2M KCl soil extracts (1:3 soil to extractant ratio) using the method described by Rayment and Higginson (1992). Soil enzyme activities were determined using the p-Nitrophenol and other colourimetric based methods originally described by Alef and Nannipieri (1995) with modifications to suit our soils.

Results and discussion

Management impacts on non-symbiotic N_2-fixation.

At an experimental site at Avon SA a nitrogen budget was calculated for 17 years of continuous wheat. The wheat crops were direct-drilled with no added nitrogen fertiliser and all stubble was retained on site. After accounting for nitrogen removed in the grain and the change in total soil nitrogen there was 334 kg of extra nitrogen/ha that was unaccounted for (Table 1). One possible explanation for the extra nitrogen is through an input from non-symbiotic N_2-fixation (NSNF). An evaluation of potential NSNF for the Avon region has been made by Gupta et al (2002) who calculated an average nitrogen input of 10-15 kg/ha for the period from January to June based on rainfall/temperature relationships and assuming the requirement for available carbon would be adequately met from stubble remaining after harvest. Given that there is also the opportunity for some NSNF during spring, the modelled NSNF figures corresponded closely to the unaccounted nitrogen in the nitrogen budgets. The amount of unaccounted nitrogen is agronomically very significant in that it is equivalent to approximately 30-50 % of the nitrogen exported in wheat crops given expected district yields of 2-3 t/ha. It is probable that the carbon inputs from the management practices of intensive cereal cropping and stubble retention have directly contributed to an increase in NSNF at this site.

Table 1. Nitrogen budget for 17 years of continuous wheat (1979-1996) with no added nitrogen fertiliser, Avon SA.

Nitrogen removal in grain			Change in soil nitrogen			Unaccounted nitrogen	
Total wheat yield (t/ha)	Average N content (%)	Total N exported (kg)	Total soil N 1979 (%)	Total soil N 1996 (%)	Change in soil N (kg)	Total N (kg/ha)	N/ year (kg/ha)
19.7	2.0	394	0.14	0.135	-60	334	19.7

Management impacts on nitrogen and phosphorus dynamics

Following four years of treatments, the 'improved systems' with more intensive cropping and higher fertiliser inputs at Waikerie produced significant improvements in soil microbial measurements compared with the district practice of low fertiliser, wheat-pasture treatment (Table 2).

Table 2. Microbial biomass carbon and nutrient levels in the surface soils (0-10 cm) of selected treatments at Waikerie core site (after four years).

Cropping system	MB-C (kg C/ha)	MB-N (kg N/ha)	MB-P (kg P/ha)	Microbial activity§ (g CO2 /m2/hour)
Pasture-Wheat (DP)	265a#	26 a	16.0a	0.105
Pasture-Wheat (HI)	370b	43 cd	21.0b	0.185
Legume-Wheat (HI)	370b	46 d	13.0a	0.210
Canola-Wheat (HI)	357b	36 bc	16.5a	0.175

#Values in each row followed by the same letter are not significant at P<0.05.
§ Average values from six *in situ* respiration measurements made in each experimental plot.

Microbial activity increased on average for all 'improved systems' by 81% over the district practice treatment and reflected the increase in plant biomass production with these treatments. Microbial biomass carbon increased 40% with higher fertiliser inputs and this increase was not influenced by rotation. Microbial biomass nitrogen was significantly influenced by rotation with an increase of 38% following continuous crop with no legume component and by 73% where there was a legume component in the rotation when compared with district practice.

The increase in microbial carbon following the 'improved systems' indicates that the carbon from the extra stubble and root residues is not just being respired as CO_2 but a proportion is being maintained within the microbial biomass. A significant increase was also seen in the populations of cellulolytic bacteria and fungi and nitrifying microorganisms, functional groups of microorganisms involved in carbon and nitrogen cycling. A key functional outcome of this is an improved net mineralisation of nitrogen between harvest and sowing (Table 3). Following a summer with a number of rainfall events (2000-01), up to an extra 30 kg/ha of nitrogen was mineralised under the 'improved systems' compared with the district practice. The higher rate of mineralisation under the 'improved systems' is also likely to provide benefits within crop, particularly during spring when extra nitrogen can be mineralised in response to rainfall events. This has the potential to provide better matching of nitrogen availability to plant requirement in the low and variable rainfall environment of the Mallee and to

allow the crop to optimise benefits from spring rains with less of the risks involved with higher fertiliser nitrogen levels.

Table 3. Amount of nitrogen mineralised (kg N/ha) during the off-season (summer and spring) as influenced by cropping system type at Waikerie SA.

Treatment 2000	Off-season mineralisation (Nov 2000-May 2001)	Treatment 2001	Off-season mineralisation (Nov 2001-June 2002)
Wheat-DP# (1)	10.3	Pasture-DP	18.5
Wheat-HI§(3)	14.0	Pasture-HI	37.0
Wheat-HI (8)	36.1	Peas-HI	35.0
Wheat-HI (9)	24.7	Canola-HI	23.1
Wheat-HI (10)	34.0	Wheat-HI	23.0
Canola-HI (11)	41.5	--	

DP: district practice fertiliser rate comprising 10 kg P/ ha; 5 kg N/ha.
§ HI: high fertiliser rate comprising 15 kg P/ha; 27 kg N/ha (N excluded for pulse crops).

The greater reserves of nitrogen in the microbial biomass also provide a buffer against leaching losses that have been measured at up to 50kg N/ha in the Mallee following large summer rainfall events. Improved microbial biomass levels can contribute significantly to the long-term nutrient efficiency of these systems as indicated by an assessment of mineral nitrogen from a depth of 2-6 metres which showed up to 600 kg/ha of accumulated nitrogen following typical district practice management.

An assessment of the activity of phosphatase enzymes has shown an increase of up to 29% with the high microbial activity of the 'improved systems'. The dynamics of phosphorus in the soil make it difficult to quantify, directly, any improved phosphorus availability, but the higher enzyme activity coupled with increased available phosphorus levels provide strong evidence for this.

Management impacts on suppression of cereal root diseases

The level of disease-suppressive activity in soils against fungal diseases is a function of the population, activity and composition of the microbial community. All soils have an inherent level of suppressive activity, but this level can be significantly modified by management practices used within a farming system. At Avon SA, disease suppression increased from a low to high level over a period of 5-10 years following a change in management practices to full stubble retention, limited grazing and higher nutrient inputs (Roget 1995). The increase in suppression provided complete control of the soil-borne diseases rhizoctonia (Roget 1995) and take-all (Roget 1997). Soils with high levels of disease suppression have also been identified in commercial farms across SA and Victoria (Roget et al 1999). The management factors consistently related to soils with improved disease suppression included intensive cropping, stubble retention, limited grazing, limited or no cultivation and above average yields (high water use efficiency). These management practices increase carbon inputs that result in changes to the composition and activity of the soil microbial community over time. These changes result in greater competition for soil resources that, along with predation and inhibition of pathogens, lead to increased suppression.

In the short-term (one to three years), the effectiveness of the disease-suppressive activity already developed in the soil can be influenced by the availability of mineral nitrogen, particularly during the summer and early autumn period. As the amount of available nitrogen (ie nitrate nitrogen) in the topsoil increases during this non-crop period, the disease suppression occurring in the following crop season decreases. The impact of higher soil nitrogen levels and its relationship to added carbon is shown in Figure 1. The underlying suppressive activity may not be lost, but may not be expressed effectively in the presence of high mineral nitrogen levels.

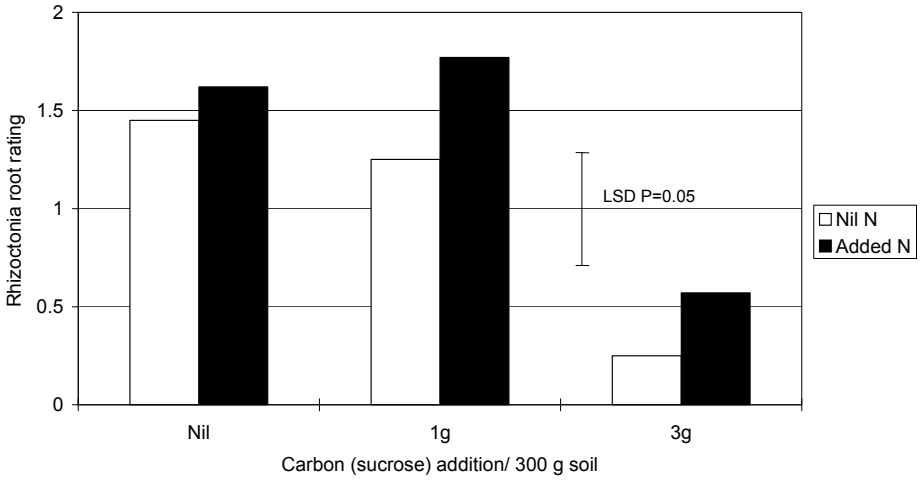


Figure1. Impact of added carbon and nitrogen on the level of rhizoctonia root damage (suppression) on soil with a natural level of inoculum of *Rhizoctonia solani*. Nitrogen added as urea at 15% of carbon addition.

Any factors that result in the accumulation of higher available nitrogen levels for longer periods of time will tend to curb the effectiveness of the suppressive activity of the soil. From the point of view of strong suppressive activity, a good farming system includes a productive intensive cropping system to provide export of nitrogen and a strong nitrogen sink through a supply of biologically available carbon (production and retention of residues) to maintain higher levels of microbial carbon turnover. This does not necessarily equate to a low fertility system but one in which the timing of nitrogen availability is more synchronised with the crop requirements. In such systems, early season nitrogen availability may depend heavily on fertiliser nitrogen but later crop requirements would be supplied by net nitrogen mineralisation through microbial turnover.

Conclusions

Soil microbial functions, including non-symbiotic N-fixation, nitrogen and phosphorus availability and disease suppression can be significantly improved through changes in onfarm management in the Mallee. Management changes that increased productivity and resulted in the return to the soil of higher levels of microbially available carbon were the driver for greater soil microbial activity and improved microbial functions. The coarse-textured Mallee soils are likely to be particularly responsive to increased carbon inputs due to the low soil carbon levels and rapid turnover of added carbon as a result of the limited protection offered by these soils. The higher carbon inputs from the 'improved systems' will most likely determine the upper limit of the improvements in microbial function. At Waikerie these improvements have been monitored for five

years, and it is not clear at this stage if the maximum level of microbial function has already been obtained or if with time there is still further improvement to come.

References

Alef K, Nannipieri P 1995. Methods in applied soil microbiology and biochemistry. Academic Press, New York.

Dudal R 1968. Definitions of soil units for the soil map of the world. World Soil Resources Report No 33. Food and Agriculture Organisation, Rome.

Gupta VVSR, Roper MM, Kirkegaard JA, Angus JF 1994. Changes in microbial biomass and organic matter levels during the first year of modified tillage and stubble management practices on a red earth. Australian Journal of Soil Research 32 1339-1354

Gupta VVSR, Roper MM, Roget DK 2002. Potential for non-symbiotic N_2-fixation in different agroecological zones of Southern Australia. Symposium papers of the 17th World Congress of Soil Science, Bangkok, Thailand. Symposium 12 Paper 1553 pp 1-10.

Joergensen R 1995. The fumigation extraction method. In Methods in applied soil microbiology and biochemistry. Eds Alef K, Nannipieri pp 375-418P. Academic Press, New York.

Northcote KH, Hubble GD, Isbell RF, Thompson CH, Bettenay E 1975. A description of Australian soils. CSIRO, Melbourne.

Rayment GE, Higginson FR 1992. Nitrogen method 7C2. In Australian laboratory handbook of soil and water chemical methods pp 47-57. Inkata Press, Melbourne.

Roget DK, Coppi JA, Herdina, Gupta VVSR 1999. Assessment of suppression to *Rhizoctonia solani* in a range of soils across SE Australia. In Proceedings 1st Australasian soil-borne disease symposium Gold Coast Qld.

Roget DK 1997. Development of take-all suppressive soils in South Australia. In Proceedings 11th APPS conference Perth WA.

Roget DK 1995. Decline of root rot (*Rhizoctonia solani* AG-8) in wheat in a tillage and rotation experiment at Avon, South Australia. Australian Journal of Experimental Agriculture 35 1009-13.

Stace HCT, Hubble GD, Brewer R, Northcote KH, Sleeman JR, Mulcahy MJ, Hallsworth EG 1968. A handbook of Australian soils pp 435. Rellim, Glenside SA.

Contact details

Mr David Roget
Principal Research Scientist
CSIRO Land and Water
PMB 2 Glen Osmond SA 5064
Phone 08 8303 8528
Fax 08 8303 8560
Email david.roget@csiro.au

Soil structure and soil biota: their interactions and implications on soil health

Yin Chan
NSW Department of Primary Industries, Richmond

Introduction

Soil is a living system and has to be managed as such in agriculture to improve sustainability. The importance of harnessing the beneficial functions of soil biota in agriculture has been recognised (eg Lee, Pankhurst 1992), but to do this we need better knowledge of the factors controlling the distribution, abundance and activity of soil biota. These factors are largely controlled by the spatial heterogeneity of the soil environment which is determined by soil structure. Hitherto, this important aspect of soil attributes has been largely ignored (Young, Ritz 2000). Such knowledge is a pre-requisite if processes mediated by soil biota are to be harnessed for agricultural production. Soil has been largely studied as a homogeneous material, where soil chemists routinely grind up the soil prior to performing analyses and microbiologists carry out their study largely on laboratory culture media.

This paper reviews the interrelationships between soil structure and soil biota which affect soil functions. It outlines the importance of soil structure on abundance, diversity and activity of soil biota, looks at the effects of soil biota in modifying soil structure, and discusses the importance of soil biota interactions on soil health and the role of soil management practices in harnessing the beneficial functions of soil biota.

Soil structure impacts on soil biota

Soil as a habitat for living organisms in agro-ecosystems

Soil provides the habitats for a whole range of living organisms (Lee, Foster 1991). Soil biota (flora and fauna) have been traditionally classified in terms of their physical size, namely micro-, meso- and macro- (Lee, Foster 1991). The architecture and the physical environment of the ecological niches are controlled by soil structure. Structure can therefore influence the type of organisms (diversity), the population density that can exist in a particular soil (abundance), and the activity of organisms. All these factors affect the biological fertility of the soil.

Soil structure is often defined as the size, shape and arrangement of aggregates and the spaces or pores in between at a given time. Therefore soil structure can be described both in terms of the pore system as well as the arrangement of primary soil particles into hierarchical structural states (Kay 1990). However, from a functional point of view in terms of soil as a habitat and the activities of the inhabitants, it is more meaningful to focus on the pore space system which can be described in terms of total porosity, pore size distribution and continuity of the pore systems.

The concept of 'habitable pore space' suggests that there is a relationship between the size of organisms and the zones of soils they are physically able to inhabit (Young, Ritz 2000). As Figure 1 shows, soil contains pore spaces and particles (aggregates) that vary in size over seven orders of magnitude ($<10^{-7}$ m to 1 m), so it provides a heterogeneous environment. Pore size distribution differs according to soil type, thus offering habitats for a diverse range of organisms. Clay soils provide better habitats for bacteria because of higher micro-porosity, so there are more micro-niches than sandy soils (Foster 1994).

Research has shown a clear correlation between soil pore volume classes and nematode and bacterial biomass in some cases (eg Hassink et al 1993).

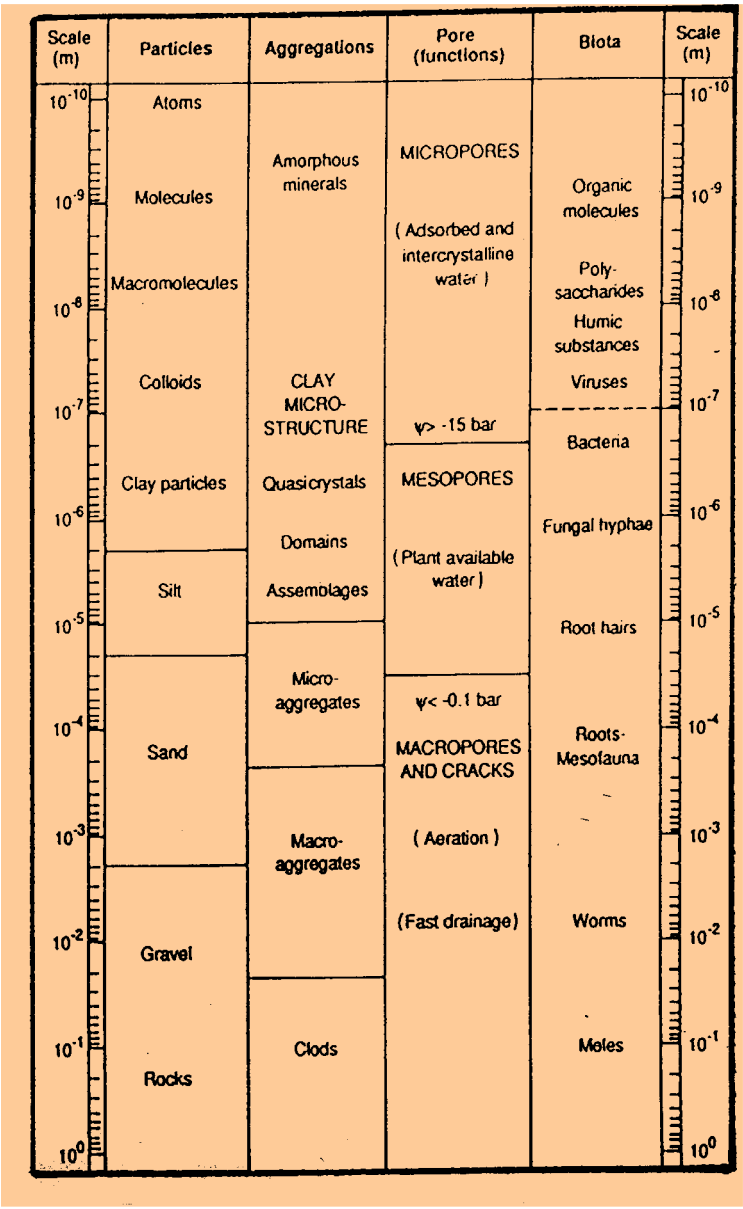


Figure 1. Soil biota in relation to different components of soil structure with different sizes and scales (Kay 1990).

Soil structure

The interaction of soil structure with soil water content determines a number of important physical properties and processes which in turn define the physical environment of the habitat (Smiles 1988).

Soil water availability and permeability

Soil water is held at different energy levels, depending on the pore size, so that water in smaller pores is held more strongly than that in larger pores. The relationship between soil water content and the energy with which the water is held by the soil (soil water potential) is referred to as 'soil moisture characteristics' and differs according to soil type. It defines the availability of water or, in other words, the moisture stress a

particular organism is subjected to at a particular soil water content. For a salt-free soil, the water potential represents the energy that an organism must expend to retain (extract) water against the attraction of the soil. It also controls the relative humidity of the soil atmosphere. The lower the water potential and the drier the soil, the greater the stress experienced by the organism. The moisture characteristics of sandy and clay soils are very different, so at the same water content, their water potentials which determine water availability will vary considerably. Ecologically, it is more meaningful to express soil water status in terms of potential rather than actual soil water content. The pore size distribution of a soil can be calculated from its moisture characteristics (Marshall, Holmes 1979). The relationship between permeability and soil water content is also determined by pore size distribution of a particular soil. As water can move much faster through larger pores than smaller pores, sandy soils have higher permeability than clay soil and the rate of water movement decreases rapidly as a soil becomes drier.

Soil aeration

The composition of soil atmosphere, and hence its aeration status, is governed by gaseous diffusion processes between the above ground atmosphere and the soil, which depend on the air-filled porosity of the soil (Smiles 1988). Air-filled porosity varies with the soil water content and the pore size distribution of the soil. Gaseous diffusion through the water phase is about four orders of magnitude less than through air and is therefore negligible. This means that at the field scale, soil is a heterogeneous environment with aerobic and anaerobic zones dispersed through out the soil volume.

Soil strength

While many of soil organisms live in and depend on pre-existing soil pores, some larger organisms can make new pores. Plant roots, earthworms and termites fall into this category. Their abilities to deform soil depend on the mechanical strength of the soil. Mechanical strength is a function of both soil structure and soil water content, and increases with bulk density (mass of dry soil per unit volume) and with decreasing soil water content.

Thermal properties

Varying soil temperature regimes within the soil environment, both diurnal and annual, directly affect biological activities. However, soil temperatures are modified by thermal conductivity and thermal capacity of soil, both of which are functions of soil structure (bulk density) and soil water content (Smiles 1988).

Soil structure and biological processes

Soil is a heterogeneous environment, spatially and temporally. Many biological activities and processes in soils are modulated by soil structure, particularly via the interactions between soil structure and water.

Activity and mobility

Different soil organisms have different activity and survival ranges in terms of soil water potential. For instance, bacteria are most active in the soil moisture potential range of -0.01 and -0.03 MPa (Lavelle, Spain 2001). In the case of earthworms, activity in surface soil ceases when drier than -150 kPa (Baker et al 1993). Soil moisture characteristics control the mobility of many small soil animals such as nematodes, motile bacteria and aquatic phycomycetes which are restricted to existing water-filled soil pores. Existence and activities of these organisms depend on availability of sequences of water-filled pores of the right sizes to permit their passage (Papendick, Campbell 1985). This is pre-determined by soil structure (pore size distribution) at a particular soil water potential. Filamentous fungi are less restricted by such factors. Permeability affects the rate of soil water movement and solute supply, and controls

many biological activities such as wilting and germination of plants, and hatching of nematode cysts and spores (Smiles 1988).

Nitrogen mineralisation and cycling

Soil is a complex interconnected framework of pores and solids. At field scale and under particular soil water status, aerobic and anaerobic zones often exist in close proximity. This controls the distribution of the types of organisms and has direct impact on processes of nitrification, denitrification and decomposition processes and therefore nitrogen/carbon cycling. Spatial heterogeneity of soil structure, distribution of plant roots and other organic debris determine how soil functions and the degree of heterogeneity of microbial and nitrogen transformation throughout the profile (Young, Ritz 2000).

Prey and predation relationship

Organisms residing in pores of appropriate size are protected from predation by organisms of larger dimension since the latter are denied physical access to their prey. This has been demonstrated in the case of prey-predator relationship for *Pseudomonas flurorescens* and ciliate protozoan *Colpoda steinii* (Wright et al 1993). When the bacteria are predominantly located in the smaller pores (<6 um), they better survive protozoa predation. Such interaction is influenced by pore size distribution and has impact on nutrient cycling, mineralisation processes and disease prevalence via its effect on composition of the food web (Lee, Pankhurst 1992).

Substrate availability and carbon sequestration

Substrates existing in pores of appropriate size are protected from breakdown by soil microorganisms because of their physical inaccessibility. As a result, even labile forms of organic carbon can be sequestered in soil. The effect of tillage and other forms of soil disturbance is to expose these substrates to the microbial population and results in the commonly observed accelerated soil organic carbon decline under conventional cultivation practices.

Soil biota impacts on soil structure

It is well documented that soil biota can directly and indirectly influence soil structure. Directly, bacteria stabilise soil aggregates by their polysaccharides gel and, in the case of fungi, by the physical entanglement of their filamentous hyphae. Direct correlation between macroaggregate stability and hyphal length has been established (Tisdall, Oades 1980). Bacteria in colonies are often found within microaggregates and in association with dispersed clay particles (Foster 1994), and their carbohydrate gel surrounding them is important in microaggregate formation (Emerson et al 1986). Mesofauna such as protozoa and nematodes do not have direct effects on soil structure but can indirectly affect aggregate structure through their regulation of bacterial and fungal populations.

While the microbes largely survive in the pre-existing soil structure, larger soil biota are known to modify soil structure in creating aggregates and burrows. Earthworms, termites and ants have been called the soil ‘ecosystem engineers’ because of their ability to modify soil structure. Casting and burrowing activities of earthworms have been well documented since the seminal work of Darwin (1811) for *Lumbricus terrestris* (Lee, Foster 1991). Earthworms are by far the most effective in transporting of soil material within the soil profile (Table 2). However, given the large range of values, the ability of earthworms to move material varies with species, abundance, abiotic factors and management.

Table 2 Soil turnover rates by different soil biota as recorded in the literature (modified from van Vliet, Hendrick 2003)

Soil biota	Soil turnover rate (t/ha/yr)
Enchytraeids	0.75-21.8
Earthworms	40-1000
Termites	0.75-45
Ants	0.42-10

Implications for soil health

Soil structure is defined as 'the spatial heterogeneity of the different components or properties of soil' (Dexter 1988). Ecologically, soil consists of very heterogeneous habitats characterised by hot spots of biological activities such as rhizospheres, drilospheres, and aggregatospheres (Lavelle, Spain 2001). A defining signature of a soil/plant/soil biota system may well be this heterogeneity (Young, Ritz 2000). Through soil structure, soil provides the physical space that accommodates many organisms in ecological niches. Through interactions with soil water, soil structure defines a range of environmental factors which are important for the growth and activities of its inhabitants. In agriculture, a good soil habitat is one that provides favourable environment for growth and development of plants and associated organisms important for ecosystem functioning. Soil structure determines the abundance, diversity and activity of the soil biota and therefore biological fertility of the soil.

On the other hand, soil biota can modify soil structure by stabilising as well as creating soil structure. Bioturbation, which results in biogenic mixing of soil materials, is an important process in soil formation. It assists interactions with inorganic fractions eg burial of litter, and is important for nutrient cycling and nutrient availability. The products of soil biota activities, eg aggregates, burrows and galleries, often have dominant effects on soil physical properties such as infiltration, aeration and soil strength (Lee, Foster 1991). Therefore, the interactions of soil structure and soil biota determine all the three aspects of soil health, physical, chemical and biological.

In natural ecosystems, many soil structural features are adaptive strategies by soil biota to increase the suitability of the soil habitat for survival (Wolters 2000). They exist in dynamic equilibria with soil biota activities, counteracting natural processes of soil chemical and physical degradation. For example, integrity of soil aggregates found under natural ecosystems depends on the continuous supply of carbohydrate gel from microbes which can be severely curtailed under annual cropping (Foster 1992). The modified soil structure found under ant mounds, (lowered bulk density and increased soil porosity) accelerates infiltration of water, alters temperature gradients and modifies pH, and is maintained by the continuous activities of the ants. These activities change the decomposition rates and functional structure of microbial communities in the soil (Wolters 2000). The transmitting burrows created by native anecic earthworms ensure that little surface runoff and soil loss occur, and are the preferred areas of root proliferation in the highly acidic subsoil (Chan 2004). Risk of runoff increased dramatically after three years of conventional tillage due to the disappearance of these burrows.

These examples illustrate the impact of agricultural management practices on soil structure - soil biota interactions. In agriculture, management practices can have direct

effects on soil biota (Clapperton et al 2003) and indirect effects due to changed soil structure. Tillage and associated field machinery traffic destroy habitats of earthworms and result in soil compaction that can render the soil too strong for these ecosystem engineers to survive. The undesirable effects of conventional management practices on soil structure have been recognised and are main impetus for conservation tillage systems. An essential feature of the latter is the harnessing of the beneficial functions of soil biota. Hitherto, attention has been focussed only on the effect of soil structure on plant growth. However, such understanding should be extended to the effect of soil structure on soil biota and their interactions. Ecological functioning by soil biota has to be studied in the context of structural heterogeneity. Better understanding of these interactions will help us develop management practices to manipulate soil biota (Elliott and Coleman 1988).

References

Baker GH, Barrett VJ, Carter PJ, Williams PML, Buckerfield JC 1993. Seasonal changes in the abundance of earthworms (*Annelida : Lumbricidae* and *Acanthodrilidae*) in soils used for cereal and lucerne production in South Australia. Australian Journal of Agricultural Research 44 1291-1301.

Chan KY 2004. Impact of tillage practices and burrows of a native Australian anecic earthworm on soil hydrology. Applied Soil Ecology (in press).

Clapperton J, Chan KY, Larney J 2004. Managing the soil habitat for enhanced biological fertility. In Soil biological fertility: A key to sustainable land use in agriculture Eds Abbott L and Murphy D pp 203-224. Kluwer Academic Publishers, Dordrecht.

Darwin C 1811. The formation of vegetable mould through the action of worms with some observations on their habits. John Murray, London.

Dexter AR 1988. Advances in characterisation of soil structure. Soil & Tillage Research 11 199-238.

Elliott ET, Coleman DC 1988. Let the soil work for us. Ecological Bulletin 39 23-32.

Emerson WW, Foster RC, Oades JM 1986. Organic-mineral complexes in relation to soil aggregation and structure. In Interactions of soil minerals with natural organics and microbes. Soil Science Society American Special Publication 17 521-548.

Foster RC 1988. Microenvironments of soil microorganisms. Biology & Fertility of Soils 6 189-203.

Foster RC 1994. Microorganisms and soil aggregates. In soil biota: Management in sustainable farming systems Eds Pankhurst CE, Doube BM, Gupta VVSR, Grace PR pp 144-155. CSIRO Australia.

Hassink J, Bouwman LA, Zwart KB, Brussard L 1993. Relationships between habitable pore space, soil biota and mineralisation rates in grassland soils. Soil Biology & Biochemistry 25 47-55.

Kay BD 1990. Rates of change of soil structure under different cropping systems. In Advances in Soil Science Ed Stewart BA 12 1-41.

Lavelle P and Spain AV 2001. Soil ecology pp 426-529. Kluwer Academic Publishers, Dordrecht.

Lee KE, Foster RC 1991. Soil fauna and soil structure. Australian Journal of Soil Research 29 745-775.

Lee KE, Pankhurst CE 1992. Soil organisms and sustainable productivity. Australian Journal of Soil Research 30 855-92.

Marshall TJ, Holmes JW 1979. Soil physics. Chapter 3 pp 58-78. Cambridge University Press, Cambridge.

Papendick RI, Campbell GS 1985. Theory and measurement of water potential. In Water potential relations in soil microbiology Eds Parr JF, Gardner WR, Elliott LF pp 1-12. Soil Science Society of America, Madison, WI.

Smiles DE 1988. Aspects of the physical environment of soil organisms. Biology & Fertility of Soils 6 204-215.

Tisdall JM, Oades JM 1980. The effect of crop rotation on aggregation in a red-brown earth. Australian Journal of Soil Research 18 423-434.

van Vliet PCJ, Hendrix PF 2004. Role of fauna in soil physical processes. In Soil biological fertility: A key to sustainable land use in agriculture Eds Abbott L, Murphy D pp 61-80. Kluwer Academic Publishers, Dordrecht.

Wolters V 2000. Invertebrate control of soil organic matter stability. Biology & Fertility of Soils 31 1-19.

Wright DA, Killham, K, Glover LA, Prosser JI 1993. The effect of the location in soil on protozoal grazing of a genetically modified inoculum. Geoderma 56 633-640.

Young IM, Ritz K 2000. Tillage, habitat space and function of soil microbes. Soil & Tillage Research 53 201-213.

Contact details

Dr Yin Chan
Principal Research Scientist (soils)
Organic Wastes Recycling Unit
NSWDPI Locked Bag 4 Richmond NSW 2753
Phone 02 4588 2108
Fax 02 4578 2528
Email yin.chan@dpi.nsw.gov.au

Impact of management practices on soil biota activity on acidic clay loams in NSW

John Kirkegaard
CSIRO Plant Industry, Canberra

Introduction

All management practices affect the numbers, diversity, and activity, both spatially and temporally, of various components of the soil biota. Apart from plant-associated organisms such as pathogens or symbionts, studies on the numbers and diversity of soil biota have, in themselves, rarely improved agricultural decision-making. What is important is how the interactions between management and soil biology affect crop performance (Figure1). What the study of soil biology offers us is insight into how we might improve agronomic practices and cultivars: that is, how we might accelerate the adaptive management of Australian farms that has enabled rises in productivity to keep ahead of the steadily falling terms of trade over the last few decades.

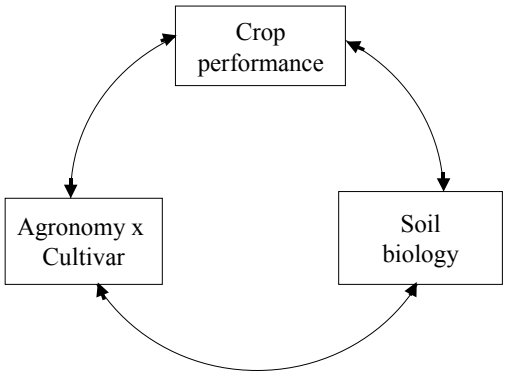


Figure 1. Interactions among management, crop performance, and soil biology that can accelerate adaptive management.

On the acidic loams of southern NSW, there is clear evidence that changes in agronomic practice have improved the productivity and sustainability of farming systems by influencing (among other things) important components of soil biology. There is substantial evidence, from fumigation experiments and from as yet unexplained agronomic responses in field trials, that there is potential to capitalise further on interactions between crop management and soil biology. It is likely that studying the rhizosphere, the interfacial region between roots and soil, will foster the fastest progress.

There is equally clear evidence that management strategies designed to improve soil 'health' may not improve productivity and can even threaten other aspects of sustainability. Growers and agricultural extension specialists should be aware that the concepts of soil 'quality' and soil 'health' that have gained popularity among soil biologists and microbiologists in the last decade or two have received trenchant criticism (eg Sojka et al 2003). The problem is that these concepts are inherently subjective, without agreed meaning, as is evidenced by trying to endow them with units. In this paper I elaborate on these points, and especially emphasise the strong

interactions between cultivars, agronomic practice, and soil biology that offer promise of further substantial improvements in sustainable productivity.

Dryland farming systems on acidic loams: recent trends

The dry-land farming system on acidic loams in southern NSW is predominately mixed farming enterprises based on cereal and sheep production, ranging in annual rainfall from 650 mm in the east to 400 mm in the west. The average farm size of around 1010 ha comprises 67% pasture and 33% crop, most of which (around 68%) is cereal, predominately wheat, the remainder being broadleaf break crops such as canola, lupin and field pea (Connell, Hooper 2002). These averages vary considerably within individual farms and paddocks, with flexibility to adjust to seasonal conditions and commodity prices. The large proportion of farm area devoted to legume-based pastures in sequence with a cropping phase is an important feature of these systems. The pasture phase results in a build-up of labile soil organic matter and biologically fixed nitrogen, as well as providing different opportunities to control problem weeds, although the acidifying effects of legume pastures must be controlled with lime. Further, when perennial species are used, especially lucerne, they enable the capture of water from the deep subsoil that may have drained beyond the reach of crop roots, thus improving the hydrologic performance of the system. The long-term productivity trends for wheat within these systems have been among the highest in Australia, especially in the 1990s (Angus 2001), and have been accompanied by several changes that improve physical resource sustainability, including increased adoption of minimum tillage (80% of crops in 2001 – Connell, Hooper 2002), increased use of lime (Angus 2001), and a move to more perennial pastures (ABS Agstats 2001). There has been recent concern about the risks of simplifying the rotations with shorter or less frequent pasture phases and continuous alternation of wheat with a broadleaf crop (eg wheat-canola-wheat-canola) (Wolfe, Cregan 2003). The static and declining canola yields in the area may indicate increasing disease problems associated with these intensive rotations, and despite impressive improvements, wheat yields in most shires remain below the water-limited potential. Future trends are likely to include an increase in the area of dual purpose winter wheat for grazing and grain production, larger farms and a continued focus on improved productivity to keep pace with the continuing 3-4% pa decline in the terms of trade.

Impacts of crop and pasture sequence on soil biology and crop production

Legumes, nitrogen and organic matter

The nitrogen-fixing activities of the legume-*Rhizobium* symbiosis in legume-based pastures and pulse crops represent one of the greatest benefits of their inclusion in the farming system. Because the amount of nitrogen fixed by legumes is related to legume biomass – 20 to 25 kg of shoot nitrogen fixed for every tonne of legume biomass produced (Peoples, Baldock 2001, Evans et al 2001), any management practices that enhance the growth of legumes will increase this nitrogen benefit within the system. Recent estimates suggest that wheat crops derive up to 40% of their nitrogen requirement from preceding legume crops (Evans et al 2001) and the impacts of various management strategies on the mineralisation and efficient use of this organic nitrogen source remain an active area of research (GRDC Project CSO000030). Importantly from a soil biology viewpoint, the frequency of highly productive legume-based pastures in the farming system of the area provides greater inputs of labile carbon and nitrogen to the system, than provided by crop residues, for sustaining microbial activity.

Break crops and soil biology

The substantial productivity improvements in wheat in southern NSW in the last decades arise from the control of cereal root diseases, primarily take-all (*Ggt*), by the use of broadleaf break crops such as canola, lupin and field pea, and the spray-topping of grass hosts in pastures in the year prior to cropping. Reductions in other soil-borne diseases such as crown-rot (*Fusarium pseudograminearum*) and stubble-borne diseases such as yellow leaf spot (*Pyrenophora tritici-repentis*) have also contributed to these benefits. Break crops result in average wheat yield increases of 19%, and cereal crops freed from root disease respond more reliably to nitrogen fertiliser and use more of the subsoil water and nitrogen, which may otherwise be leached (Angus et al 2001). The combination of break crops, improved nitrogen management and liming has lifted productivity to a new level. Diseases common to wheat and canola (eg rhizoctonia, lesion nematodes *Pratylenchus neglectus*) or other inhibitory organisms may continue to reduce yield in the area, as suggested by significant responses to soil fumigation (see later).

Non-hosting of disease pathogens is not the only way in which break crops can influence the soil biology and crop production. Such crops can also influence the populations of specific rhizosphere organisms which may compete, antagonise or suppress pathogens, influence nutrient transformations or affect plant growth directly. We have conducted much research in southern NSW to investigate the impacts of canola on some of these other aspects of soil biology. A brief summary follows.

Biofumigation

When wheat grew better after canola than after other broadleaf break crops in southern Australia during the early 1990s it prompted speculation that chemicals called isothiocyanates (ITCs) released from canola roots may suppress disease organisms, a process termed biofumigation. Early pot studies showed that cereal pathogens such as take-all were sensitive to ITCs, but subsequent field studies found only limited benefits to following wheat crops from a biofumigation effect (Kirkegaard et al 2000, Smith et al 2004). Similar studies in northern NSW also failed to find direct biofumigation effects on crown rot, although the disease was lower after canola than after chickpea (Kirkegaard et al 2004). It now seems that any impacts of canola on root disease in addition to the non-host effect, relate to more general changes in rhizosphere bacteria (eg Rumberger, Marschner, 2003) or antagonistic organisms such as *Trichoderma* (Kirkegaard et al 2004) rather than direct killing of disease inoculum by ITCs.

N mineralisation

More evidence that break crops can significantly affect soil biology was that different amounts of mineral nitrogen accumulated in the summer fallow following different crops (Kirkegaard et al 1999). Surprisingly, more mineral nitrogen accumulated following canola (94 kg/ha) than following field pea (50 kg/ha) and the differences could not be explained by the amount, nitrogen content or carbon:nitrogen ratio of the crop residues. What caused this effect is uncertain, but populations of organisms associated with nitrogen-cycling such as free-living nitrogen-fixing bacteria, *Azospirillum* species and ammonium-oxidising bacteria were generally *lower* following canola, while total bacterial populations did not differ. The effect was shown to be transitory under laboratory conditions which accelerated mineralisation, but in the field it strongly influenced the growth of subsequent wheat crops.

Mycorrhizal fungi (AMF) and other beneficial organisms

The realisation that canola may release fungicidal compounds from the roots prompted speculation that more frequent use of canola may influence the levels of beneficial organisms such as arbuscular mycorrhizal fungi (AMF) or *rhizobia*. Low colonisation of wheat crops following canola or fallow results in poor growth of wheat on low phosphorus soils in the northern wheat belt (Thompson et al 2001). Recent studies on wheat in southern Australia showed that lower AMF colonisation in wheat following brassicas (and fallow) did not reduce growth or yield in subsequent wheat crops despite strong phosphorus limitations on crop growth and yield (Ryan, Angus 2003). The authors hypothesised that AMF may even be parasitic on these crops prior to spring, utilising carbohydrates from the seedlings to support their growth. Surprisingly, it is possible that wheat in southern NSW may grow better after canola partly as a result of lower AMF colonisation. Smith et al (2004) found no evidence that canola influenced the nitrogen-fixing capacity of subsequent pea crops. Populations of the disease antagonist *Trichoderma* have increased more in wheat following canola than following chickpea or wheat (Kirkegaard et al 2004).

Tillage management and the soil biology: good and bad

Conservation cropping systems involving direct-drilling and stubble retention have been developing for over 30 years in Australia, initially in response to concerns about fuel costs and more recently to reduce erosion, maintain organic matter and soil structure, and conserve water for crop use. During that period, the systems have been tuned to specific regions, especially by growers adapting sowing equipment to their circumstances (Cornish, Pratley 1987). In southern NSW, particularly in higher rainfall areas, adoption has been slower and the benefits to crop yields flowing from improved soil conditions more difficult to demonstrate (Kirkegaard 1995, 2001).

Research carried out over the last 15 years at a site near Harden in southern NSW helps illustrate the role of soil biology in the productivity and sustainability of conservation cropping. From the outset, a consistent problem with direct-drilled wheat in southern Australia was the reduced early vigour of crops compared with those sown into cultivated soil, a phenomenon which has not diminished over time (Kirkegaard et al 1995, Simpfendorfer 2002). The surprising results of Chan et al (1987) and later Kirkegaard et al (1995), showing that soil fumigation could overcome the early growth reductions pointed to the role of soil biological constraints. A subsequent investigation at 39 farm sites over three years in southern NSW (Simpfendorfer et al 2002) showed that the problem was widespread (62% of sites), was not related to any of the major soil-borne cereal disease organisms, nor to general changes in soil biology, but was strongly related to the inhibitory activity of *Pseudomonas* bacteria isolated from the rhizosphere of wheat seedlings at each site. The reduced early vigour at the Harden site reduced yield by 11% in wheat over that period and increased the amount of residual water and nitrogen left in the subsoil (Kirkegaard et al 2001).

During the course of our research to understand what reduced growth in direct-drilled wheat, the Harden site was used by many other researchers investigating the impact of management on other aspects of the soil biology. The widely promoted benefits to soil 'health' under direct-drill systems were evident there. Increases in soil organic matter (Pankhurst et al 2002), microbial biomass (Gupta et al 1994), populations of earthworms (Doube et al 1994), nematode diversity (Hodda et al 1997), faunal diversity (Longstaff et al 1997) and disease suppression (Pankhurst et al 2002) were all seen on

direct-drill/stubble retained treatment compared with late burn/single tine cultivation treatment, the most widely practised management in the area.

The dilemma for researchers and advisers highlighted by this example is that in spite of the apparent improvements in soil 'health', the growth and productivity of crops throughout the 15 year period were lower on the direct-drill/stubble retained treatment, and this was associated with more water and mineral nitrogen left in the subsoil, representing an increased risk of deep drainage/nitrogen leaching. This scenario may not be true for all farming systems, but it highlights the need to be pragmatic about the benefits of management to preserve various components of the soil biology for their own sake, and to keep in mind the other important aspects of the production system.

Plant root systems: an important part of soil biology

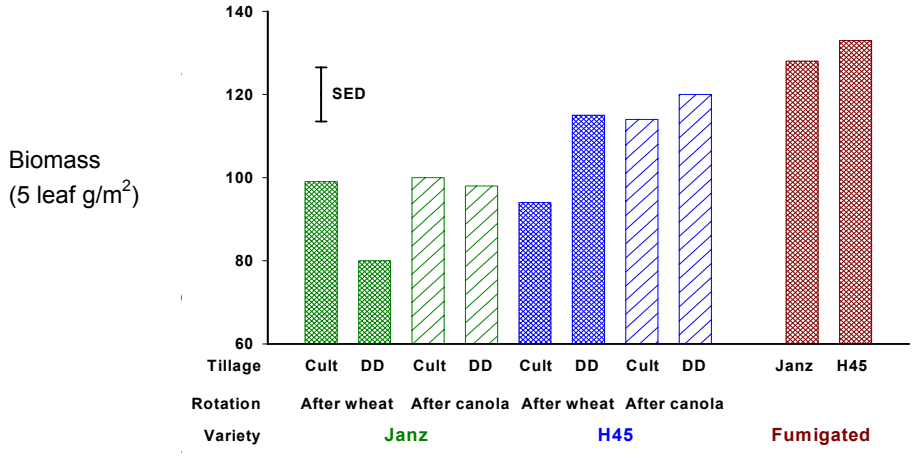
The most likely mechanism underlying the build-up of inhibitory bacteria on direct-drilled crops occurs in the rhizosphere, and is related to an interaction between soil hardness and an accumulation of pseudomonads on constrained, slow-growing roots (Watt et al 2003). Direct-drilled wheat roots grow at half the rate of roots in cultivated soil, contort around soil structure twice as often and have much shorter growth zones around the tips (Watt et al 2001 and unpublished). *Pseudomonas* bacteria accumulate on these slow-growing tips while the general bacterial population do not, likely related to growth and patterns of compounds released from roots (Watt et al 2003). If slow-growing roots are indeed more prone to being colonised by inhibitory pseudomonads, it could explain why management strategies such as early sowing into warmer soils and cultivation below the seed, both of which increase seedling root growth rates, may reduce the impact of direct-drilling on early growth. It could also mean that selecting wheat varieties with inherently fast rates of root growth may help reduce the impacts of hard soil on crop growth.

There are other examples of the ways in which plant roots can generate specific rhizosphere populations (excluding disease) which can have significant impacts on crop production. A recent example is that reported by Gupta et al (2003) in which specific populations of bacteria associated with some wheat varieties can persist in soil and influence the growth of subsequent wheat crops. Understanding the basis for these effects may provide opportunities to select or develop crop varieties that possess beneficial characteristics, and to exploit important interactions by matching the right variety to appropriate management. We recently investigated the interactions between tillage, rotation and wheat varieties in a field experiment in southern NSW, using fumigation as a tool to investigate the role of soil biology in the observed responses (Figure 2). The yield was drought affected, but the early biomass data demonstrated significant interactions between variety, tillage and rotation in the unfumigated treatments (on the left).

Janz biomass was reduced under direct-drilling following wheat but not following canola, while H45 had a different response to these management practices. Under fumigation there was no management or variety effects observed (only means for each variety are shown) indicating the role of the soil biology in the responses in the unfumigated soil.

The results suggest there is considerable potential to exploit the interactions between variety and management within cropping systems although an understanding of the mechanisms involved will be necessary to provide a basis for varietal selections. In the case of direct-drilling it may be the rate of early root growth which benefits some varieties, while different root exudates may influence the bacterial populations in the rhizosphere of different wheat varieties. In addition, the results indicate that biological constraints may still exist for some varieties even with good rotation and tillage management (eg cf Janz/ DD/Canola with Janz fumigate). Fumigation eliminated these interactions (overall means shown on right).

Figure 2. Interactions between tillage, rotation and variety on wheat biomass (left).



Organic farming

Organic/biodynamic farming places particular emphasis on creating a diverse and vigorous soil biological community. It is often assumed that the soil biology on organic farms will adapt to the elimination of soluble fertilisers and other inputs to play a larger role in plant nutrition and growth. In some of the most comprehensive comparisons of organic/biodynamic with conventional systems on both grain and dairy farms on acidic loams in south-eastern Australia, it was phosphorus deficiency which most limited the development of an effective soil biological community (Ryan 2003). There was no evidence that biodynamic preparations enhanced the biomass or functions of the soil biological community, and it was wrong to assume that inputs permitted on organic farms are friendly towards the soil biota. Furthermore, there was no evidence that organic management had a consistent positive benefit for grain nutritive value. While there may be economic, philosophical or personal reasons for adopting organic, biodynamic or similar farming systems, there is no clear evidence that a more effective soil biological community results, and on the generally phosphorus-deficient acidic loams, a real risk that overall system productivity and sustainability will be limited if phosphorus inputs are not matched with outputs.

Conclusions

Management practices (including varietal selection) can substantially affect the sustainability and profitability of farming systems via interactions with important components of the soil biology. Recent research offers promise of further improvements in this regard. The vague term ‘soil health’ does not relate well to either the broader environmental performance or the productivity of farming systems. The capacity of systems to achieve and sustain well-established physiologically-based productivity potentials remain the most useful benchmarks.

References

Angus JF 2001. Nitrogen supply and demand in Australian agriculture. Australian Journal of Experimental Agriculture 41 277-288.

Angus JF, Kirkegaard JA, Peoples MB 2001. Rotation, sequence and phase: research on crop and pasture systems. 10th Australian agronomy conference. www.regional.org.au/au/asa/2001/plenery/4/angus.htm

Chan KY, Mead JA, Roberts WP 1987. Poor early growth and yield of wheat under direct-drilling. Australian Journal of Agricultural Research 38 791-800.

Connell P, Hooper S 2002. Australian grains industry 2002. Report of the Australian agricultural and grazing industries survey of grain farms, ABARE Canberra.

Cornish PS, Pratley JE 1987. Tillage: New directions for Australian agriculture. Inkata Press.

Doube BM, Buckerfield JC, Kirkegaard JA 1994. Short-term effects of tillage and stubble management on earthworm populations in cropping systems in southern New South Wales. Australian Journal Agricultural Research 45 1587-1600.

Evans J, McNeill AM, Unkovich MJ, Fettell NA, Heenan DP 2001. Net nitrogen balances for cool-season grain legume crops and contributions to wheat nitrogen uptake: A review. Australian Journal of Experimental Agriculture 41 347-359.

Gupta VVSR, Roper MM, Kirkegaard JA, Angus JF 1994. Changes in microbial biomass and organic matter levels during the first year of modified tillage and stubble management practices on a red earth. Australian Journal of Soil Research 32 1339-54.

Gupta VVSR, Roget DK and Coppi JA 2004. Identification of a previously unrecognised biological constraint to yield in sequential wheat crops. Proceedings of the 3rd Australasian soil-borne disease conference, Adelaide.

Hodda M, Stewart E, FitzGibbon F, Reid I, Longstaff BC and Packer I 1997. The identification of free-living soil-dwelling nematode assemblages as indicators of sustainable soil use. RIRDC project report CSE-73A.

Longstaff BC, Greenslade PJM, Colloff M, Reid I, Hart P, Packer I 1997. The impact of soil tillage practice on soil fauna in the NSW wheatbelt. RIRDC project report 99/18.

Kirkegaard JA 1995. A review of trends in wheat yield responses to conservation cropping in Australia. Australian Journal of Experimental Agriculture 35 835-48.

Kirkegaard JA, Howe GN, Mele P 1999. Enhanced accumulation of mineral-N following canola. Australian Journal of Experimental Agriculture 39 587-593.

Kirkegaard JA, Sarwar M, Wong PTW, Mead A, Howe G, Newell M 2000. Field studies on the biofumigation of take-all by Brassica break crops. Australian Journal of Agricultural Research 51 445-56.

Kirkegaard JA Howe GN Simpfendorfer S, Angus JF, Gardner PA, Hutchinson P 2001. Poor wheat yield response to conservation cropping: Causes and consequences during 10 years of the Harden tillage trial. 10th Australian agronomy conference. www.regional.org.au/au/asa/2001/session4/c/5-177-KirkegaardJ.htm.

Kirkegaard JA, Simpfendorfer S, Holland J, Bambach R, Moore KJ, Rebetzke GJ 2004. Effect of previous crops on crown rot and yield of durum and bread wheat in northern NSW. Australian Journal of Agricultural Research 55 321-334.

Pankhurst CE, McDonald HJ, Hawke BG, Kirby CA 2001. Effect of tillage and stubble management on chemical and microbiological properties and the development of suppression towards cereal root disease in soils from two sites in NSW, Australia. Soil Biology & Biochemistry 34 833-840.

Peoples MB, Baldock JA 2001. Nitrogen dynamics of pastures: Nitrogen fixation inputs, the impact of legumes on soil nitrogen fertility, and the contributions of fixed nitrogen to farming systems. Australian Journal of Experimental Agriculture 41 327-346.

Rumberger A, Marschner P 2003. 2-phenylethylisothiocyanate concentration and microbial community composition in the rhizosphere of canola. Soil Biology & Biochemistry 35 445-452.

Ryan MH, Angus JF 2003. Arbuscular mycorrhizae in wheat and field pea crops on low P soil: increased Zn-uptake but no increase in P-uptake or yield. Plant & Soil 250 225-239.

Ryan MH 2003. Going 'organic' may not improve soil health. Farming Ahead 137 36-39.

Simpfendorfer A, Kirkegaard JA, Heenan DP, Wong PTW 2002. Reduced early growth of direct-drilled wheat in southern New South Wales: Role of root inhibitory pseudomonads. Australian Journal of Agricultural Research 53 323-331.

Smith BJ, Kirkegaard JA, Howe GN 2004. Impacts of Brassica break crops on soil biology and yield of following wheat crops. Australian Journal of Agricultural Research.

Sojka RE, Upchurch DR, Borlaug NE 2003. Quality soil management or soil quality management: Performance vs semantics. Advances in Agronomy 79 1-68.

Thompson JP, Owen KJ, Clewett TG 2001. Pre-cropping with canola decreases vesicular-arbuscular mycorrhiza and growth of wheat in low P soil. Proceedings of the 2nd Australasian soil-borne root disease symposium Lorne 2001 pp 150-151.

Watt M, Kirkegaard JA, Simpfendorfer S, Rebetzke G, McCully ME 2001. Variation in wheat genotypes to no-till farming: Responses to soil biology and structure. Proceedings 6th International Society of Root Research (ISRR) symposium Nagoya Japan pp 424-425.

Watt M, McCully ME, Kirkegaard JA 2003. Soil strength and rate of rot elongation alter the accumulation of *Pseudomonas* spp. and other bacteria in the rhizosphere of wheat. Functional Plant Biology 30 483-491.

Wolfe E C and Cregan P D 2003. Smart rotations: Farming systems for the future. In Principles of field crop production, 4th edition. Ed Pratley JE pp 294-320. Oxford University Press, Sydney.

Contact details

Dr John Kirkegaard
Principal Research Scientist
CSIRO Plant Industry
GPO Box 1600 Canberra ACT 2601
Phone 02 6246 5080
Fax 02 6246 5399
Email John.Kirkegaard@csiro.au

Soil biology and crop production in Western Australian farming systems

Dan Murphy, Nui Milton, Mahdi Osman, Frances Hoyle, Lyn Abbott, Richard Cookson, Sigit Darmawanto
University of Western Australia

Introduction

Agricultural management practices ultimately seek to optimise plant and animal productivity within the overriding constraints of both climate and the capacity of the soil (physical, chemical and biological attributes) to support plant growth (Abbott, Murphy 2003). While optimal physical and chemical conditions of the soil for plant growth are often well defined, we have a much poorer understanding of the control that biological factors, particularly non-pathogenic associations, have on plant growth. The objective of this paper is to examine the relative contribution of soil biological attributes to crop production in Western Australian farming systems. Once these key attributes have been identified, management practices can be selected that take into account the potential for enhanced soil biological fertility and improved yield.

Western Australian farming system

The grain production zone (wheat belt) in Western Australia covers an area of more than seven million ha. Grain production is primarily restricted to areas where average annual rainfall is between 325 and 750 mm, the majority of which falls during the growing season (late autumn-late spring) in the south-west of Australia. Major soils in this region (Chromosols, Sodosols, Kandosols) are highly weathered with low surface clay and soil organic matter contents. The summer weather pattern is typified by hot dry conditions with infrequent storm events, largely restricting production to an annual winter cropping phase. Low winter rainfall and dry summers therefore constitute the primary constraint to organic matter production and accumulation. A lack of new plant residues and root exudates to provide a carbon food source in the soil, and problems associated with desiccation over summer as surface soil temperature peaks above 40°C, present significant challenges to the buildup of biological components in soil compared with temperate environments. However, this does not mean that soil biology is not important. Indeed, the Western Australian farming system is reliant on a cyclic pattern of biological activity which ‘explodes into action’ with rainfall and then slows at the onset of soil drying.

The relatively low growing-season rainfall and the inherently low capacity of major soil types in WA to retain water and plant nutrients are realised in poorer crop growth. Low potential yields have thus resulted in relatively low input systems, and these systems are therefore more reliant on biologically fixed nitrogen and organic matter decomposition to supply plant available nutrients and support crop production. In southern Australia for example, Angus (2001) calculated that, on average, 80% of crop uptake was supplied via biological processes, so the amount of nitrogen cycling through a WA soil during the growing season can be more than enough to satisfy crop nitrogen demand (43-122 kg/N ha, Murphy et al 1998), even where no fertiliser is applied. The exceptions to this are soils with a high leaching potential, which can result in the loss of both water and mobile nutrients below the rooting zone, and soils where microbial immobilisation of nitrogen out-competes plants for nitrogen availability (eg decomposing plant residues with high carbon:nitrogen ratio). Strategically timed or split

fertiliser applications (generally 20-80 kg N/ha) are therefore used to overcome the difficulties of matching biological nutrient supply with plant demand. Developing management strategies to improve asynchrony (microbial nutrient supply occurring when plant demand is low) and synlocation (plant-available nutrients being located in the soil matrix where there are no plant roots) is often difficult but essential for future sustainable production (Murphy et al 2004, Ridley et al 2004, Hoyle, Murphy this proceedings).

Identifying soil constraints to crop production

From 1960 to 1990, the average wheat grain yield in 62 WA shires was 1.9 t/ha, with less than 5% of shires assessed in 1990 having reached 50% of their rainfall-limited yield potential (Hoyle, Anderson 1993). In our current research we have used the WA-Wheat model (Department of Agriculture), which has been developed as a front-end system for the APSIM model, to target districts that consistently under-perform. To do this, WA-Wheat was used to initialise (seeding date, varietal maturity, fertiliser application, actual rainfall, soil type) model simulations (1960-2001) on a shire basis for comparison against actual historical yields. Where potential yield is not achieved our approach has been to assume that this is the result of inappropriate management practices and/or soil physical, chemical or biological constraints to crop production (Figure 1).

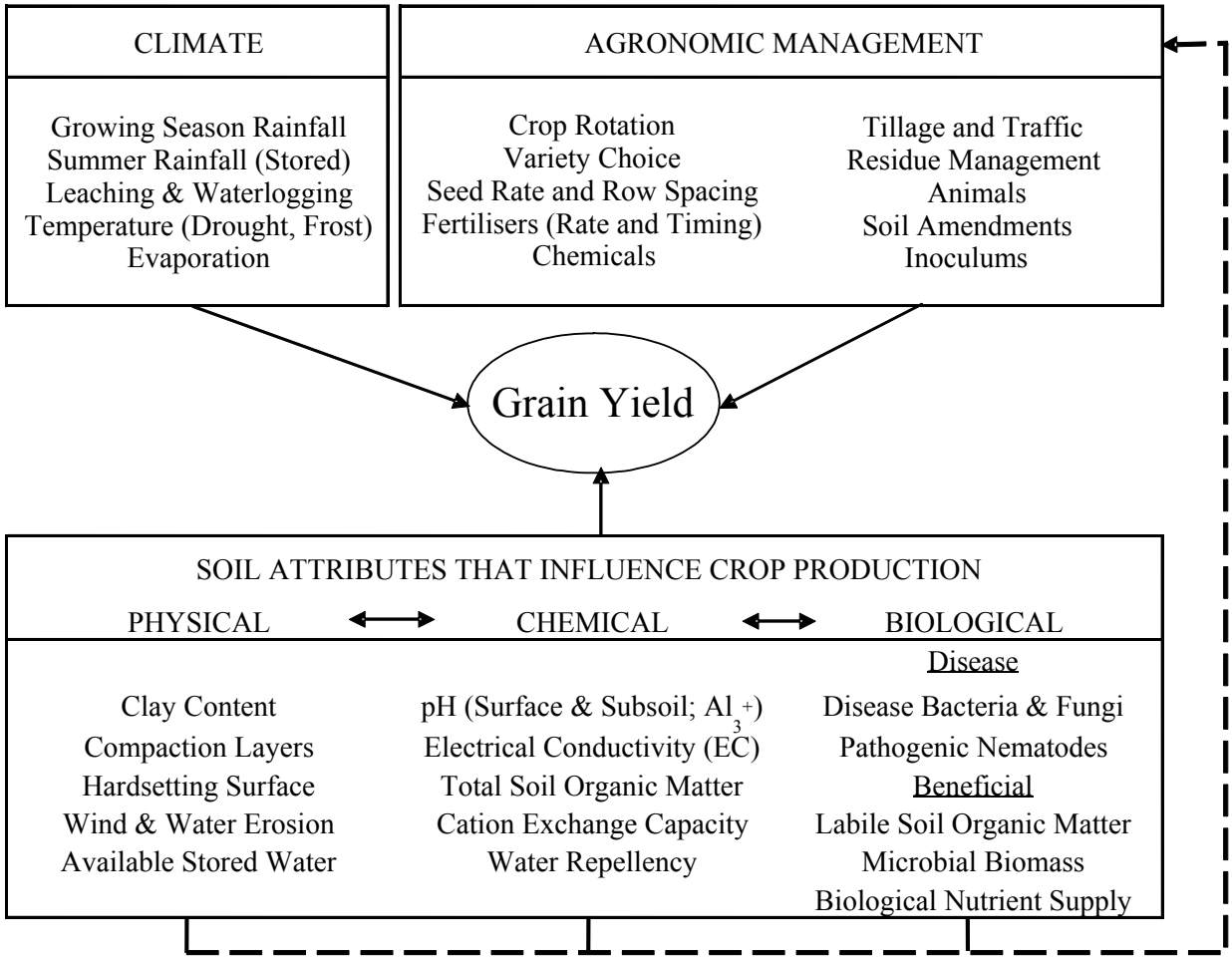


Figure 1. A conceptual model of climatic and agronomic factors along with key soil physical, chemical and biological constraints to yield production in Western Australian farming systems.

Once soil constraints are identified their economic importance can be assessed, so that the cost and practicality of removing the constraint versus potential yield benefit is known before implementing changes in agronomic practice. This approach focuses on discrete soil attributes that have a known direct impact on crop production, and can be measured and interpreted in the context of management solutions. This approach provides an economic evaluation of 'cause' and 'effect', enabling prioritisation of high return solutions to overcome major agronomic and soil limitations instead of placing effort in further detailed site characterisation which is not feasible over a large scale.

Identifying soil constraints to crop production: a case study

Evaluation of the 'soil indicator' package described in Figure 1 was achieved by collecting climatic, agronomic and soil data from 40 paddocks on 20 farms in two adjoining catchment groups (named 'A' and 'B' for simplicity). Paddocks were located within a 10 x 20 km region and were chosen in consultation with growers to either compare high and low yielding areas, or encompass soils that consistently under/over performed against expected yields. Within each paddock three field replicates were established, and within each replicate area soil was collected in 0-5, 5-10, 10-30, 30-60 and 60-90 cm layers for laboratory analysis (in triplicate). Rainfall was recorded at each farm and agronomic data supplied through a one-on-one interview and questionnaire process with the principal grower in each farming unit. Grain yield cuts were taken by hand within a few days prior to machine harvest.

Using figures from the shire that includes A and B catchments, we compared the WA-Wheat model's predicted achievable grain yield against historical records (1960-2001) of actual average grain yield (Figure 2). In approximately 50% of years, we observed good agreement between actual and predicted yield, but in 20 of the 43 years there was a difference of greater than 0.8 t/ha in predicted yield compared with actual yield. Given the low average historical grain yield for wheat in this region (1.58 t/ha), this would represent a significant yield benefit if obtainable. Actual yield data from the 40 paddocks illustrate that on a site by site basis actual yield can vary considerably (mean = 2.5 t/ha, min = 0.44 t/ha, max = 4.74 t/ha) within a season (Figure 2) and can reach the same upper range as predicted by the model .

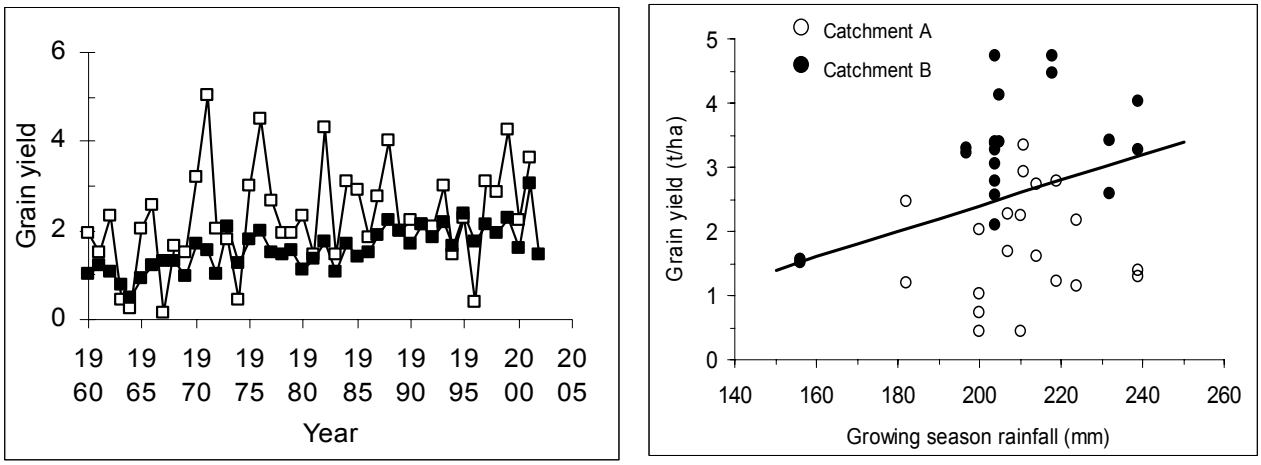


Figure 2. Left: Actual (filled squares) and modelled (open squares) grain yield (t/ha) for the shire that contains catchments A and B. Right: Measured grain yield from the 40 paddocks plotted against growing season rainfall for each site. The solid line represents an achievable grain yield. Paddocks below this line are underperforming and those above the line are above reasonable expectation.

The independent influence of rainfall, inorganic nitrogen fertiliser and soil constraints (as listed in Figure 1) on grain yield was determined using bivariate regression analysis (Table 1). In this regression analysis data for diseases (take-all, rhizoctonia) and pathogenic nematodes (*Pratylenchus neglectus*, *P. thornei*) were excluded as their occurrence was below detection limits or low in 38/40 paddocks. Biological nutrient supply was assessed solely as potentially mineralisable nitrogen in the regression analysis. Mycorrhizal bioassays were performed to determine their importance to plant nutrient supply. More than 30% of root length colonisation is required to obtain benefits of plant nutrient acquisition from mycorrhizal associations (Abbott, unpublished critical value). However, mycorrhizal root length colonisation in the plant bioassays performed was between 0-30% as the paddocks were sufficient in bicarbonate-extractable phosphorus.

Table 1. Mean values for attributes determined in catchments A and B and results of bivariate regression analysis whereby climatic, agronomic and soil physical, chemical and biological attributes were assessed for their individual influence on wheat grain production across the 40 paddocks. Average grain yield was 1.76 and 3.24 t ha/ in catchments A and B respectively. All significant attributes have been presented; most non-significant attributes assessed have been removed. (The same letter denotes no significant difference between catchments for that attribute.)

	Attribute	Catchment		Coefficient[a]	P-value[b]	Variability
		A	B			Explained[c]
Climate	Rainfall (mm)	211a	206a	-	ns	3.7
Agronomy	N fertiliser (kg N/ha)	20a	24a	0.02	0.055*	9.4
Physical	Clay content[d] (%)	11.0a	10.4a	0.08	0.062*	9.1
Chemical	Total carbon (t C/ha)	9.0a	10.8b	-	ns	0.2
	pH ($CaCl_2$)	5.7a	5.6a	-	ns	0.4
	EC[d] (mS/m)	80a	63b	-	ns	0.1
Biological	Labile C (kg C/ha)	83a	118b	0.01	0.041**	10.5
	Microbial biomass C (kg C/ha)	107a	183b	0.01	0.001***	30.3
	PMN (kg N/ha)	7.0a	10.1b	0.14	0.003***	21.2

[a]The coefficient can be interpreted as t/ha grain yield change per unit change in attribute.
[b*] = significant P<0.10; ** = significant P<0.05; *** = significant P<0.01; ns = not significant.
[c]The variability explained has a maximum of 100% and is not additive between individual attributes.
[d]clay and EC data were assessed using robust regression analysis due to unusual data points. EC = Electrical conductivity. .

Measured yields from catchment B were significantly higher than in catchment A, which is reflected in some, but not all of the soil attributes used in the regression analysis (Table 1). It is notable that the biological attributes explained the greatest amount of variability in yield between the 40 paddocks. For example nitrogen fertiliser and clay content each explained 9% of the variability. Potentially mineralisable nitrogen, an index of biological nitrogen supply, explained 21%. Microbial biomass explained 30% (Table 1). Growing season rainfall was not significantly related to grain yield, although we have already argued that rainfall is the primary driver of production in this environment. However, this was not surprising as we would only expect a strong relationship between growing season rainfall and yield if there were no other constraints to crop production. Over a 10 mm growing season rainfall gradient (200 to 210 mm), there was a grain yield variation from 0.5 to nearly 5.0 t/ha (Figure 2). Thus there was

certainly either poor agronomic management and/or the influence of soil constraints on crop production.

Combinations of significant factors that influenced grain yield were then determined using ordinary least square multiple regression analysis. Using a multiple regression model that included all nine parameters listed in Table 2 we were able to explain 42% of the yield variability (regression model not shown).

Several soil attributes were identified that did not have a significant direct influence on grain yield; but may have had an indirect influence through their effect on the size of the microbial biomass (Figure 3). In this case, 66% of the variability in microbial biomass could be explained by clay content (log transformed data), pH and labile carbon. In other words, providing an optimal physical and chemical soil matrix along with an available carbon (food) source was the primary basis for improving the mass of soil microorganisms in these soils. This is logical given microorganisms, like all other living organisms, function more effectively within an optimal environment and provided with a suitable food source. Removing attributes that were either directly related to microbial biomass, or those that were not significantly affecting grain yield from the initial model, resulted in the development of a simpler model to explain the variability in grain yield (Figure 3). This model, which consisted of growing season rainfall, nitrogen fertiliser and microbial biomass as the only three attributes used, still explained 40% of the variability in grain yield. This means that by removing six attributes from the initial model we only lost 2% of explained variability; but removed a considerable amount of the analytical measurements that would be required.

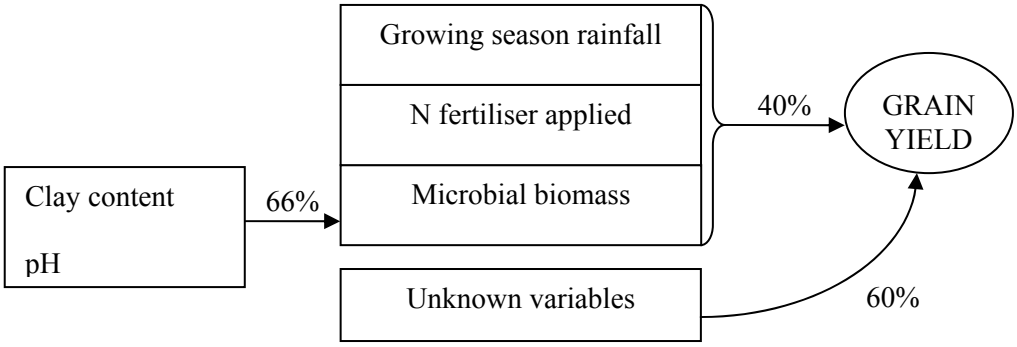


Figure 3. A schematic representation of the multiple regression analysis models used to describe microbial biomass (explanatory variables: nlogClay content*pH*Labile Carbon) and grain yield (explanatory variables: Growing season rainfall*N fertiliser applied*Microbial Biomass).

Further analysis indicated that the influence of the microbial biomass on yield was predominately due to the strong relationship ($r^2 = 0.77$) to potentially mineralisable nitrogen. Thus the model used to describe grain yield could alternatively be expressed as growing season rainfall, nitrogen fertiliser and potentially mineralisable nitrogen with a similar percentage of the grain yield still being explained (data not shown). This provides a simple water and nitrogen availability story as the key drivers of grain production in this environment, which is supported by the fact that water is essential for plant growth and that nitrogen is the primary nutrient limiting crop production throughout the world.

Rapid prediction of potentially mineralisable nitrogen using mid infrared technology

Our current research has demonstrated that potentially mineralisable nitrogen (PMN) can be successfully predicted using mid infrared technology (Murphy et al 2004, Murphy, Milton this proceedings). The major advantage of mid infrared prediction over conventional laboratory analysis of PMN is that it enables rapid (two minute) and cost efficient analysis of a soil biological attribute that has a direct impact on yield production. For example, a one unit increase in PMN caused a 0.14 t/ha yield increase, Table 1. The accuracy of mid infrared to predict the within-paddock variability in PMN is illustrated in Figure 4.

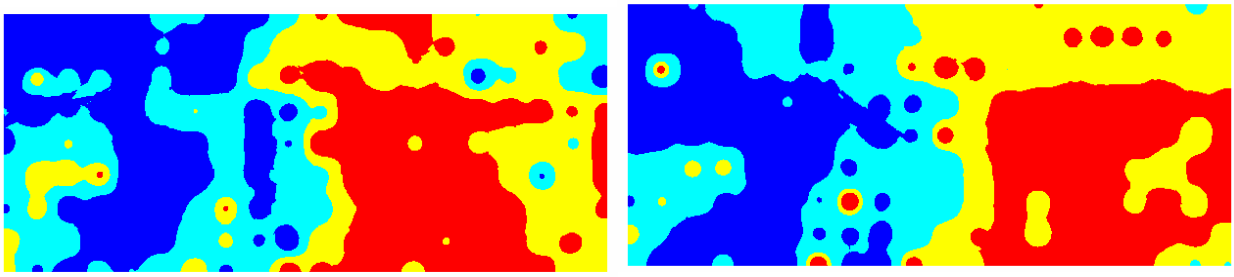


Figure 4. Spatial maps (10 ha) of potentially mineralisable ntirogen determined analytically using traditional biochemical analysis (left) and predicted using mid infrared technology (right) for the 0-10 cm layer of a Western Australian agricultural soil. Data categorised into four categories (Murphy et al 2004).

Soil was collected using a 25m x 25m sampling grid (180 sampling points over 10 ha) from one of the 40 paddocks. Over this 10 ha area PMN ranged from 4-32 kg N/ha. PMN was determined using conventional biochemical laboratory analysis and also predicted (on the same soil samples) using a mid infrared calibration curve that was developed from an independent data set. There was good agreement between mid infrared predicted and measured PMN ($r^2 = 0.70$) which is illustrated by the degree of similarity in the measured and mid infrared predicted spatial maps (Figure 4). While mid infrared is not 100% accurate at predicting PMN, it is of sufficient accuracy for categorising soils or zones within a paddock into poor, low, moderate and high biological soil nitrogen supply, which could be used to adjust for inorganic nitrogen fertiliser application rates.

Management options to enhance soil biological fertility

Despite the identification of known soil constraints to grain yield, 60% of the variability in wheat grain yield is still not explained within these catchments. This highlights the complexity of soil-plant-microbe interactions and the difficulty in identifying drivers of grain yield within different environments. However, the fact that biological attributes had a greater quantifiable influence than chemical or physical attributes on yield variability in this case study provides justification to the development of agricultural farming systems that encourage soil biological fertility (Abbott, Murphy 2003). However, there are few, if any, quick fix solutions to improving soil biological fertility. Research trial data from WA (Table 2) demonstrates that it can take many years for differences in attributes of soil biological fertility to occur upon implementation of management practices. Soil biological attributes are generally highly variable spatially over small distances (see Case study 4, Table 2), with changes in the chemical and physical attributes of the soil often having a greater influence than imposed agronomic management practices on soil biological fertility. Therefore, it is difficult to measure

significant differences between treatments even when changes seem quite large (eg Case study 3 microbial biomass, Table 2).

Table 2. Impact of agronomic management practices on microbial biomass, biological soil nitrogen supply (PMN) and diversity (catabolic diversity, range possible 0-24 with higher number indicating more diverse population) of microorganisms in four trials from WA that represent the major soil groups (Chromosols, Sodosols, Kandosols). Values with the same letter are not significantly different (P<0.10) within the same trial for the biological attribute specified.

Case study #	Agronomic management	Microbial biomass	PMN	Catabolic diversity
		kg C/ha	kg N/ha	min = 1 max = 24
1	Harvest stubble burnt	98a	No data	14.5a
	Harvest stubble retained	153b	No data	15.5b
2	Continuous wheat rotation	308a	30a	15.8a
	Faba beans:Wheat:Canola:Wheat	317a	30a	16.4a
	Medic (grazed) : Wheat	421b	25a	18.0b
	Annual pasture - Ryegrass (grazed)	417b	45ab	16.5a
	Perennial pasture - Lucerne (grazed)	421b	67b	16.5a
3	Lupin - brown manure	140a	13a	15.9a
	Oat - brown manure	76a	14a	17.6b
	Mustard - brown manure	119a	15a	19.4c
4	Variability within 10 ha; n = 220 pts	22 to 1000	4 to 32	No data

1: Data collected after 17 years of imposed treatments, 0-5 cm, Chromosol, Merredin WA.
2. Data collect after 4 years of imposed treatments, 0-5 cm, Sodosol, Mindarabin WA.
3. Data collected after 2 months of imposed treatments, 0-10 cm, Kandosol, Meckering WA.
4. Minimum and maximum data from 220 composite bags of soil collected under a barley crop on a 25 m grid over 10 ha; 0-10 cm; Dangin WA.

Seasonal variability in the data collected is also a major issue in deciding when to sample soil for biological attributes. This is illustrated in Table 3 where it can be seen that the seasonal (sowing, tillering, flowering, harvest) differences in measured soil biological attributes are considerable.

Table 3. Impact of season on the microbial biomass, potentially mineralisable nitrogen (PMN) and the actual daily rate of inorganic nitrogen release through microbial decomposition of soil organic matter and residues (gross nitrogen mineralisation). Six conventional farms were paired with two farms of each of the other farming systems listed. S = sowing, T = tillering, F = flowering, H = harvest.

Farming system	Microbial biomass-N (kg N/ha)					PMN (kg N/ha)					Gross mineralisation (kg N/ha/day)			
	S	T	F	H		S	T	F	H		S	T	F	H
Conventional	52	42	20	12		42	34	40	36		7.1	6.6	1.2	1.0
Integrated	60	32	17	11		58	44	54	47		5.8	6.1	1.2	0.8
Organic	72	46	19	10		54	44	53	48		3.6	6.4	1.6	1.0
Bio-dynamic	72	37	26	11		54	46	58	54		5.3	5.5	1.3	1.1

However, it should be noted that PMN was more stable through the season than measurement of microbial biomass or microbial activity (gross nitrogen mineralisation), suggesting that it is an easier soil biological attribute to interpret between and within seasons. Data in Table 3 also illustrates that seasonal changes in the measured biological attributes are greater than measured differences between farming system type. Thus the capacity to alter soil biological fertility within a region is primarily constrained by water and temperature with agronomic practice as a secondary factor.

Conclusion

Soil biological fertility was significantly correlated to grain production in WA. The benefit was predominately associated with the size of the microbial biomass, which was directly related to their capacity to decompose soil organic matter and fresh residues to release plant available nitrogen. These findings confirm our view that WA farming systems are highly reliant on biological nitrogen supply and that farming systems need to be modified where possible to fully benefit from water availability and microbial nutrient supply. However to achieve this, limitations associated with both the asynchrony and synlocation of water and nutrients need to be further addressed. This will require improved soil management to identify and remove soil constraints to plant growth and rooting depth, new plant breeding to improve plant root architecture in order to capture water and nutrients, a flexible fertiliser strategy (type, split applications, delivery), developing an economic role for deep rooted plants and improved plant residue management (carbon:nitrogen ratio of decomposing material, level and timing of incorporation) and identifying novel methods for manipulating microbial processes.

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References

Abbott LK and Murphy DV Eds 2003. Soil biological fertility: A key to sustainable land use in agriculture. Kluwer Academic Publishers, Dordrecht.

Angus JF 2001. Nitrogen supply and demand in Australian agriculture. Australian Journal of Experimental Agriculture 41 277-288.

Hoyle FC and Anderson WK 1993. Yield performance and variability of winter cereals in Western Australia 1960-1990. Technical Report No. 61, Department of Agriculture Western Australia (ISSN 1037-9789).

Murphy DV, Fillery IRP and Sparling GP 1998. Seasonal fluctuations in gross N mineralisation, ammonium consumption and microbial biomass in a Western Australian soil under different land uses. Australian Journal of Soil Research 49 523-535.

Murphy DV, Stockdale EA, Hoyle FC, Smith JU, Fillery IRP, Milton N, Cookson WR, Brussaard L and Jones DL 2004. Matching supply with demand: Principle, practice and prediction. In Controlling nitrogen flows and losses. Eds Hatch DJ, Chadwick DR, Jarvis SC, Roker JA. Wageningen Academic Publishers, in press.

Ridley AM, Mele PM and Beverly CR 2004. Legume-based farming in Southern Australia: Developing sustainable systems to meet environmental challenges. Soil Biology & Biochemistry, in press.

Contact details

Dr Daniel Murphy
Research Fellow
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia Crawley, WA 6009
Phone 08 6488 7083
Fax 08 6488 1050
Email dmurphy@agric.uwa.edu.au
Websites www.clr.uwa.edu.au, www.soilhealth.com

Impact of fertilisers on soil biota

Else K. Bünemann and Annie McNeill
University of Adelaide

Introduction

Fertilisers

Fertilisers are an integral component of agricultural production systems and are applied with the main goal of maximising yields and economic returns. An underlying principle governing fertiliser application rate should be the replacement of nutrients removed in the harvested product. Fertilisers include mineral products such as urea, ammonium nitrate, sulfates and phosphates, and organic fertilisers such as animal manures and biosolids. Since most organic fertilisers are waste products, their application rate is often determined by availability rather than demand. Currently, the use of mineral fertilisers in broadacre Australian agriculture outweighs the use of organic fertilisers.

Soil biota

While the application of fertilisers usually aims at meeting plant demands, it can also have an impact on soil biota. Soil biota consist of the microflora (bacteria and fungi) and the soil fauna (protozoa and invertebrate groups such as nematodes, mites and earthworms). They influence the availability of nutrients for crop production via a range of activities such as the decomposition of crop residues, immobilisation (microbial uptake) of nutrients, mineralisation (transformation of organic nutrients into plant available inorganic forms), biological nitrogen fixation, and bioturbation. The soil fauna are crucial for the initial comminution of residues and mixing into the soil, while the microflora have a greater suite of enzymes for chemical breakdown of organic material (Paul, Clark 1996). Bacteria and fungi are often considered as a labile pool of nutrients (carbon, nitrogen, phosphorus and sulfur) called the soil microbial biomass that has a pivotal role in nutrient immobilisation and mineralisation. The release of nutrients from the microbial biomass is partly regulated through grazing by the soil fauna.

Methods to determine fertiliser effect on soil biota

The effect of fertilisers on soil biota can be measured either as changes in the amount of single organisms, organism groups or methodologically defined pools such as the microbial biomass, or as changes in biological activity such as soil respiration, enzyme activities and root colonisation rates. Variable effects of fertilisers on different organisms may change the composition of the microbial (or faunal) community without changing total amounts or activities. However, the majority of studies have focussed on the soil microbial biomass as the central pool in nutrient cycling.

Approach of this review

In this paper we summarise the current understanding of fertiliser effects on soil biota, based on the concept that fertilisers can affect soil biota through direct or indirect effects (Table 1). Direct effects via changes in nutrient availability are likely to become obvious in the first season after their application, or in the longer term if repeated additions are required to reach a threshold above which effects are seen. Indirect effects will usually take more than one season to establish, especially when changes in soil organic matter levels are involved. In the case of long-term data, it can be difficult to separate direct and indirect effects. The evidence from Australia is rather limited, and therefore the review includes literature from overseas, in an attempt to establish the

main principles of fertiliser effects on soil biota and to draw some conclusions applicable to agro-ecosystems in Australia.

Table 1: Potential effects of fertilisers on soil biota

Direct effects	Time frame
Increased amount and/or activity after removal of nutrient limitations	Short[1]- to long-term[2]
Decreased activity due to high nutrient availability	
Decreased amount and/or activity due to toxicity	
Indirect effects	
Change in pH	Long-term
Change in productivity, residue inputs and soil organic matter levels	

1 One season
[2] More than one season

Direct effects of mineral fertilisation

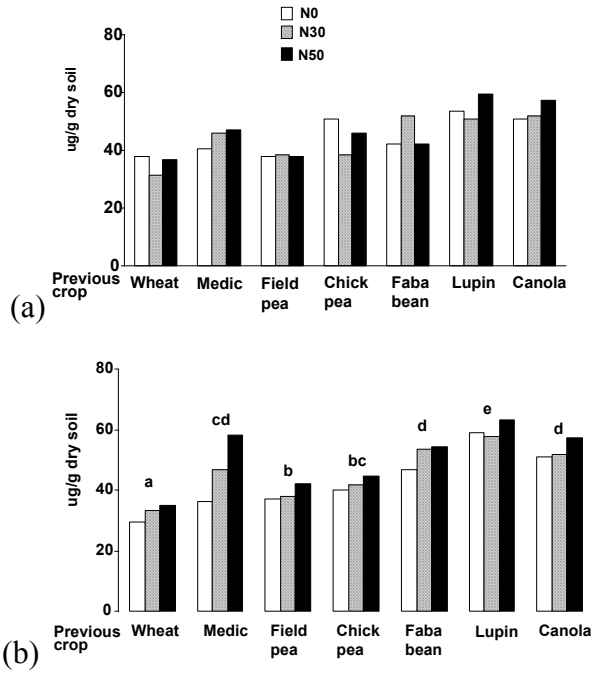
Short-term stimulation of soil biota due to mineral nitrogen or phosphorus fertilisers

Increased amounts or activity of soil biota as a direct result of mineral fertilisation have been reported in several studies. For example, Sarathchandra et al (2001) observed a short-term increase in gram-negative bacteria and rock-phosphate-dissolving bacteria two weeks after application of superphosphate or rock phosphate to a permanent pasture in New Zealand. Application of ammonium nitrate to pasture in the UK led to a short-term increase in the microbial processes of nitrification and ammonification, although the size of microbial biomass (carbon and nitrogen) did not change (Lovell, Hatch 1997). Indeed, a 'priming effect' of nitrogen addition on soil respiration is often observed (Kuzyakov et al 2000), and there is recent evidence that nitrogen addition might specifically stimulate the decomposition of older, stabilised soil organic matter (Waldrop, Firestone 2004).

No short-term effect or depression of soil biota due to mineral nitrogen fertilisers

While such findings suggest a positive impact of mineral fertilisers on soil biota, probably via the removal of nutrient limitations, many more studies report no change or even a decrease in amounts or activity of soil biota following mineral fertiliser application. For example, a study in Western Australia (McNeill et al 2000) found little influence of fertiliser nitrogen, applied to wheat at sowing, on microbial nitrogen measured at tillering or anthesis (Figure 1a,b), compared with the effect of the previous crop on the microbial biomass (Figure 1b). Likewise, the addition of 120 kg N/ha as ammonium nitrate to a potato crop in the UK had no effect on soil respiration, dehydrogenase activity or microbial carbon and nitrogen throughout the growing season (Ritz et al 1992), although the addition of sucrose (as an energy/carbon source) in conjunction with nitrogen did increase all of these parameters. Nitrogen fertilisation did not affect the size and activity of the microbial biomass under permanent pasture, regardless of whether or not the soil had received nitrogen fertilisation for the previous 15 years (Hatch et al 2000). Similarly, addition of ammonium sulfate to soils under pasture in NZ had little effect on microbial use of added energy (glucose), although it appeared to actually decrease amounts of microbial phosphorus (Saggar et al 2000).

Figure 1: Microbial N (ug/g) in 0-15 cm soil depth (Goomalling WA 1998) under a wheat crop after application of urea (0, 30 or 50 kg N/ha at sowing) and following different break crops (wheat, medic, field pea, chickpea, faba bean, lupin or canola): (a) at tillering in August and (b) at peak biomass of the crop in October. Different letters atop bars indicate significant differences between previous crop across N rates (p<0.001).



No short-term effect or depression of soil biota due to mineral phosphorus or sulfur fertilisers

There is even less information available on the direct effects of mineral phosphorus and sulfur fertilisers on soil biota than there is for nitrogen fertilisers. A Canadian study did not find any immediate nor residual effect of sulfur fertilisers on the microbial biomass under a wheat-canola rotation, but observed a slight decrease in diversity as assessed by the utilisation of carbon substrates (Lupwayi et al 2001). A reduction in specific organisms such as arbuscular mycorrhizal (AM) fungi by phosphorus fertilisation appears to be fairly well established. In a comparison of Australian pastures under conventional and biodynamic management, Ryan et al (2000) noted a negative relationship between available phosphorus and colonisation rates of clover roots with AM fungi. However, AM colonisation rates of ryegrass were not affected by phosphorus addition (Ryan, Ash 1999). These findings agree with the variable effect of nitrogen, phosphorus, potassium (NPK) fertilisation on percent root colonisation by AM in different grassland species observed by Rillig et al (1998), suggesting that in the case of mycorrhizal symbioses, indirect effects through changes in plant growth and metabolism rather than direct effects on the fungi might prevail.

Short-term toxicity effects of mineral fertilisers

A decreased amount or activity of soil biota after mineral fertilisation could be due to the toxicity of metal contaminants contained in mineral fertilisers. In general, nitrogen and potassium fertilisers contain very low levels of contaminants, whereas phosphorus fertilisers often contain significant amounts of cadmium, mercury and lead (McLaughlin et al 2000). Metal contaminants are most prevalent in waste products from urban and industrial areas. Long-term chronic toxicity due to gradually accumulating metals appears to be far more common than immediate, acute toxicity (Giller et al 1998).

Therefore, toxicity effects on soil organisms will be discussed in the section on long-term effects.

Direct effects of organic fertilisation

Short-term stimulation of soil biota due to organic fertilisers

The majority of studies report a positive direct effect of organic fertilisers on soil biota. For example, the application of farmyard or poultry manure to barley in the UK increased microbial nitrogen immediately and microbial carbon in the second year of the trial (Ritz et al 1997). Microbial carbon and gross nitrogen immobilisation were also increased under dung pats applied to pasture in the UK, which was attributed to a temporary increase in available soil carbon (Hatch et al 2000). In another study, the microbial biomass as well as soil respiration clearly increased after mixing of dung into the soil, but not under dung pads in the field (Lovell, Hatch 1997). Likewise, no change in microbial phosphorus under dung pads was observed in Victoria, except for an increase at the last sampling after 60 days (Aarons et al 2004).

Role of carbon inputs with organic fertilisers

The relatively greater influence of organic versus inorganic fertilisers was emphasised in a study where three months after application of 200 kg N/ha as ammonium nitrate to maize, soil respiration, acid phosphatase and dehydrogenase activity were higher than in the non-fertilised control. However, all these changes were even more pronounced when the same amount of nitrogen was added as dairy manure or composted sewage sludge (Marinari et al 2000). These differences are certainly related to carbon inputs with organic fertilisers. In contrast to plants, soil organisms are heterotrophic and need carbon as the most essential nutrient. A large fraction of soil organic carbon is poorly available to soil organisms. Additions of labile carbon have therefore also been observed to induce 'priming effects' on the decomposition of soil organic matter (Kuzyakov et al 2000).

Indirect effects of mineral and organic fertilisers

Absence of long-term effects of mineral phosphorus fertilisers

Preliminary data from two trials in Australia indicate the absence of phosphorus fertiliser effects on microbial phosphorus in spite of significantly increased phosphorus availability after many years of phosphorus addition (Table 2). This agrees with findings from pastures in NZ, where no significant long-term effects of phosphorus additions on microbial phosphorus or earthworm abundance were noted (Sarathchandra et al 1993). The same was observed during six years of phosphorus fertiliser inputs to pastures in Victoria (Aarons 2001).

Change in pH – a long-term effect of fertilisation

While phosphorus fertilisation often does not have any effect on soil biota, mineral nitrogen fertilisation sometimes has a negative effect. For example, repeated additions of ammonium nitrate to various wheat rotations in South Australia decreased microbial carbon as well as pH (Ladd et al 1994). In a long-term trial in Sweden, Witter et al (1993) identified a change in soil pH due to repeated addition of ammonium sulfate as an indirect effect of fertilisation on soil biota. Urea application also decreased soil organic carbon, microbial carbon and pH under permanent pasture, while superphosphate applications affected none of these parameters (Sarathchandra et al 2001). While a decrease in pH appears to be the primary explanation for negative effects of mineral fertilisers on soil biota, it does not always correspond with a decrease in microbial biomass (Belay et al 2002, Moore et al 2000, Peacock et al 2001).

Table 2: Effect of mineral phosphorus fertilisation on available and microbial phosphorus (Bünemann et al, unpublished)

Site	Land use	P inputs	Available P[1]	Microbial P[2]	Organic C[3]
		kg/ha/yr	mg P/kg soil	mg P/kg soil	g/kg
Walpeup, Vic	Wheat-fallow rotation[4]	0	4.4 ± 0.2	1.0 ± 0.1	0.63 ± 0.07
		135	18.0 ± 0.8	1.5 ± 0.8	0.60 ± 0.07
Otterbourne, NSW	Permanent pasture	0	1.4 ± 0.1	3.9 ± 0.2	1.99 ± 0.11
		125	4.0 ± 0.1	4.4 ± 0.7	2.18 ± 0.08
		250	9.1 ± 0.1	3.0 ± 0.9	2.17 ± 0.02

1 P extractable with anion-exchange resin membranes
2 P-flush after hexanol fumigation
3 Organic C predicted by mid-infrared spectroscopy (MIR)
4 Permanent phosphate trial (McClelland 1968)

Change in soil organic matter - a long-term effect of fertilisation

Graham et al (2002) investigated the amounts of microbial carbon and nitrogen under sugarcane after 59 years of differential crop residue management and NPK fertilisation, showing that the microbial biomass was directly influenced by residue management and indirectly by NPK fertilisation through increased residue inputs. In fact, many long-term experiments with different forms and rates of fertilisation have rendered strikingly good relationships between the size of the microbial biomass and soil organic matter contents (Houot, Chaussod 1995, Leita et al 1999, Moore et al 2000, Witter et al 1993). Usually, the treatments with mineral fertilisation are at the lower end of the correlation, as they increase soil organic and thus microbial carbon to a lesser degree than organic amendments such as farmyard manure and compost (Leita et al 1999). The example provided by Houot and Chaussod (1995) is especially interesting as it shows how the excellent correlation found after more than one hundred years of constant management practices remained disturbed two years after a change in crop rotation and crop residue management. The time required to reach a new equilibrium is a factor that may confound the results from many short-term studies.

Nevertheless, most studies that show a positive effect of mineral or organic fertilisation on soil organisms also record an increase in soil organic matter. Indirect effects of fertilisers on soil biota through an increase in plant productivity and returned crop residues may also explain results from the Mallee region in South Australia. Gupta et al (2004) showed that after four years of a low input system with fertiliser rates of 10 kg P/ha and 5 kg N/ha, microbial biomass size and activity were lower than for a high input system with rates of 15 kg P/ha, 27 kg N/ha and 1.5% zinc (Table 3). On the other hand, the absence of phosphorus fertiliser effects on microbial phosphorus shown in Table 2 may be due to the fact that no significant changes in soil organic carbon content were observed.

Long-term toxicity effects of fertilisers

The accumulation of toxic metals in soil due to repeated fertiliser applications is of concern in the long-term. It has been shown that specific organisms such as nitrogen fixing rhizobia are far more sensitive to metal toxicity than clover as their host plant. This resulted in nitrogen deficiency of clover due to ineffective rhizobia in sludge-amended soils (Giller et al 1998). Such observations warrant strict regulations of fertiliser quality and applied quantity, especially of waste products such as sewage

sludge and biosolids, in order to minimise contamination of agricultural land with toxic metals.

Table 3: Microbial biomass carbon (MB-C), nitrogen (MB-N), phosphorus (MB-P), and respiration as a measure of microbial activity in the surface soils (0-10 cm) of selected treatments at the Mallee Sustainable Farming Systems Waikerie core site after 4 years. DP is district practice and HI is high input as referred to in the text.

Cropping system	MB-C(kg C/ha)	MB-N (kg N/ha)	MB-P (kg P/ha)	Respiration (g CO2 /m2/hour)
Pasture-Wheat (DP)	265	26	16.0	0.105
Pasture-Wheat (HI)	370	43	21.0	0.185
Legume-Wheat (HI)	370	46	13.0	0.210
Canola-Wheat (HI)	357	36	16.5	0.175

From Gupta et al 2004.

Conclusions

The availability of carbon substrates is more important for soil biota than that of nutrients such as nitrogen, phosphorus, potassium and sulphur. Therefore, organic fertilisers usually have greater impact on soil biota than mineral fertilisers. Direct effects of mineral fertilisers on soil biota seem to be variable but perhaps less important than indirect effects. The main indirect effects are a depression of soil biota due to a decrease in soil pH, and an increase in biological activity with increasing plant productivity, crop residue inputs and soil organic matter levels. As Australian soils are generally low in organic matter and nutrient contents, any increase in soil organic matter is desirable in view of the important role of soil biota in nutrient cycling.

References

Aarons SR 2001. Cycling of organic phosphorus in grazed dairy systems. Final report July 2001. Agriculture Victoria, Ellinbank Victoria.

Aarons SR, Hosseini HM, Dorling L, Gourley CJP 2004. Dung decomposition in temperate dairy pastures: II. Contribution to plant-available soil phosphorus. Australian Journal of Soil Research 42 115-123.

Belay A, Claassens AA, Wehner FC 2002. Effect of direct nitrogen and potassium and residual phosphorus fertilisers on soil chemical properties, microbial components and maize yield under long-term crop rotation. Biology & Fertility of Soils
35 420-427.

Giller KE, Witter E, McGrath SP 1998 Toxicity of heavy metals to microorganisms and microbial processes in agricultural soils: a review. Soil Biology & Biochemistry 30 1389-1414.

Graham MH, Haynes RJ, Meyer JH 2002. Soil organic matter content and quality: effects of fertiliser applications, burning and trash retention on a long-term sugarcane experiment in South Africa. Soil Biology & Biochemistry 34 93-102.

Gupta VVSR, Roget DK, Drew E 2004. Beneficial functions of soil biota in dryland agricultural systems. In SANTFA workshop Tanunda, February 2004.

Hatch DJ, Lovell RD, Antil RS, Jarvis SC 2000. Nitrogen mineralisation and microbial activity in permanent pastures amended with nitrogen fertiliser or dung. Biology & Fertility of Soils 30 288-293.

Houot S, Chaussod R 1995. Impact of agricultural practices on the size and activity of the microbial biomass in a long-term field experiment. Biology & Fertility of Soils
19 309-316.

Kuzyakov Y, Friedel JK, Stahr K 2000. Review of mechanisms and quantification of priming effects. Soil Biology & Biochemistry 32 1485-1498.

Ladd JN, Amato M, Zhou LK, Schultz JE 1994. Differential effects of rotation, plant residue and nitrogen fertiliser on microbial biomass and organic matter in an Australian Alfisol. Soil Biology & Biochemistry 26 821-831.

Leita L, De Nobili M, Mondini C, Muhlbachova G, Marchiol L, Bragato G, Contin M 1999. Influence of inorganic and organic fertilisation on soil microbial biomass, metabolic quotient and heavy metal bioavailability. Biology & Fertility of Soils 28 371-376.

Lovell RD, Hatch DJ 1997. Stimulation of microbial activity following spring applications of nitrogen. Biology & Fertility of Soils 26 28-30.

Lupwayi NZ, Monreal MA, Clayton GW, Grant CA, Johnston AM, Rice WA 2001. Soil microbial biomass and diversity respond to tillage and sulfur fertilisers. Canadian Journal of Soil Science 81 577-589.

Marinari S, Masciandaro G, Ceccanti B, Grego S 2000. Influence of organic and mineral fertilisers on soil biological and physical properties. Bioresource Technology
72 9-17.

McClelland VF 1968. Superphosphate on wheat: the cumulative effect repeated applications on yield response. Australian Journal of Agricultural Research 19 1-8.

McLaughlin MJ, Hamon RE, McLaren RG, Speir TW, Rogers SL 2000. Review: A bioavailability-based rationale for controlling metal and metalloid contamination of agricultural land in Australia and New Zealand. Australian Journal of Soil Research 38 1037-1086.

Moore JM, Klose S, Tabatabai MA 2000. Soil microbial biomass carbon and nitrogen as affected by cropping systems. Biology & Fertility of Soils 31 200-210.

Paul EA, Clark FE 1996. Soil microbiology and biochemistry. Academic Press: San Diego.

Peacock AD, Mullen MD, Ringelberg DB, Tyler DD, Hedrick DB, Gale PM, White DC 2001. Soil microbial community responses to dairy manure or ammonium nitrate applications. Soil Biology & Biochemistry 33 1011-1019.

Rillig MC, Allen MF, Klironomos JN, Chiariello NR, Field CB 1998. Plant species-specific changes in root-inhabiting fungi in a California annual grassland: Responses to elevated CO_2 and nutrients. Oecologia 113 252-259.

Ritz K, Griffiths BS, Wheatley RE 1992. Soil microbial biomass and activity under a potato crop fertilised with N with and without C. Biology & Fertility of Soils 12 265-271.

Ritz K, Wheatley RE, Griffiths BS 1997. Effects of animal manure application and crop plants upon size and activity of soil microbial biomass under organically grown spring barley. Biology & Fertility of Soils 24 372-377.

Ryan M, Ash J 1999. Effects of phosphorus and nitrogen on growth of pasture plants and VAM fungi in SE Australian soils with contrasting fertiliser histories (conventional and biodynamic). Agriculture Ecosystems & Environment 73 51-62.

Ryan MH, Small DR, Ash JE 2000. Phosphorus controls the level of colonisation by arbuscular mycorrhizal fungi in conventional and biodynamic irrigated dairy pastures. Australian Journal of Experimental Agriculture 40 663-670.

Saggar S, Hedley CB, Giddens KM, Salt GJ 2000. Influence of soil phosphorus status and nitrogen addition on carbon mineralisation from 14C-labelled glucose in pasture soils. Biology & Fertility of Soils 32 209-216.

Sarathchandra SU, Ghani A, Yeates GW, Burch G, Cox NR 2001. Effect of nitrogen and phosphate fertilisers on microbial and nematode diversity in pasture soils. Soil Biology & Biochemistry 33 953-964.

Sarathchandra SU, Lee A, Perrott KW, Rajan SSS, Oliver EHA, Gravett IM 1993. Effects of phosphate fertiliser applications on microorganisms in pastoral soil. Australian Journal of Soil Research 31 299-309.

Waldrop MP, Firestone MK 2004. Altered utilisation patterns of young and old soil C by microorganisms caused by temperature shifts and N additions. Biogeochemistry 67 235-248.

Witter E, Mårtensson AM, Garcia FV 1993. Size of the soil microbial biomass in a long-term field experiment as affected by different N-fertilisers and organic manures. Soil Biology & Biochemistry 25 659-669.

Contact details

Dr Else K. Bünemann
Research Fellow
School of Earth and Environmental Sciences
Prescott Building DP 636
The University of Adelaide, Waite Campus
Glen Osmond SA 5064
Phone 08 8303 7306
Fax 08 8303 6511
Email else.bunemann@adelaide.edu.au

Impacts of pesticides on soil biota

Lukas Van Zwieten
NSW Department of Primary Industries, Wollongbar

Introduction

There is significant uncertainty in the prediction of impacts of external stressors such as pesticide application on landscape effects. For example, a pesticide may be toxic to a soil organism under laboratory studies, but does field application result in long-term population changes, and if so what is the significance of these population changes? If an applied pesticide removes a particular species from a site, and the species returns to the site from neighbouring populations, has there been any harm to soil health?

Recent developments in aquatic ecotoxicology have combined ecotoxicological principles (eg direct toxicity measurements to individual species), with ecological knowledge to enhance the realism of risk assessment (Liess 2004). In other words, an understanding of the entire system is required to gain an accurate picture of potential impacts. Many studies often evaluate short-term effects on individual indicators, so cannot realistically predict the true risk of a chemical or management practice on soil biota.

The use of pesticides has long been a feature of conventional agricultural practice and their use has made it possible to increase crop yields and food production (Lee 1985). However, many of these chemicals have toxic effects that are not confined to their target species, and their application may have impacts on organisms that benefit the wider agroecosystem. The paradigm of Newton's Third Law, although referring to motion and energy, is quite applicable to the management of soils: ‘To every action there is an equal and opposite reaction.’ Every management decision a farmer implements will impact on the health of the soil, resulting in either a beneficial or detrimental outcome.

This paper discusses the potential impacts of several pesticides on soil biota and highlights implications for the management of these soils.

What are soil biota and how can impacts be measured?

There are many excellent reviews on soil biota (see Pankhurst 1997, Rovira 1994) and their interactions in farming systems. Soils contain microorganisms including bacteria, fungi, yeasts; photosynthetic organisms including algae, and macroorganisms such as protozoa, nematodes, mites, springtails, spiders, insects and earthworms. The functions of this complex array of biota, often referred to as the ‘soil food web,’ are diverse, and include residue decomposition, nutrient storage and release, soil structure and stability, resistance against disease and degradation or immobilisation of pesticides and other pollutants. In the literature, soil biota are measured or studied in many different ways, including direct observation under microscopes and other direct counting mechanisms, analysis of intra- and extracellular enzymes, respiration and biomass carbon, fatty acid profiles, and DNA fingerprinting. Unlike the field of aquatic ecotoxicology where many standard tests are applied to measure impacts of stressors, there are no standard reporting mechanisms for measuring soil biota. This renders the direct comparison between different pesticides and their potential impacts impossible.

What processes can affect soil biota?

Many factors can have negative impacts upon soil health. These factors include loss of organic carbon (Islam and Weil 2000), compaction (Singleton and Addison 1999), disruption of soil macroaggregates (Islam and Weil 2000), pesticides (Mitra and Raghu 1998) and pesticide breakdown products (Cernakova and Zemanovicova 1998), inorganic pollution arising through fertilisers, fungicides and sludge application (Merry et al 1986, Gong et al 1997), the use of fertilisers (Stamatiadis et al 1999) and non-pesticide organic pollution including surfactants (Wilke 1997). Other causes of reduced soil health can arise through water and wind erosion (Garcia et al 1997), grazing (Anon 2001), loss of organic matter due to fire, deforestation and tillage (Islam and Weil 2000).

How can pesticides influence soil biota?

Pesticides can impact on soils through direct or indirect mechanisms. For example, the use of herbicide may not directly influence soil biota, but the process of removing vegetative material from the soil can influence availability of rhizosphere exudates and organic material to biota, and can reduce soil stability, leading to erosion and compaction. Direct impacts of pesticides can occur when the chemical reaches the soil, either due to direct targeted deposition such as the case of soil fumigants, or pre-emergent herbicides, or through indirect deposition from spray and spray drift, dripping from plant material, and contaminated plant material falling to the soil.

There are several factors influencing the amount of pesticide reaching the soil, and potentially impacting on soil biota. During application, pesticide may degrade in the spray mix and/or undergo photodegradation (ie sunlight). The pesticide may drift off target and affect non-target sites. These processes reduce the amount of active chemical impinging on the soil surface.

On the soil, pesticides dissipate though several processes, including volatilisation, leaching and surface run-off. Degradation can occur through biological and non-biological processes. The biological availability of the pesticide may be reduced by its binding to components in soil, in particular, organic matter and clay.

Effects of selected pesticides on soil biota

Table 1 summarises the effects of a few selected pesticides on soil biota. It can be observed that negative impacts range from negligible to very significant, while some pesticides even appear beneficial to soil biota!

Although the list of chemicals mentioned is not exhaustive, it covers a range of herbicides, fungicides, insecticides, veterinary care products and soil fumigants. Often data is not available on the effects of pesticides on soil biota, as registration requirements do not require extensive soil biota tests to be undertaken.

Table 1. Impacts of selected pesticides on soil biota

Active chemical	Effects	Reference
Herbicides		
glyphosate	Bacterial populations reduced. Fungi and actinomycetes increase, with 9-19% increase in microbial activity. Increased rate of glyphosate degradation over time.	Araujo et al 2003
glyphosate	Short-term changes to community structure. Increased microbial activity and no long-term changes to community structure	Busse et al 2001
glyphosate and paraquat	Activation of urease and invertase soil enzymes, but suppression of phosphatase enzyme	Sannino and Gianfreda 2001
pendimethalin	Soil nematodes and other invertebrates reduced, plant-rhizobium symbiosis affected.	Strandberg and Scott-Fordsmand 2004
atrazine and metolachlor	Altered community structure of several groups of bacteria and actinomycetes.	Seghers et al 2003
atrazine	Significant activation of soil urease activity, and suppression of invertase enzyme.	Sannino and Gianfreda 2001
butachlor	Significant effects on earthworms and soil health	Panda and Sahu 2004
isoproturon	Affects earthworms at very high soil concentrations (not agricultural rates) with LC50 for *Eisenia fetida* >1000mg/kg	Mosleh et al 2003
oxyfluorfen	Stimulates microbial populations, and increases availability of phosphorus in rhizosphere soil.	Das et al 2003
Insecticides/ nematicides		
chlorpyrifos	Reduced bacterial numbers, but significantly increased fungal numbers.	Pandey and Singh 2004
carbofuran	Significant impacts on acetylcholinesterase activity in earthworms.	Panda and Sahu 2004
dimethoate	Short-term reduction in microarthropod numbers (Collembola), but recovery in numbers after time.	Martikainen et al 1998
malathion	Short-term impacts on earthworm population.	Panda and Sahu 1999
DDT and arsenic contamination	Changes in microbial properties.	Edvantoro et al 2003
DDT	Reduced bacterial and soil algal populations, but may have increased fungal counts.	Megharaj et al 2000
arsenic	Reduced performance of soil functions resulting in reduction of DDT degradation.	Van Zwieten et al 2003
Fungicides		
copper	Earthworm populations avoid soils with concentrations as low as 34mg/kg. Lack of breakdown of organic	Van Zwieten et al 2004

	carbon. Potential long-term implications reported.	
copper	Increased respiration indicating microbial stress. Significantly reduced microbial biomass.	Merrington et al 2002
copper	Reduced performance of soil functions resulting in reduction of DDT degradation.	Gaw et al 2003
metalaxyl	Reduced enzyme activity, in particular dehydrogenase. Toxic to nitrogen fixers.	Monkiedje et al 2002
benomyl	Suppression of respiration, stimulation of dehydrogenase, effects were less noticeable with organic matter addition	Chen et al 2001
benomyl	Significant long-term impacts on mycorrhizal colonization (80% reduction), reduction in fungal to bacterial ratios and nematode numbers.	Smith et al 2000
captan	Suppression of respiration and dehydrogenase, but increases in ammonium nitrogen.	Chen et al 2001
chlorothalonil	Suppression of respiration, stimulation of dehydrogenase.	Chen et al 2001
Antimicrobials		
tylosin, oxytetracycline, sulfachloropyridazine	Tylosin and sulfachloropyridazine significantly impact on gram positive bacteria, while oxytetracycline inhibits general microbial respiration a levels as low a 1mg/kg in soil.	Vaclavik et al 2004
tylosin	Long-term changes to microbial community structure, and short-term reduction in total microbial numbers.	Westergaard et al 2001
Fumigants		
propargyl bromide and 1,3dichloropropen	Significant effects on respiration and dehydrogenase activity.	Dungan et al 2003

Few studies demonstrate long-term impacts of the pesticide application, and even less discuss measured or observed changes to soil processes. Very significant effects were demonstrated with copper-based fungicides, where Van Zwieten et al (2004) demonstrated reductions in earthworm populations in soil. The authors also mentioned a buildup of organic material on the soil surface, most likely as a result of the lack of bioturbation (mixing of the soil by biota). Likewise, Gaw et al (2004) described the lack of pesticide residue breakdown in soils where copper residues were co-contaminating. Merrington et al (2002) further demonstrated significant impacts on soil microbial processes such as respiration and biomass carbon, and showed conclusively that copper residues resulted in stressed microbes. These impacts are unlikely to change in the near future, as copper accumulates in surface soils and is not prone to dissipative mechanisms such as biodegradation. Also having very significant effects are the soil fumigants, whose purpose in soil is to eliminate biology and any competition for soil resources by the crop. The long-term effects of fumigants however was reduced by the addition of commercially available composted steer manure, where normal biological activity was observed 8-12 weeks following application of high rates of the fumigant (Dungan et a 2003). In the absence of the organic amendment, little recuperation of soil function was detected even after 12 weeks.

Impacts of biological pesticides on soil biota

Microorganisms have been knowingly used to control plant diseases for over 100 years (Winding et al 2004). However, risks of using biological control agents are often forgotten. While the microbes selected may naturally occur in the environment, there are concerns that altering the proportion of these microbes will result in environmental impacts on non-target species including mycorrhizal and saprophytic fungi, soil bacteria, plants, insects, aquatic and terrestrial animals and humans (Brimner and Boland 2003). The authors argue dry-spored biocontrol agents could potentially become a problem (eg allergen to humans) as these spores are more suited to air transport than wet-based spores and therefore more likely to be spread widely.

Recently, there has been evidence that significant non-target effects may occur with bacterial biocontrol agents (Winding et al 2004), however effects were generally observed as short-term and they did not impact on soil health.

Implications for farm management

There is clear evidence that soil biota are impacted by pesticides. Some impacts include the short-term stimulation of enzymatic activity and bacterial numbers, while other impacts include the long-term elimination of earthworm populations. While not an exhaustive list, some important issues that need to be considered are outlined below.

Nutrient management

Altering microbial populations in soil can affect the availability of nutrients. For example, Taiwo and Oso (1997) report the reduction in phosphorus availability to plants following application of pesticides including pyrethrin, atrazine and metolachlor. Oxyfluorfen has been shown to increase phosphorus availability in rhizosphere soil (Das et al 2003).

Arbuscular mycorrhizal associations

Fungicides such as benomyl have been shown to have very significant effects on AM fungi (Smith et al 2000), possibly altering the uptake of nutrients by the plant, and affecting natural disease control mechanisms.

Breakdown of organic material

Residues of copper (Van Zwieten et al 2004) and other pesticides such as malathion (Panda and Sahu 1999) reduce earthworm populations, thus reducing bioturbation and breakdown and incorporation of organic material in soil.

Breakdown of soil contaminants

Metal based pesticides, in particular copper (Gaw et al 2004) and arsenic (Van Zwieten et al 2003) have been shown to increase the persistence of recalcitrant contaminants such as DDT.

While uncertainty still exists on the implications on many of the effects described in the literature, what can Australian farmers realise from the current state of knowledge? If we think about the paradigm of Newton's Third Law 'To every action there is an equal and opposite reaction' it is almost certain that the application of a pesticide will have some impact on soil biota. Whether this impact is significant for the future health of the agro-ecosystem is still unknown in most cases. The achievement of sustainable agriculture was 'let down' in the 20th century when research focused strongly focused on soil chemical and physical factors, and neglected biological factors (Sherwood and Uphoff 2000). Concern about soil health is motivated by present and future interest in

both agricultural productivity and profitability. Research on the implications of agricultural management on soil biology is in its infancy, and many important findings will become evident in the coming years.

References

Anon 2001. Rangeland soil quality: Soil biota. USDA Natural Resources Conservation Service, Rangeland Sheet 8.

Araujo ASF, Monteiro RTR, Abarkeli RB 2003. Effect of glyphosate on the microbial activity of two Brazilian soils. Chemosphere 52 799-804.

Brimner TA, Boland GJ 2003. A review of the non-target effects of fungi used to biologically control plant diseases. Agriculture, Ecosystems and Environment 100 3-16.

Busse MD, Ratcliff AW, Shestak CJ, Powers RF 2001. Glyphosate toxicity and the effects of long-term vegetation control on soil microbial communities. Soil Biology & Biochemistry 33 1777-1789.

Cernakova M, Zemanovicova A 1998. Microbial activity of soil contaminated with chlorinated phenol derivatives. Folia Microbiologia 43 411-416.

Chen S-K, Edwards CA, Subler S 2001. Effects of the fungicides benomyl, captan and chlorothalonil on soil microbial activity and nitrogen dynamics in laboratory incubations. Soil Biology & Biochemistry 33 1971-1980.

Das AC, Debnath A, Mukherjee D 2003. Effect of the herbicides oxadiazon and oxyfluorfen on phosphate- solubilising microorganisms and their persistence in rice fields. Chemosphere 53 217-221.

Dungan RS, Ibekwe AM, Yates SR 2003. Effect of propargyl bromide and 1,3-dichloropropene on microbial communities in an organically amended soil. FEMS Microbiology Ecology 42 75-87.

Edvantoro BB, Naidu R, Megharaj M, Singleton I 2003. Changes in microbial properties associated with long-term arsenic and DDT contaminated soils at disused cattle sip sites. Ecotoxicology and Environmental Safety 55 344-351.

Garcia C, Gong P, Sun TH, Beudert G, Hahn HH 1997. Ecological effects of combined organic or inorganic pollution on soil microbial activities. Water, Air, and Soil Pollution 96 133-143.

Gaw SK, Palmer G, Kim ND, Wilkins AL 2003. Preliminary evidence that copper inhibits the degradation of DDT to DDE in pip and stonefruit orchard soils in the Auckland region, New Zealand. Environmental Pollution 122 1-5.

Gong P, Sun TH, Beudert G, Hahn HH 1997. Ecological effects of combined organic or inorganic pollution on soil microbial activities. Water, Air, and Soil Pollution 96 133-143.

Islam KR, Weil RR 2000. Land use effects on soil quality in a tropical forest ecosystem of Bangladesh. Agriculture, Ecosystems & Environment 79 9-16.

Lee KE 1985. Earthworms: their ecology and relationships with soils and land use. Academic Press, Australia.

Liess M 2004. Enhancing realism and practicability in ecotoxicological risk assessment. Proceedings of Interact 2004, Gold Coast, Australia, 123.

Martikainen E, Haimi J, Ahtiainen J 1998. Effects of dimethoate and benomyl on soil organisms and soil processes: A microcosm study. Applied Soil Ecology 9 381-387.

Megharaj M, Kantachote D, Singleton I, Naidu, R 2000. Effects of long-term contamination of DDT on soil microflora with special reference to soil algae and algal transformations of DDT. Environmental Pollution 109 35-42.

Merrington G, Rogers SL, Van Zwieten L 2002. The potential impact of long-term copper fungicide usage on soil microbial biomass and microbial activity in an avocado orchard. Australian Journal of Soil Research 40 749-759.

Merry RH, Tiller KG, Alston AM 1986. The effects of soil contamination with copper, lead and arsenic on the growth and composition of plants. Plant and Soil 95 255-269.

Mitra J, Raghu K 1998. Long-term DDT pollution in tropical soils: Effect of DDT and degradation products on soil microbial activities leading to soil fertility. Bulletin of Environmental Contamination Toxicology 60 585-591.

Monkiedje A, Ilori MO, Spiteller M 2002. Soil quality changes resulting from the application of the fungicides mefenoxam and metalaxyl to a sandy loam soil. Soil Biology & Biochemistry 34 1939-1948.

Mosleh Y, Paris-Palacios S, Couderchet M, Vernet G 2003. Effects of the herbicide isoproturon on survival, growth rate, and protein content of mature worms (*Lumbricus terrestris L.*) and its fate in the soil. Applied Soil Ecology 23 69-77.

Panda S, Sahu SK 1999. Effects of malathion on the growth and reproduction of *Drawida willsi* (Oligochaete) under laboratory conditions. Soil Biology & Biochemistry
31 363-366.

Panda S, Sahu SK 2004. Recovery of acetylcholine esterase activity of *Drawida willsi* (Oligochaete) following application of three pesticides to soil. Chemosphere 55 283-290.

Pandey S, Singh DK 2004. Total bacterial and fungal population after chlorpyrifos and quinalphos treatments in groundnut (*Arachis hypogaea L.*) soils. Chemosphere 55 197-205.

Pankhurst CE 1997. Biodiversity of soil organisms as an indicator of soil health. In Biological indicators of soil health. Eds Pankhurst CE, Doube BM, Gupta VVSR pp 297-324. CAB International Wallingford.

Rovira AD 1994. The effect of farming practices on the soil biota. In Soil biota: Management in sustainable farming. Ed Pankhurst CE pp 81-87. CSIRO Australia.

Sannino F, Gianfreda L 2001. Pesticide influence on soil enzymatic activities. Chemosphere 45 417-425.

Seghers D, Verthe K, Reheul D, Bulcke R, Siciliano SD, Verstraete W, Top EM 2003. Effect of long-term herbicide applications on the bacterial community structure and function in an agricultural soil. FEMS Microbiology Ecology 46 139-146.

Sherwood S, Uphoff N 2000. Soil health: Research, practice and policy for a more regenerative agriculture. Applied Soil Ecology 15 85-97.

Singleton PL, Addison B 1999. Effects of cattle treading on physical properties of three soils used for dairy farming in the Waikato, North Island, New Zealand. Australian Journal of Soil Research 37 891-902.

Smith MD, Hartnett DC, Rice CW 2000. Effects of long-term fungicide applications on microbial properties in tallgrass prairie soil. Soil Biology & Biochemistry. 32 935-946.

Stamatiadis S, Werner M, Buchanan M 1999. Field assessment of soil quality as affected by compost and fertiliser application in a broccoli field (San Benito County, California). Applied Soil Ecology 12 217-225.

Strandberg M, Scott-Fordsmand JJ 2004. Effects of pendimethalin at lower trophic levels-A review. Ecotoxicology and Environmental Safety 57 190-201.

Taiwo LB, Oso BA 1997. The influence of some pesticides on soil microbial flora in relation to changes in nutrient level, rock phosphate solubilisation and P release, under laboratory conditions. Agriculture, Ecosystems and Environment 65 59-68.

Vaclavik E, Halling-Sørensen B, Ingerslev F 2004. Evaluation of manometric respiration tests to assess the effects of veterinary antibiotics in soil. Chemosphere (in press).

Van Zwieten L, Ayres M, Morris S 2003. Influence of arsenic co-contamination on DDT breakdown and microbial activity. Environmental Pollution 124 331-339.

Van Zwieten L, Rust J, Kingston T, Merrington G, Morris, S 2004. Influence of copper fungicide residues on occurrence of earthworms in avocado orchard soils. Science of the Total Environment 63 59-68.

Westergaard K, Muller AK, Christensen S, Bloem J, Sorensen SJ 2001. Effects of tylosin as a disturbance on the soil microbial community. Soil Biology & Biochemistry
33 2061-2071.

Wilke B-M 1997. Effects of non-pesticide organic pollutants on soil microbial activity. Advances in GeoEcology 30 117-132.

Winding A, Binnerup SJ, Pritchard H 2004. Non-target effects of bacterial biological control agents suppressing root pathogenic fungi. FEMS Microbiology Ecology 47 129-141.

Contact details

Dr Lukas Van Zwieten
Research Scientist
Wollongbar Agricultural Institute
NSWDPI 1243 Bruxner Highway, Wollongbar NSW 2477
Phone 02 6626 1126
Fax 02 6628 3264
Email lukas.van.zwieten@dpi.nsw.gov.au

Can we manipulate resource availability to drive changes in microbial carbon assimilation and nitrogen cycling?

Frances Hoyle, Dan Murphy
University of Western Australia

Introduction

Decomposition of soil organic matter (SOM) is controlled by microorganisms and leads to the release of nitrogen and other nutrients. However, soil microbial activity is often limited by the absence of readily available carbon-based substrates. This paper discusses the effect of stubble management (stubble burning vs. stubble retention) and temperature on microbial carbon respiration and nitrogen cycling. We then examine the application of labile carbon substrates under controlled conditions on microbial process rates.

Nitrogen supply in dryland agricultural systems is derived predominantly from microbial decomposition of SOM and crop residues as a response to water-limited yield potentials and low inorganic fertiliser inputs (eg less than 50 kg N/ha for wheat production in Western Australia). In the typically low input agricultural systems of WA, up to 80% of soil nitrogen supply in wheat is therefore obtained from microbial transformation of SOM (Angus 2001), compared with 50% in temperate soils (Jenkinson 2001). In many farming systems the amounts of nitrogen cycling through soils during a year are more than enough, even where no fertiliser is applied, to satisfy crop nitrogen demand (see Table 1, Murphy et al, this proceedings). In WA, the role of soil microorganisms can therefore be highly significant in determining the amount and timing of biologically derived nitrogen. Thus an economic benefit can be associated with management of soils for optimal biological fertility influencing potential yield. It is therefore important to identify and understand the primary factors influencing microbial mass and activity in this environment and therefore the potential limitations to gross nitrogen transformation rates (ie total supply of inorganic nitrogen from microbes).

Effect of residue management on carbon and nitrogen cycling

Resource availability influences both carbon and nitrogen dynamics in soil. By limiting readily available carbon, reduced residue inputs can also influence population density and immobilisation of nitrogen which is essential for maintaining microbial growth and activity. Burning stubble for example, has been associated with depletion of SOM levels and declines in microbial biomass and/or activity (Powlson 1987).

Soil nitrogen released from crop residues and soil organic matter results primarily from the activity of microorganisms. Therefore any change in their mass (microbial biomass), or their activity (CO_2-C evolution) can result in changes to the rate of biological soil nitrogen supply. Plant residues are a primary form of organic matter utilised by microorganisms for microbial growth and activity. Since microorganisms are usually starved in soil because they lack available carbon food sources, retention of crop residues provides a practical means of increasing the size of the microbial population. This section investigates the effect of retaining or burning stubble on microbial carbon and nitrogen process rates in a low rainfall (< 325 mm annual rainfall) environment on a red-brown earth (Red Chromosol). Additional trial data and agronomic responses are presented in the poster abstract (Hoyle, Murphy this proceedings).

Stubble retention increased microbial biomass in the surface layer (0-5 cm) of the soil by up to 45% compared with burnt treatments, though no significant difference in biomass was observed below 5 cm (Figure 1). Microbial biomass carbon measured to 30 cm depth in this trial was 423 kg C/ha in stubble retained treatments (ie total mass of microorganisms was equivalent to 18 sheep/ha) and 310 kg C/ha in burnt treatments (equivalent to 13 sheep/ha). Microbial biomass nitrogen was measured at 89 and 67 kg N/ha for stubble retained and burnt treatments respectively (data not presented). This means that there is equivalent to 192 kg/ha (stubble retained) and 146 kg/ha (stubble burnt) of urea contained within the soil microorganisms, a significant source of potentially plant available nitrogen.

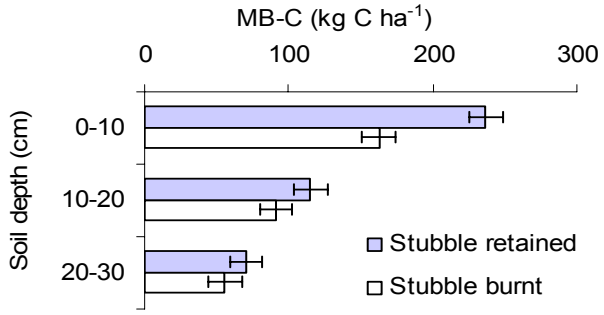


Figure 1. The effect of long-term (17 year) stubble management (retained versus burnt) on microbial biomass carbon (MB-C) at different soil depths (Hoyle et al unpublished).

Cumulative activity (measured as CO_2-C respiration) was up to 219% greater in stubble retained treatments compared with burnt stubble treatments (Figure 2) after seven days, demonstrating that changes in microbial processes were attributable to the quantity and quality of organic carbon. The average daily rate for CO_2-C evolution measured was 1.27 µg CO_2-C/g/soil $^{\circ}$C for stubble retained and 0.63 µg CO_2-C/g/soil $^{\circ}$C for burnt treatments (data not presented). These results also illustrate that stubble retention promoted both a larger but also more active microbial community compared with burning.

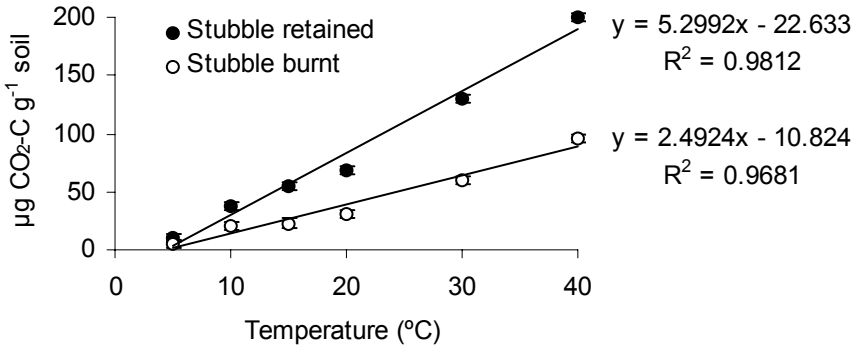


Figure 2. The effect of long-term stubble management (retained versus burnt) and incubation temperature on microbial activity (cumulative CO_2-C respired) after seven days incubation (Hoyle et al unpublished).

Effect of temperature on nitrogen cycling

The influence of environmental regulators such as temperature and moisture must also be considered as they control microbial activity and regulate substrate availability. For

example, substrate availability in semi-arid soils may be constrained by slower diffusion of nutrients and dissolved OM in soils, with poor connectivity between water films. Few studies have so far examined the effects of high soil temperatures, as typically experienced in Mediterranean or semi-arid environments, on carbon and nitrogen cycling. In particular there is a lack of data on the kinetics of gross mineralisation, immobilisation and nitrification rates at high temperature (> 20°C). This section investigates the effect of stubble retention vs. burning on short-term gross nitrogen transformation rates at constant temperatures of 5, 10, 15, 20, 30 and 40°C using ^{15}N isotopic pool dilution (Murphy et al 2003) on a red-brown earth (Red Chromosol).

Increases in the inorganic nitrogen concentration measured in our study (data not presented), suggest both mineralisation and nitrification processes remain active at higher temperature. Gross nitrogen mineralisation increased linearly between 5 and 40°C in burnt stubble treatments and between 5 and 30°C in stubble retained treatments (Figures 3a, 3b). In stubble retained treatments, a rapid increase in gross mineralisation rates measured at 40°C resulted in significantly ($P<0.001$, LSD=2.921) higher mineralisation rates than either those observed in burnt stubble treatments (14.8 and 2.7 µg N/g soil respectively), or at other incubation temperatures (0.8-3.8 µg N/g soil). Microbial immobilisation reached a plateau at 15°C (Figures 3a, 3b), while mineralisation continued to increase. This indicates a separation in the mineralisation (microbial nitrogen supply) immobilisation (microbial demand) turnover that was more apparent at high temperature and in stubble retained treatments. Therefore below 20°C, as is commonly experienced in temperate systems, microbial supply and demand are relatively well matched, so farming systems are less likely to lose nitrogen at these temperatures. However, in warmer climates this means that if water is not a limiting factor at high temperature (eg summer rainfall events), mineralisation can occur, but microbial immobilisation is constrained, leading to an accumulation of inorganic nitrogen susceptible to leaching.

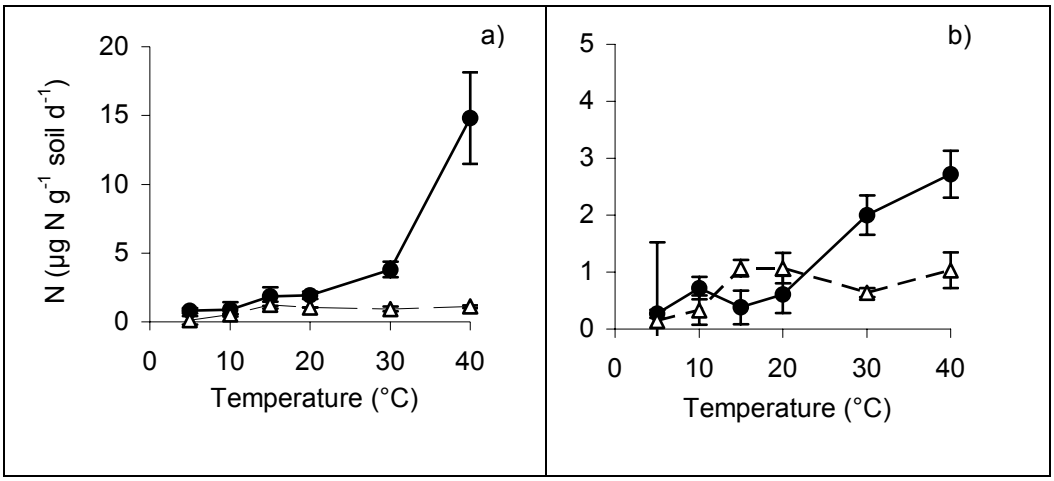


Figure 3. Effect of temperature on gross nitrogen mineralisation rate (●) and immobilisation (Δ) in (a) stubble retained and (b) burnt treatments (Hoyle et al unpublished). Capped bars may be smaller than symbols and represent the 95% confidence interval.

Previous research has demonstrated that below a carbon:nitrogen ratio of approximately 22, nitrogen is in excess of microbial demand and released, resulting in an oversupply of nitrogen for microbial use. This means there is more nitrogen available for nitrification and thus the nitrification: immobilisation (N/I) ratio increases. This relationship can be used to determine the risk of increasing nitrate losses within a soil,

and the impact of changing management practice on nitrogen retention vs. loss pathways. Stockdale et al (2002) demonstrate this relationship in temperate grassland soils with greater loss of nitrate associated with increasing N/I ratio (Figure 4a). In our study, soil temperature had a greater influence on the N/I ration than stubble management treatments, indicating potential losses of nitrate are greater in soils at low (5°C) and high (30 and 40°C) temperature (Figure 4b). This illustrates that environmental variables often override management practices in terms of microbial activity.

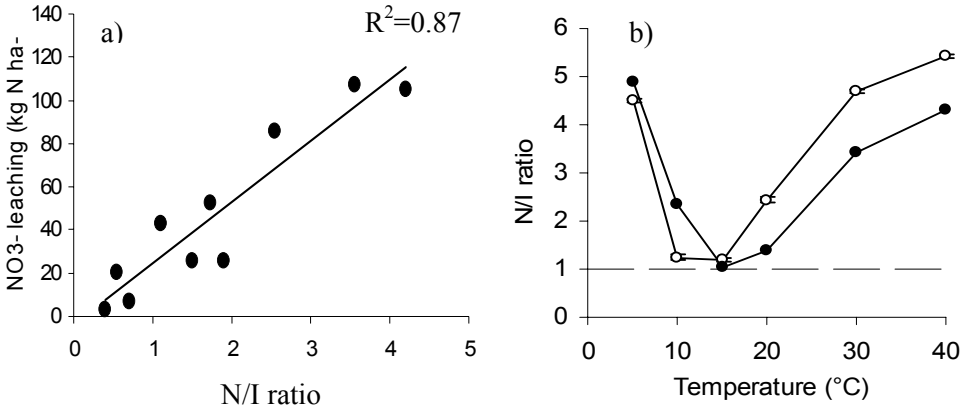


Figure 4. (a) Relationship between N/I ratio and nitrate leaching losses for a temperate grassland (Stockdale et al 2002). (b) Effect of temperature on the N/I ratio in stubble retained (○) and stubble burnt (●) treatments in WA (Hoyle et al unpublished). The dashed line represents a ratio of 1 (ie gross nitrification=microbial immobilisation).

Studies of the processes controlling soil nitrogen supply confirm that there is a strong influence of soil temperature, water potential and residue quality on gross nitrogen fluxes in soil. Therefore, by changing either the physical status of soil (ie soil pore structure, size and connectivity, water holding capacity), or increasing food supply (ie through increased organic matter returns or application of carbon substrates), we can alter the rate of decomposition and shift the dominance of rates for mineralisation, immobilisation, and nitrification. Since mineralisation and immobilisation are carbon driven and nitrification is nitrogen driven, we can potentially alter the N/I ratio in favour of nitrogen retention by microbes instead of nitrogen loss via leaching. This data illustrates that the relative dominance of these microbial nitrogen pathways defines the amount of plant available nutrients, as well as the capacity of a soil to leach nitrate.

What is the capacity to manipulate microbial carbon and nitrogen processes?

The microbial assimilation of nitrogen is closely linked to the availability of organic carbon to sustain growth and energy requirements. Therefore changes in the mass of microorganisms and their activity are commonly reflected in changes to the soil supply rate of both carbon and nitrogen. Labile carbon substrates may therefore be used in theory to manipulate immobilisation (consumption of ammonium (NH_4) by microbes) in favour of nitrification (loss process), and hence alter the timing of inorganic nitrogen release. The intent is to drive microbial processes toward immobilisation and away from nitrification (which causes nitrate losses).

Although significant quantities of SOM are usually present, it is largely resistant to decomposition and does not provide sufficient maintenance energy for the soil microbial biomass. De Nobili et al (2001) showed that additions of labile carbon substrates (11.3-34 mg C/kg soil; ≈ 8 and 24 kg C/ha) such as glucose, amino acids or root exudates to temperate soils caused more CO_2-C to be evolved than was contained in the original substrate (ie theoretically 'feeding the microbes'). They proposed that this increased CO_2 production may result from an evolutionary strategy used by soil microorganisms to become 'metabolically alert' in response to a forthcoming food event. In this trigger molecule theory, De Nobili et al (2001) hypothesised that detection of these molecules would indicate the pending arrival of fresh substrate and cause the microbial biomass to activate itself in anticipation of a more significant forthcoming food event. Thus the microbial biomass, by investing more energy than was contained in the original substrate, may gain a later benefit. Such a microbial response can in theory also be used to manipulate the dominant nitrogen transformation pathway in favour of nitrogen microbial retention instead of nitrogen loss. However, suitable products or management strategies are yet to be well defined.

This section investigates the effect of glucose-carbon to determine whether microorganisms can be manipulated in such a way on low fertility coarse textured soil from WA. In our experiment, the addition of glucose-carbon to non-cellulose (labile carbon only) and cellulose amended (labile carbon + stable carbon applied) soils resulted in significantly greater ($P<0.05$) evolution of cumulative CO_2-C compared with the control (data not presented). However, the cumulative amounts of respired carbon after addition of glucose-carbon to these soils indicated no additional release of CO_2-C from the microorganisms following application of glucose to either non-cellulose amended or cellulose amended soils (Figure 5, Hoyle et al, submitted). In cellulose amended soils, CO_2-C evolution was often lower than in control soils suggesting the addition of glucose inhibited cellulase activity, possibly by end-product inhibition (Figure 5).

An alternative interpretation of our findings and those of De Nobili et al (2001), suggest any delayed CO_2-C evolution is therefore more likely an indirect result of the applied glucose. For example CO_2-C evolution may result from differences in basal CO_2-C evolution measured between glucose-amended and non-amended soil, due to the activation of different microbial populations on addition of glucose-carbon, or an accelerated turnover of microbial biomass-carbon, or the formation of a glucose-derived metabolite more readily decomposed by a greater diversity of microorganisms. Therefore although the 'trigger molecule' concept proposed by De Nobili et al (2001) was not evident in soils tested in WA, the application of a labile carbon substrate (in this case, glucose-carbon) resulted in changes to microbial activity and carbon cycling. The implications of these findings to WA farming practices are currently under investigation.

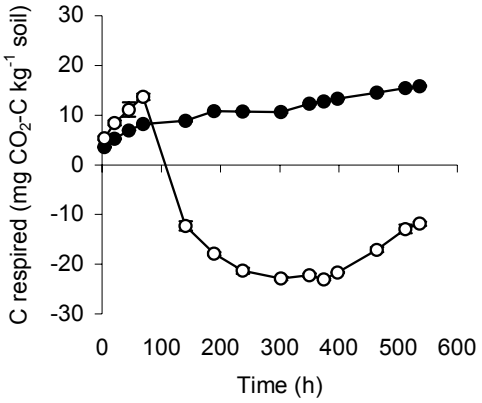


Figure 5. Cumulative CO_2-C evolved (treatment minus control) after addition of a single glucose-carbon solution at 30 mg C/kg soil (≈ 21 kg C/ha) at 0 hours to cellulose amended (○) and non-cellulose amended soil (●) in a coarse textured soil from WA (data from Hoyle et al submitted). Capped bars representing standard errors (n=3) are plotted for each sampling point, and may be smaller than symbols.

Asynchrony of N cycling

Carbon substrates, plant residues or soil amendments that change the number of biologically active days, or alter activity and function may enhance the timing of release and/or plant nutrient uptake. Plant residues are a primary form of organic matter utilised by microorganisms for microbial growth and activity. Since microorganisms are usually starved in soil because they lack available carbon food sources, crop residues provide a practical means of increasing the size and/or activity of the microbial population. However, nutrients released from residue decomposition may be underutilised by a growing crop, as supply and demand are often not in synchrony (Murphy et al 2004, Ridley et al 2004).

Differences in plant residues and management influence organic matter quality, decomposition rate, and hence the timing and amount of nutrient release. For example, greater contact between plant residue and soil microorganisms, as may occur in a green manure (incorporated) phase can cause faster residue decomposition and hence greater nutrient release than the same residue retained with little or no disturbance (brown manure, desiccation only) as is demonstrated in Figure 6a. Asynchrony is illustrated in this example with high inorganic nitrogen levels early in the season when crop demand is low, demonstrating the potential for high nitrogen loss. Rainfall patterns (amount and distribution) will also significantly influence the release and location (synlocation) of nitrogen within the soil profile (Figure 6b) and hence the capacity of a growing crop to capture nitrogen. In the example demonstrated (Figure 6b), high rainfall is associated with greater distribution of inorganic nitrogen to depth. In this case, the wheat crop gains access to only 19% of the total nitrogen available within the soil profile 40 days after sowing in a high rainfall (leaching) year, compared with 50% under a non-leaching year. Similar improvements in nitrogen capture may also be achieved through management strategies such as deep ripping to improve root exploration (see Murphy et al 2004). These results illustrate that there is potential to improve the synchrony between plant growth and nitrogen uptake in WA farming systems, thereby reducing potential nitrogen losses.

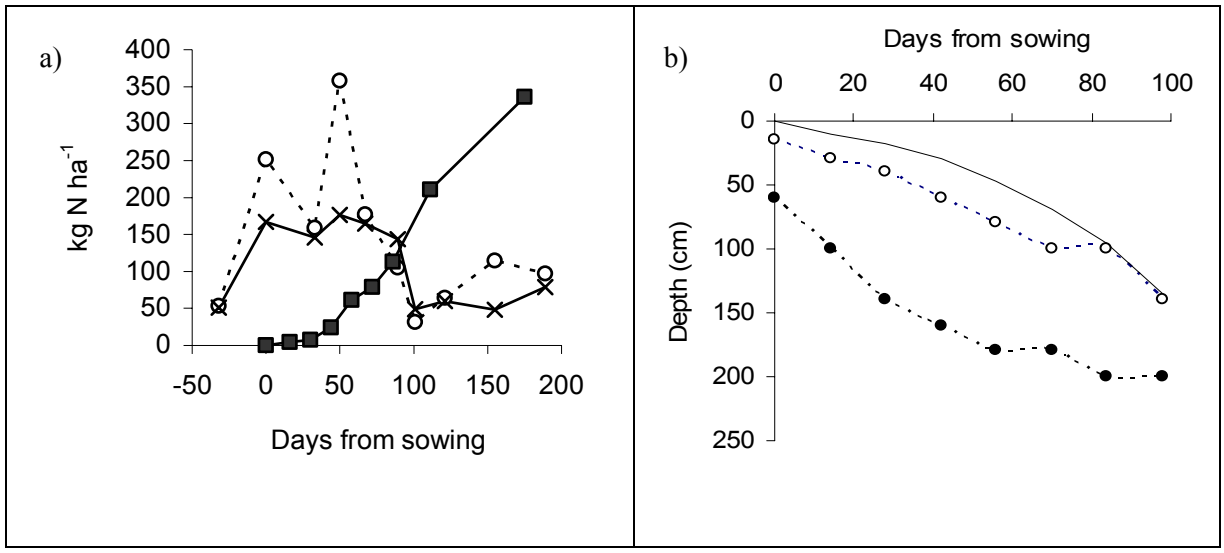


Figure 6. (a) Effect of field pea residue management (green manure (○) vs. brown manure (x) crop) on nitrogen release (0-50 cm) and synchrony with nitrogen uptake (■) in a subsequent wheat crop at East Beverley in 2002. Asynchrony is illustrated here with high inorganic nitrogen levels early in the season when crop demand is low. (b) Location of inorganic nitrogen in relation to the rooting zone (solid line) of a wheat crop using actual rainfall (216 mm) from East Beverley in 2002 (○) or simulating a high rainfall (444 mm) season (●). Data simulated using SYN (Hoyle and Murphy, unpublished).

Conclusion

Stubble retention has been shown to increase the amount of microorganisms in soil compared with stubble burning, resulting in greater soil nitrogen supply. Residue incorporation can also be an effective means of increasing biological soil nitrogen supply and potential grain production. However, potential yield depends on the ability of the subsequent grain crop to utilise this inorganic nitrogen, and is therefore influenced significantly by both rainfall pattern and intensity as this will alter the location of inorganic nitrogen in the soil profile compared with plant root development. Management strategies to optimise the synchrony between nutrient release and crop demand must therefore be considered.

Acknowledgements

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References

Angus JF 2001. Nitrogen supply and demand in Australian agriculture. Australian Journal of Experimental Agriculture 41 277-288.

De Nobili M, Contin M, Mondini C, Brookes PC 2001. Soil microbial biomass is triggered into activity by trace amounts of substrate. Soil Biology & Biochemistry 33 1163-1170.

Hoyle FC, Murphy DV, Brookes PC (submitted). Microbial response to addition of glucose. Soil Biology & Biochemistry (submitted).

Jenkinson DS 2001. Nitrogen in a global perspective, with focus on temperate areas – state of the art and global perspectives. Plant & Soil 228 3-15.

Murphy DV, Recous S, Stockdale EA, Fillery IRP, Jensen LS, Hatch DJ, Goulding KWT 2003. Gross nitrogen fluxes in soil: Theory, measurement and application of 15N pool dilution techniques. Advances in Agronomy 79 69-118.

Murphy DV, Stockdale EA, Hoyle FC, Smith JU, Fillery IRP, Milton N, Cookson WR, Brussaard L, Jones DL 2004. Matching supply with demand: Principle, practice and prediction. In Controlling nitrogen flows and losses. Eds Hatch DJ, Chadwick DR, Jarvis SC, Roker JA. Wageningen Academic Publishers, in press.

Powlson DS, Brookes PC, Christensen BT 1987. Measurement of microbial biomass provides an early indication of changes in total soil organic matter due to straw incorporation. Soil Biology & Biochemistry 19 159-164.

Ridley AM, Mele PM, Beverly CR 2004. Legume-based farming in Southern Australia: Developing sustainable systems to meet environmental challenges. Soil Biology & Biochemistry (in press).

Stockdale EA, Hatch DJ, Murphy DV, Ledgard SF, Watson CJ 2002. Verifying the nitrification to immobilisation ratio (N/I) as a key determinant of potential nitrate loss in grassland and arable soils. Agronomie 22 831-838.

Contact details

Frances Hoyle
Research Officer
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia
35 Stirling Hwy Crawley WA 6009
Phone 08 6488 7345
Fax 08 6488 1050
Email fhoyle@agric.uwa.edu.au

Registration of soil biological products

Colin Byrnes
Australian Pesticides & Veterinary Medicines Authority, Canberra

Introduction

The Australian Pesticides & Veterinary Medicines Authority (APVMA) was formerly known as the National Registration Authority for Agricultural and Veterinary Chemicals (NRA). The Authority was established in 1993 to administer the National Registration Scheme for agricultural and veterinary chemicals.

The principal legislation the APVMA administers is the Agricultural and Veterinary Chemicals Code Act 1994. The Agricultural and Veterinary Chemicals Code, known as the Agvet Code, provides for a common basis for evaluation, registration and control of supply of these products to apply in all states and participating territories.

Scope of the Agvet Code

The definition of an agricultural chemical product in the Agvet Code is quite broad. It covers substances or mixtures of substances, not just formulated chemicals. It includes an organism or part of an organism, material that is produced by an organism or matter whose production involves use of an organism. Plant extracts, pheromones, plant hormones, enzymes and vitamins, microbial pesticides and inserted genes that code for the production of pesticidal substances are agricultural chemical products if they are intended to be used for the purposes described below.

The definition captures substances that destroy, repel, stupefy, inhibit the feeding of, prevent attacks of or infestation of any pest in relation to a plant, place or thing, or that attract a pest for the purpose of its destruction. It also includes substances that destroy a plant or modify the physiology of a plant or pest to alter its natural development, productivity, quality or reproductive capacity, or modify an effect of another agricultural chemical product. Product types that fit the definition include insecticides, fungicides and herbicides, plant growth regulators, vertebrate pest control lures and baits, insect repellents or attractants, adjuvants that affect the activity of another product such as spreaders and stickers, wood preservatives, algaecides, swimming pool chemicals and antifouling paints.

Products do not have to provide complete control to come under the definition. Products that reduce the severity of a pest or disease attack are considered to require registration.

Certain substances can be declared not to be agricultural chemical products. There are a number of substances so declared in the Agricultural and Veterinary Chemicals Code Regulations. These include any soil ameliorant, conditioner or fertiliser if the product is not claimed to have any effect as a regulator of plant growth, any predatory insect, predatory mite or macroscopic parasite, and any hay inoculant, silage inoculant or legume inoculant, if the product is based on bacteria and/or enzymes.

The definition does not rest entirely on the claims to be made for the product. It also includes products represented, imported, manufactured, supplied or used for the purposes listed in the definition. Merely leaving claims off the label for a product that is clearly intended to be used for purposes defined for agricultural chemicals does not take the product outside the definition.

Specifically in regard to soil biological products, any product that is for use to control soil pests, weeds or diseases would require registration. Products that provide a population of beneficial organisms at the expense of detrimental organisms are considered to be exercising control and require registration. Products that produce hormones that stimulate plant growth require registration. Products that only produce a better nutrient balance in the soil do not require registration, nor do products that only aggregate soil particles to provide better soil structure (ie soil conditioners).

If a product comes under the definition of an agricultural chemical product, it must be registered under the Agvet Code before being supplied in Australia. The Agvet Code does not control the use of agricultural and veterinary chemical products. Each state and territory has its own control of use legislation.

Registration

Under the Agvet Code the APVMA in empowered to approve the active constituent(s) for use in the formulated product, register the formulated product and approve the labels for containers of the product.

The APVMA must assess an application for registration against criteria that include safety to users, other people and the environment, residues in food, trade impacts, effectiveness and safety to treated plants. The product must have a label that includes all the information required by the APVMA and that is specifically approved by the APVMA.

The APVMA has published *Guidelines for registering agricultural chemicals* to provide details of the data required to support applications for registration. A similar set of requirements has been published for veterinary products. These publications are available on the APVMA website (www.apvma.gov.au) which also includes labelling codes, application forms and other information to assist registrants.

In general, a submission to APVMA is divided into the following parts:

- Part 1 Application overview
- Part 2 Chemistry and manufacture
- Part 3 Toxicology
- Part 4 Metabolism and toxicokinetics
- Part 5A Residues
- Part 5B Overseas trade aspects of residues in food
- Part 6 Occupational health and safety
- Part 7 Environment
- Part 8 Efficacy and safety
- Part 9 Other trade aspects
- Part 10 Special data requirements

In many cases, biological products have different properties from conventional chemical products, so the APVMA has developed separate guidelines and data requirements to more appropriately address the potential risks posed by biological agricultural products. *Guidelines for the registration of biological agricultural products* are also available on

the APVMA website. These guidelines categorise biological agricultural products into the following four groups:

- Group 1— biological chemicals (eg pheromones, hormones, growth regulators, enzymes and vitamins)
- Group 2 — extracts (eg plant extracts, oils)
- Group 3 — microbial agents (eg bacteria, fungi, viruses, protozoa)
- Group 4 — other living organisms (eg microscopic insects, plants and animals plus some organisms that have been genetically modified)

Data requirements have been tailored to meet the specific issues and properties of the products in each of these groups. The guidelines are to be considered to provide guidance and not as absolute requirements. The APVMA recognises the need for flexibility in determining the data requirements for biological products. Where certain data are not considered necessary, relevant scientific arguments for their omission should be put forward.

Not all data must be generated in Australia. It is common for laboratory data, for example from toxicology studies, to be generated overseas. Australian data are usually required for residues in food and efficacy/plant safety aspects. Overseas data may also be submitted but its relevance to Australian conditions must be argued.

The APVMA expects that all laboratory studies should be conducted in accordance with an acceptable code of good laboratory practice (GLP). At this stage, GLP is mandatory only for residues studies. Field trials should be designed so that the data can be statistically analysed. Reports of all studies must be fully documented as for a scientific experiment.

Registration process

An application for registration of an agricultural chemical product is screened for general compliance with requirements, including correct form, fee, provision of draft proposed labels and data or argument on the various criteria the APVMA must assess as described above.

When the application passes screening it is evaluated by APVMA. This usually involves relevant data being considered by the Commonwealth Department of Health and Ageing and the Department of Environment and Heritage as well as by expert groups within APVMA that deal with chemistry and food residues aspects. Advice on efficacy aspects is usually sought from state departments of agriculture/ primary industries, although advice from other sources such as universities may sometimes be sought.

If the product is, or contains, a GM product as defined under the Gene Technology Act 2000 the APVMA must consult the Gene Technology Regulator in writing before granting the application and must take into account any advice received from the Gene Technology Regulator.

If the APVMA has not evaluated the active constituent previously a Public Release Summary outlining the APVMA's findings is prepared, its availability is published in the APVMA Gazette and there is a 28-day period for public comment.

The APVMA agrees to wording/format of the label and when copies of the final printed label are received registration is granted. Details of all new product registrations are

published each month in the APVMA Gazette. Details of registered products are available on the APVMA website. This is updated every 24 hours.

Permits

The APVMA can issue permits that allow supply and use of unregistered products or use of registered products for purposes or in a manner different from the instructions shown on the approved label.

There are three main types of permits.

Research permits

These permits allow the supply and use of products for scientific purposes, usually for the generation of data required for registration.

A general permit (PER7250) applies to small-scale research trials on research facilities or in other situations. This permit is subject to a number of conditions including constraints on the scale of the trials (for example an area of less than 5 ha nationally for food or fibre crops or 500 plants nationally for other crops/situations) and restriction on disposal of produce. Full details are specified in the permit. No application to APVMA is required in order to conduct trials that comply with this permit.

For other trials, an application to APVMA is required so a permit can be issued.

Minor use permits

These permits allow the use of registered products for purposes or in a manner not covered by the approved label. They are usually for crops where the area grown is small or the pest is of limited or localised incidence. The APVMA website has criteria for what constitutes a minor use. This includes a list of crops or situations classed as major, some parameters for determining what is a minor use in a major crop/situation (eg the lesser of either 10% of the national area of crop or 10,000 ha) and an option to provide an argument that registration would not produce sufficient economic return to the manufacturer.

Emergency use permits

These permits allow the supply and/or use of products to address unforseen problems such as the outbreak of an exotic pest or disease.

In assessing applications for each of these categories of permit, the APVMA must be satisfied of similar criteria to those applying to registration. In some cases the data requirements are similar, but in most cases the limited nature of the use allows some lesser data requirements to apply. It is also possible for the APVMA to conditionally grant a permit subject to further data being generated. This is not done for product registration.

Details of data requirements and other aspects relevant to permits are available on the APVMA website. The website also enables a search to be made for current minor use and emergency use permits.

Compliance

The APVMA is a partnership between the Commonwealth and the States/Territories under which the APVMA was established as a Commonwealth Statutory Authority, with responsibility for the evaluation, registration and review of agricultural and

veterinary chemicals, and their control up to the point of retail sale. The States and Territories retain responsibility for control-of-use activities, such as licensing of pest control operators and aerial spraying.

If products are not registered the APVMA has the power to require their recall from the market. Prosecution for an offence under the Agvet Code may also be undertaken.

Misuse of products is a matter for the relevant State/Territory authorities. It is generally an offence to use an unregistered agricultural chemical product.

Adverse Experience Reporting Program

The APVMA has operated an Adverse Experience Reporting Program (AERP) for a number of years for veterinary products. A similar scheme for agricultural chemical products commenced late last year.

The scope of the AERP is broad and allows for the receipt of adverse experience reports involving registered agricultural chemical products (as defined in the Agvet Code), when used according to label or APVMA permit directions, for:

- human health issues, where people are exposed to these products either by using them, consuming treated produce, or as bystanders
- animal health issues, including both domestic and native birds and animals
- crop and plant damage
- residue issues
- problems that lead to unacceptable exposure to users
- environmental damage
- lack of efficacy.

The scope of the program does not include:

- registered veterinary medicines (these are dealt with as part of the adverse experience reporting program for veterinary products)
- trade issues (these are dealt with under other programs within the APVMA)
- household or home garden product issues (such as damaged packaging of home-use pesticides not caused by the product itself, minor efficacy issues), which are dealt with under other protection laws such as consumer affairs, trade practices legislation etc
- packaging design faults
- illegal off-label uses (contrary to label or APVMA permit directions)
- products not registered by the APVMA.

Information on how to submit a report is available through the APVMA web site, www.apvma.gov.au, which makes the reporting system as accessible as possible for chemical users and the general public.

The web site also defines levels of adverse experiences, and provides guidelines for submission and evaluation.

Based on the assessment of adverse experience reports certain risk mitigation strategies or corrective actions may be required. These may include, but are not restricted to, the following:

- registration amendments, such as label changes, changes to the method of manufacture or product's physical or chemical design, changes to container design, changes to production line processes, or suspension and/or cancellation of registration and approval
- referral for action, such as compliance action, including product and batch recalls, referral to state authorities for action, or nomination of products or active constituents for formal chemical review by the APVMA, (note that once the recommendation for review has been made by the AERP the review team in the Pesticides Program will consult further with advisory agencies and other experts to determine whether a review is necessary and, if so, the scope of that review)
- education and publicity, such as providing scientific papers or articles on issues identified for relevant journals, magazines or newspapers.

Conclusion

Many soil biological products would fall under the definition of agricultural chemical products in the Agvet code. These products are required to be registered before being supplied or used in Australia.

The registration process evaluates the safety, environmental effects, food residues, trade and efficacy aspects of the product in the interests of the user of the product, the consumer or exporter of any produce from treated soil and the community generally. The APVMA provides a mechanism for users to report adverse experiences with registered products and for these to be evaluated.

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References

Australian Pesticides and Veterinary Medicines Authority 2004. Adverse Experience Reporting Program for Agricultural Chemicals, APVMA Canberra, http://www.apvma.gov.au/qa/aerp_ag.shtml

Australian Pesticides and Veterinary Medicines Authority 2004. Ag manual: The requirements manual for agricultural chemicals. APVMA, Canberra. http://www.apvma.gov.au/guidelines/ag_manual.shtml.

Australian Pesticides and Veterinary Medicines Authority 2004. Ag requirements series: Guidelines for registering agricultural chemicals. APVMA, Canberra. http://www.apvma.gov.au/guidelines/requirements_ag.shtml.

Australian Pesticides and Veterinary Medicines Authority 2001. Ag labelling code: Code of practice for labelling agricultural chemical products. APVMA, Canberra. http://www.apvma.gov.au/publications/ag_labelling_code.shtml.

Australian Pesticides and Veterinary Medicines Authority 2000. Guidelines for the registration of biological agricultural products. APVMA, Canberra. http://www.apvma.gov.au/guidelines/bioagprod.shtml.

Contact details

Colin Byrnes
Manager Fungicides
Pesticides Program
APVMA
PO Box E240 Kingston ACT 2604
Phone 02 6272 4850
Fax 02 6272 3218
Email colin.byrnes@apvma.gov.au

Development of experimental protocols for evaluating beneficial soil biological products

Robert Hannam
R J Hannam & Co Pty Ltd (For GRDC)

Introduction

A dominant research theme in the GRDC Soil Biology Initiative is to support development of prospective soil-borne microorganisms for beneficial application in the grains industry. There are three areas of interest where inoculated microorganisms may be beneficial:

- suppression of soil-borne root diseases
- stimulation of plant growth
- enhancing access of plant roots to soil-bound nutrients

Some microorganisms may provide one or more of these benefits. However, in the context of this paper, we are concerned mainly with those that are antagonistic to soil-borne cereal diseases.

Australian and overseas research, sometimes in collaborative arrangements, is active in identifying, isolating and developing beneficial microorganisms for application against a range of diseases, mainly in intensive horticulture and broadacre cropping systems. Hence the Australian grains industry has potential access to organisms from Australian soils and from other countries. There is also potential for the development of genetically modified organisms (GMOs) with specifically introduced traits if and when the industry accepts them. For any of these organisms to be marketed in Australian environments, they need to comply with the Agricultural and Veterinary Chemicals Code which seeks to protect the health and safety of people, animals and the environment and the domestic and export market potential of our agricultural industries. The code is administered by the Australian Pesticides and Veterinary Medicines Authority (APVMA).

The GRDC is currently developing a business model through which to commercialise relevant outputs from their research investments. The beneficial microorganisms are key candidates for this process and must comply with the necessary registration requirements. The APVMA has developed guidelines designed specifically for registration of biological agricultural products, largely on the basis that they are deemed to represent a lower risk than synthetic pesticide chemicals. Under these guidelines, bio-inoculant microorganisms are assigned to a group 3 category of 'microbial agents' (bacteria, fungi, viruses, protozoa).

Bacterial legume inoculants (*rhizobium*), and products that stimulate plant growth and make no claims for pest control or specific growth regulation do not require registration. Products based on plant hormones do require registration. Biologically derived chemicals which have toxic effects are usually not classed as biological products and are subject to normal agricultural chemical provisions. GMOs require registration and also more stringent technical data relating to the genetic manipulation, traits, stability and environmental expression.

The key elements of registration under the soil biological guidelines for which details may be required include:

- active agent description, properties, formulation, storage
- toxicology or pathogenicity to humans and other mammals, metabolism and residues of compounds if applicable
- occupational health and safety: risks of exposure to biological products.
- environmental risks: toxicity (plants and animals), pathogenicity, fate, behaviour, survival, hazards
- efficacy and safety: justification, performance, lab-pot-field data, side effects, integration with pest management, safety to target plants, phytotoxicity to non-target crops, animal safety.

This paper deals mainly with gathering the efficacy information for beneficial soil-borne organisms for cereals.

In the research programs, it is common that the effect of beneficial microorganisms against soil-borne diseases is demonstrated first in laboratory and pot-based experiments. Usually, isolates of a single species are tested against a single pathogen and those that show promise are tested under field conditions against the target disease. However the variability in spatial distribution of pathogens in field soils and in seasonal expression of disease makes it difficult to reliably demonstrate responses to inoculants, which experience has shown are usually in the order of 5-10%. Experience with this approach indicates that positive responses are measured only in around 10-20% of trials due to these factors, which may mask the true potential of the beneficial bio-inoculants and encourage a negative marketing image.

To demonstrate efficacy, the APVMA requires statistically positive results over at least two seasons and in representative market areas in at least two States. If beneficial bio-inoculants are to be accepted by Australian grain growers, reliable, credible response data is essential.

To embrace this challenge, the GRDC is currently establishing a system to provide independent comparative evaluation of the effect of beneficial organisms on common cereal root diseases. The aim is to establish a rigorous series of laboratory, bio-assay and field-based experimental regimes that satisfy APVMA requirements and support market development of the better performing organisms. The system is being developed in 2004 for application in 2005 and beyond.

Materials and methods

The system for screening and proving efficacy of beneficial bio-inoculants has three linked components planned for development. The target pathogens initially are rhizoctonia, take-all and pythium.

Laboratory tests

Protocols for two assays are being developed to enable initial comparative assessment of prospective beneficial organisms for their anti-pathogenic activity and effects on seedling growth and disease resistance when applied to wheat seeds.

Quantitative in vitro anti-fungal assays (direct interactions)
The most appropriate media (with advice from the submitting organisation) and growth conditions for both pathogens and beneficial organisms will be determined, and will include media that potentially enhance production of the biocontrol agent or are similar in composition to soil solution. The beneficial organisms will be tested against a number of strains of pathogens.

Miniaturised in planta assay (indirect interactions)
A range of beneficial microorganisms will be inoculated onto seed infected with target pathogens. Seedling growth and development of disease symptoms on their roots will be monitored.

Once established, this system should be able to screen a significant number of organisms (and combinations if required) in a relatively rapid throughput system which can have a number of cycles per year. The better performing candidates, taking into account any other relevant information which may be provided by the developers, will move through to the pot bioassay system.

Pot bioassays

The bioassay system will be used to further evaluate and shortlist potential beneficial organisms for inclusion in field trials. An outdoor terraced pot culture system similar to a high throughput system used to screen breeder's lines for resistance to cereal cyst nematode will be developed. Seed inoculated with different beneficial microorganisms will be sown into a range of natural soils inoculated with take-all, rhizoctonia or pythium. Seeding times will be chosen to optimise conditions for disease expression. The number of pot replicates and disease levels to produce a reliable quantitative bioassay will be determined and appropriate controls will be applied.

A range of four soil types which represent the broad variation experienced in the field will be used initially. These are a calcareous sand, acid sand, black earth and red brown earth which will be sourced in South Australia due to quarantine restrictions. Disease impact will be based on dry matter responses and appearance of whiteheads (take-all). Root disease assessments will be limited and chosen from across a range of controls and selected treatments.

Field trials

Field trials need to reliably measure grain yield differences of at least 5-10% to test the effect of biocontrol agents in reducing soil-borne disease. Hence field trial designs need to minimise the field variability of the target soil-borne pathogens and establish adequate levels of pathogens in soil to reliably test the biocontrol microorganisms. We are attempting to develop a field trial protocol in which the take-all and rhizoctonia are artificially introduced into field plots to create a robust test environment with manageable replication. The more extensive distribution of pythium in soils is such that artificial inoculation of the pathogen is not warranted.

This year we have established six trials in South Australia at locations with high yield potential to help differentiate modest yield differences. One rhizoctonia and one take-all trial have been established at each of two locations on contrasting soil types, a red brown earth and a brown calcareous loam over limestone. Two pythium trials have been established on a red brown clay loam and a brown gravelly clay. There are six replicates in all trials. Beneficial organisms from several research programs are being compared

for relative effects against one or all diseases by being inoculated onto seed before or at seeding.

All plots were seeded with a district standard CCN-resistant wheat variety and a moderate plane of nutrition. Seed was pickled with standard fungicides and an appropriate weed management strategy employed. We have provided for the take-all sites to be irrigated in spring if necessary to encourage development of the disease in test plots.

Rhizoctonia infection levels will be estimated with dry matter cuts at late tillering and take-all infection levels will be evaluated by appearance of whiteheads in spring. Disease symptoms on roots will be assessed only in pathogen inoculum control plots of both the rhizoctonia and take-all trials due to the constraints on undertaking root assessments on all plots. For pythium, disease levels will be estimated by rhizosphere soil dilution and an incubation technique developed by Paul Harvey (CSIRO Land & Water). Grain yield and quality will be measured at crop maturity.

To the extent that resources allow, a DNA assay of root plus attached rhizosphere soil for the target pathogens will be developed and evaluated as an objective measure of the level of pathogen present and the extent to which the anti-fungal organisms are able to reduce pathogen levels in and on roots.

The trial sites at which rhizoctonia or take-all have been artificially inoculated will be managed as a chemical fallow for one season following the completion of the trials to eliminate disease hosts and allow pathogen levels to decline to paddock background levels.

Progress

At the time of submitting this paper, only the field trials have been established. Development of the laboratory assay and pot bioassay projects are subject to funding being approved by GRDC.

Discussion

The protocols developed in this project should provide the basis for rigorous comparative evaluation of beneficial microorganism inoculants which are able to reduce soil-borne root diseases of cereals. Transition through a series of assays including laboratory, pot culture and field trials should allow for the better performing organisms with commercial potential to emerge.

It may be possible to establish some key performance benchmarks for candidate organisms to achieve before they are deemed suitable for commercialisation. These benchmarks may take the form of performance relative to a well proven bio-inoculant (similar to wheat variety performance comparison against a nominated variety) or to a fungicide seed dressing treatment of known performance.

While the system seeks to identify the most promising organisms for commercialisation and application on-farm, the process is also designed to assist with compliance with APVMA registration requirements.

Organisms showing promise from both Australian and international research programs would be possible candidates for this system. However, imported organisms would need to comply with Australian quarantine regulations.

While these protocols will assist with proof of efficacy for the beneficial bio-inoculants, there are many other factors which may affect the acceptance of any organisms within the Australian grains industry. Some examples are:

- development of a reliable formulation for carrying and supporting the bio-inoculant through commercial seeding operations
- the levels of bio-inoculants that need to be applied to seed to ensure adequate numbers survive storage, handling and seeding operations in a commercial system
- capacity of the bio-inoculant to tolerate commercial seed pickles – a reality
- well developed instructions for the preparation, storage and application of the organisms
- risk of translocation of any toxic compounds into plant tops and seed and the implications for mammalian health
- risk to operators in handling the bio-inoculant
- need for evaluation of the risk of non-target negative effects of the introduced organisms
- possible interactions of bio-inoculants with commercial herbicides and pesticides.

The stage in the product development pipeline at which these and other important factors are determined or resolved remains to be determined. Perhaps in future GRDC contracts supporting the development of beneficial bio-inoculants, the critical factors for early resolution should be identified in project milestones.

Ideally, candidate organisms submitted to the field component of the beneficial organism evaluation system described above should at least:

- have strong evidence of benefits against target pathogens
- have a good understanding of the critical factors associated with targeting where they have application or where they should not be used
- have advanced instructions on product storage, handling and application
- be provided in a suitable formulation for field use
- be proven to tolerate commercial grain pickles
- have the confidence of the submitting researcher or organisation that they can be applied in the field by independent operators.

The primary aims of developing an independent comparative evaluation system for advanced beneficial biological products are to:

- confirm relative efficacy among a range of candidate organisms for target pathogens
- comply with APVMA efficacy registration requirements
- provide robust, independent data to support market development efforts.

As new bio-inoculants become available in the market place, there will need also to be a high level, up-to-date technical resource base to support their reliable application in the field. For example, market development of those organisms targeting cereal root diseases will require complementary technology to estimate the presence and risk of the important pathogens in paddock soils and also a knowledge base on best practice cereal root disease management to ensure that the application of the beneficial organisms can be well targeted to best realise their potential benefits.

The DNA-based cereal root disease assays and the cereal root disease resource manual and training provide a valuable foundation on which to build the commercialisation of beneficial organisms targeting root diseases.

Reference

Australian Pesticides and Veterinary Medicines Authority 2000. Guidelines for the registration of biological agricultural products. APVMA, Canberra. http://www.apvma.gov.au/guidelines/bioagprod.shtml.

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Contact details

Robert Hannam
RJ Hannam & Co Pty Ltd
PO Box 587 Magill SA 5072
Phone 0407 606 383
Fax 08 8364 5005
Email rhannam@ozemail.com.au

Behaviour of *Penicillium* fungi in soils

Steven Wakelin, VVSR Gupta, Paul Harvey, Maarten Ryder
CSIRO Land and Water, Glen Osmond SA

Introduction

Penicillium is the name given to an important group of micro-fungi. Isolates of *Penicillium* fungi can be recognised by the production of a characteristic reproductive structure terminating with a '*penicillius*', Latin for 'little brush' (Figure 1). With over 200 recognised species and a ubiquitous distribution on land and in soil, the *Penicillium* are one of the largest groups of fungi and among the most common eukaryotic life forms on earth (Pitt 2000).

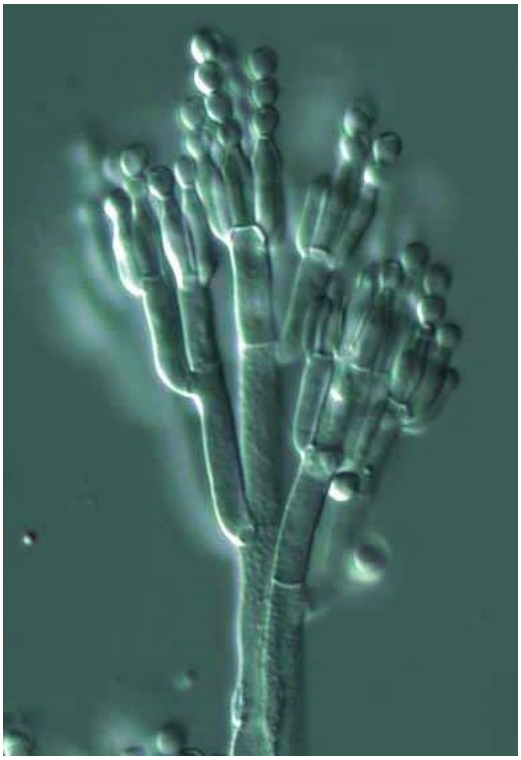

Figure 1. Penicillius of *Penicillium roquefortii*, a cheese-ripening fungus (courtesy of Ailsa Hocking, Food Science Australia, North Ryde NSW).

Penicillium fungi are familiar to most people as the blue and green moulds that occur on citrus (*P. digitatum* and *P. italicum*) and as the maturing agent of various cheeses and meats (eg *P.roqueforti* var. *roqueforti* and *P. camemberti*). Alexander Fleming made the genus famous through the discovery of the antibiotic penicillin from a culture of *Penicillium chrysogenum* (then called *P. notatum*) he observed inhibiting the growth of the pathogenic bacterium *Staphylococcus aureus*. We now know that many *Penicillium* species produce antibiotic compounds and also a wide range of other biologically-active metabolites. These compounds include toxins (mycotoxins) which can cause considerable disease in animals and humans. Accordingly, great care needs to be taken when selecting strains for potential industrial or agricultural application.

Although we are most familiar with *Penicillium* in or on our foods, the vast majority of species inhabit soil and are not commonly encountered. In soils, *Penicillium* species are ubiquitously distributed and can be isolated with relative ease in the laboratory. Their

successful colonisation of virtually all soils is largely attributable to their undemanding nutritional requirements and their ability to grow over a range of temperatures, water potentials and physicochemical conditions. The ability of *Penicillium* to produce a wide arsenal of biologically-active secondary metabolites is also likely to be associated with their ability to capture and compete for resources in soil.

Activities of *Penicillium* species in soil

Saprophytic decomposition of organic materials

In the soil ecosystem, nearly all *Penicillium* species are regarded as ubiquitous, opportunistic saprophytes. As such, they receive their nutrition through the decomposition of (mostly) plant material in the soil and play an important role in the fundamental process of nutrient cycling. To appreciate the importance of this, one must only consider the quantity of crop residues such as stubble, leaf trash, and root material that are decomposed into soil annually. Ultimately the nutrients contained in these materials, such as carbon, nitrogen and phosphorus, are recycled to the soil ecosystem increasing fertility.

Mobilisation of inorganic minerals

In addition to their important role in the recycling of organic material in soil, the *Penicillium* fungi are one of the relatively few groups of microflora capable of primary weathering of soil rock and minerals. The capacity for *Penicillium* species to solubilise (release) minerals from inorganic materials can be mostly credited to their ability to produce an arsenal of powerful organic acids. These acids increase mineral dissolution by reducing pH at discrete microsites of fungal activity.

However, they also function as powerful cation-complexing agents that can directly dissolve minerals and precipitates or can chelate with cations and release minerals and nutrients into solution. *Penicillium* species can degrade the surfaces of many rocks, including carbonate, marble and granite (Sterflinger 2000), serpentine (releasing silicon and magnesium), muscovite (releasing aluminium, potassium and silicon) (Crawford et al 2000), and basalt (Metha et al 1979). They are important in the bio-solubilisation of various forms of coal (Kitamura et al 1993, Polman et al 1994) and can solubilise a wide range of rock-phosphates (Whitelaw 2002).

Mineral weathering activity by *Penicillium* species (and other soil inhabitants) is an important primary step in the formation of soil and transformation of soil structure. Furthermore, the release into the soil ecosystem of nutrients that were previously biologically unavailable is a fundamental process in the development and maintenance of soil fertility. This is particularly significant for ‘poorer’ soils and in ecosystems developing on primary mineral substrates.

Agents of soil-borne plant disease

Like many saprophytic fungi, *Penicillium* species can be weakly parasitic to crop plants under certain conditions. Surprisingly, however, only a few have become parasitic to actively growing plant tissue. Most notably, *P. gladioli* causes a rot of the corms of *Gladiolus* and related species. Although many *Penicillium* species may be commonly isolated from diseased plant tissue, their infestation is usually regarded as secondary to a principal infecting agent.

Utilising *Penicillium* to increase plant growth

Biological control agents

Penicillium species are often investigated for the biological control of a range of soil phytopathogenic fungi. Research so far has shown strains of *Penicillium* to have biocontrol activity against *Phytophthora* root rot of azalea and orange (Fang, Tsao 1995), damping-off of chickpea and cucumber (Kaiser, Hannan 1984, Carisse et al 2003), Fusarium wilt of tomato (De Cal et al 1995), and various root rots of pea and bean (Kommedahl, Windels 1978, Windels 1981). Disease suppression by *Penicillium* species could occur by a variety of mechanisms: direct pathogen inhibition (antibiotic production), competition with pathogens for energy in the soil (saprophytic competition) or for infection sites on the root, or by inducing resistance in the plant.

In Australia, Dewan and Sivasithamparam (1988) investigated the potential for *Penicillium* species to control the take-all disease of wheat. The authors concluded that 'although… certain species of *Penicillium* are capable of providing some protection of wheat and ryegrass from the take-all fungus, the toxic effects produced by these fungi to wheat seedlings certainly outweigh the benefits of disease reduction'. Therefore, although isolates of *Penicillium* did exhibit biocontrol activity, the metabolites produced by these fungi were toxic to plants. This work clearly demonstrates the importance of carefully selecting isolates of *Penicillium* species for use as seed inoculants, as the side effects of inoculation may outweigh the benefits. Despite a substantial amount of research effort, there are currently no commercially-available biocontrol products based on *Penicillium* fungi.

Releasing soil phosphate for plant uptake

Penicillium fungi are a key group of soil microflora involved in phosphorus cycling (recently reviewed by Whitelaw 2000). Certain species of *Penicillium* species have also been shown to be intimately associated with the roots of crop plants (Wakelin et al 2004). In this microhabitat, expression of phosphorus-solubilising activity by *Penicillium* species has the potential to influence phosphorus nutrition of plants. The potential use of such fungi as phosphorus-solubilising inoculants has been demonstrated by the successful commercial release of *P. bilaiae* (JumpStart™, Philom Bios Inc, Saskatoon, Canada) and *P. radicum* (PR70RELEASE™, Bio-Care Technology Pty Ltd, Somersby, Australia).

General plant growth promoters

Solubilisation of soil phosphorus minerals by *P. radicum* can explain, at most, only part of the plant growth promotion (PGP) effect observed in the field (Whitelaw et al 1997). When investigating other possible mechanisms of plant growth promotion, Anstis (2004) detected in vitro production of precursors of plant hormone by *P. radicum*. In the rhizosphere, the microbial production of phytohormones has been shown to stimulate root branching (Patten, Glick 2002), resulting in significant increases in plant growth. Similarly, the phosphate-solubilising fungus *P. bilaiae* has also been found to increase production of root hairs when inoculated onto pea (Gulden, Vessey 2000). By increasing the root area, these *Penicillium* inoculants could enable plants to explore more of the soil and the nutrients therein. Although this form of plant growth promotion does not make soil nutrients more available to a plant (eg it is not phosphorus solubilisation or nitrogen fixation), it may nevertheless be an important mechanism through which crop production can be increased.

Co-inoculation with Rhizobium

In addition to stimulating root hair production, *P. bilaiae* has also been shown to increase nodulation and nitrogen uptake of pea and lentil (Gleddie 1993). The formation of nodules on leguminous plant roots begins with the infection of root hairs by *Rhizobium* bacteria. When inoculated onto seed, nodulation of a plant is often limited by root hair availability immediately below seed. Accordingly, the stimulation of root hair production in this region containing high quantities of *Rhizobium* bacteria may be a mechanism by which *Penicillium* fungi can increase nodulation and legume nitrogen fixation.

Strains of *Penicillium* can be easily formulated for co-inoculation with *Rhizobium* in peat-based carriers (Rice et al 1994), and co-inoculation of legumes using this technology is extensively used in Nth America. Peat-based legume inoculants containing the phosphorus-solubilising fungus *P. bilaiae* (marketed as TagTeam™, Philom Bios Inc) are available for a wide range of legume crops.

Compatibility with seed fungicides/pickles

The compatibility of biological inoculants with chemical seed dressings would appear to be a critical issue affecting their use. Many seeds, particularly grains, are commonly treated with various fungicides for the control of smuts, bunts, damping-off and associated seedling diseases. Fortunately, *Penicillium* species appear to be relatively insensitive to many of these classes of chemicals. Furthermore, it is a relatively simple process to test the compatibility of new chemicals as they become available, and make recommendations based on this information. The Australian *Penicillium* inoculant, *P. radicum*, is cited as being compatible with Premis®, Real®, Vitaflo®, Vitaflo C®, Raxil® and Jockey® provided the pickle is dry prior to applying the fungus (Bio-Care Technology Pty Ltd).

Current and future prospects for *Penicillium* in Australia

Penicillium fungi have much to offer us, in terms of disease control, nutrient mobilisation, increasing the efficacy of current *Rhizobium* products, and as general plant growth promoters. Presently, however, there is only one *Penicillium*-based product registered in Australia: PR70RELEASE™ (Bio-Care Technology), a formulation of the fungus *P. radicum*.

Penicillium radicum

PR70RELEASE™ is marketed as a phosphate-solubilising inoculant for broadacre grain crops (particularly wheat). The formulation contains dry spores of the fungus and is mixed with a liquid wetting agent (containing a sticker and food base for the fungus) prior to treating seed. *P. radicum* can solubilise a range of phosphorus-containing minerals in laboratory conditions (Whitelaw et al 1999, Figure 2) and stimulate wheat growth in the field (Whitelaw et al 1997). However, the actual contribution of phosphorus-solubilisation towards promoting plant growth is questionable, as it appears likely that *P. radicum* stimulates plant growth via a number of different mechanisms. Further research establishing the relative contribution of each mechanism to plant growth promotion is needed, as it will allow growers to target soil types and better predict inoculation response.

Penicillium bilaiae

Penicillium bilaiae (alternatively called *P. bilaii* or *P. bilaji*) is widely used in Nth America as a phosphorus-solubilising inoculant for a range of crops (JumpStart™,

Philom Bios Inc). The fungus, originally from Canada, underwent some limited testing in Australia but failed to show any benefit. Given its success overseas, it is possible that the lack of efficacy for this isolate in Australia could be due to its evolution under different agro-ecological conditions.

An extensive survey of root-associated *Penicillium* with phosphate-solubilising activity was recently conducted in Australia (Wakelin et al 2004). After screening over 800 isolates, the fungus with strongest phosphorus-solubilising activity was identified as a strain of *P. bilaiae* (Figure 2). This isolate was tested in glasshouse trials and field trials for plant growth promotion, and has shown significant benefits on crop growth over a variety of legume species. Given that the potential for commercial development of *P. bilaiae* has been well established overseas, the discovery of a locally-adapted strain of *P. bilaiae* has important implications for Australian industry and growers.

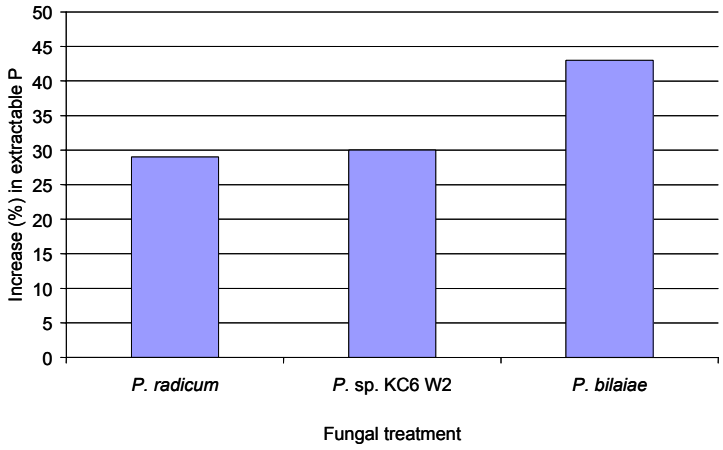

Figure 2. Effect of addition of fungal-colonised rye-grass seeds to Tarlee soil on HCO_3-extractable phosphorus (microcosm incubation experiment). Percentage increases in extractable phosphorus with respect to the non-colonised rye-grass control.

New phosphorus-solubilising Penicillium *species*

In addition to discovering a strain of *P. bilaiae* with strong phosphorus-solubilising activity, a potentially new species of *Penicillium* with plant growth promotant activity (isolate KC6W2) was unearthed (Wakelin et al 2004). In glasshouse conditions, *P.* sp. KC6W2 has been shown to increase the growth of wheat, lentil and medic (unpublished data), and increase the amount of extractable phosphorus following soil incubation (Figure 2). The commercial use of this fungus to increase the efficiency of phosphate fertilisers, release phosphorus 'locked' in soil and increase plant growth and yield warrants investigation.

Biological-control isolates

There are currently no *Penicillium* species registered for control of soil-borne plant diseases. Part of the reason may be due to stringent regulatory requirements. Disease control agents must be registered as pesticides and are consequently subject to tighter registration than organisms used as 'biological fertilisers'. Nevertheless, there appears to be a strong interest in the development of *Penicillium* as biocontrol agents.

The potential use for *Penicillium* fungi to increase crop health and yields and boost the efficiency of fertiliser inputs is almost completely unexploited. As agricultural production in Australia is increasingly expected to adhere to accredited environmental

management systems, the use of new technologies must be investigated. The harnessing of soil biological activity through seed inoculation with specific biological treatments warrants further investigation.

Acknowledgements

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References

Anstis ST 2004. *Penicillium radicum*: Studies on the mechanisms of growth promotion in wheat. PhD thesis, School of Earth and Environmental Sciences, University of Adelaide, Australia.

Carisse O, Bernier J, Benhamou N 2003. Selection of biological agents from composts for control of damping-off of cucumber caused by *Pythium ultimum*. Canadian Journal of Plant Pathology 25 258-267.

Crawford RH, Floyd M, Li CY 2000. Degradation of serpentine and muscovite rock minerals and immobilisation of cations by soil *Penicillium* species Phyton-Annales Rei Botanicae 40 315-321.

De Cal A, Pascual S, Larena I, Melgarejo P 1995. Biological control of *Fusarium oxysporum f.sp. lycopersici*. Plant Pathology 44 909-917.

Dewan MM, Sivasithamparam K (1998) Occurrence of species of *Aspergillus* and *Penicillium* in roots of wheat and ryegrass and their effect on root rot caused by *Gaeumannomyces graminis* var. *tritici*. Australian Journal of Botany 36 701-710.

Fang JG, Tsao PH 1995. Efficacy of *Penicillium funiculosum* as a biological control agent against *Phytophthora* root rots of azalea and citrus. Phytopathology 85 871-878.

Gleddie SC 1993. Response of pea and lentil to inoculation with the phosphate-solubilising fungus *Penicillium bilaii* (PROVIDE™). In Proceedings soil and crops workshop Saskatoon, Canada pp 47-52.

Gulden RH, Vessey JK .2000. *Penicillium bilaii* inoculation increases root-hair production in field pea. Canadian Journal of Plant Science 80 801-804.

Kaiser WJ, Hannan RM 1984. Biological control of seed rot and pre-emergence damping-off of chickpea with *Penicillium oxalicum*. Plant Disease 68 806-811.

Kitamura K, Ohmura N, Saiki H 1993. Isolation of coal-solubilising microorganisms and utilisation of the solubilised product. Applied Biochemistry & Biotechnology
38 1-13.

Kommedahl T, Windels CE 1978. Evaluation of biological seed treatment for controlling root diseases of pea. Phytopathology 68 1087-1095.

Metha AP, Torma AE, Murr LE 1979. Effect of environmental factors on the efficiency of biodegradation of basalt rock by fungi. Biotechnology & Bioengineering 21 875-885.

Patten CL, Glick BR 2002. Role of *Pseudomonas putida* indoleacetic acid in development of the host plant root system. Applied & Environmental Microbiology
68 3795-3801.

Polman JK, Stoner DL, Delezene-Briggs KM 1994. Bioconversion of coal, lignin, and dimethoxybenzyl alcohol by *Penicillium citrinum*. Journal of Industrial Microbiology 13 292-299.

Pitt JI 2000. A laboratory guide to common *Penicillium* species. 3rd edn. Food Science Australia, North Ryde NSW.

Rice WA, Olsen PE, Leggett ME 1994. Co-culture of *Rhizobium meliloti* and a phosphorus-solubilising fungus (*Penicillium bilaii*) in sterile peat. Soil Biology & Biochemistry 27 703-705.

Sterflinger K 2000. Fungi as geologic agents. Geomicrobiology Journal 17 97-124.

Wakelin SA, Warren RA, Harvey PR, Ryder MH 2004. Phosphate solubilisation by *Penicillium* species closely associated with wheat roots. Biology & Fertility of Soils
40 36-43.

Whitelaw MA 2000. Growth promotion of plants inoculated with phosphate-solubilising fungi. Advances in Agronomy 69 99-151.

Whitelaw MA, Harden TJ, Bender GL 1997. Plant growth promotion of wheat inoculated with *Penicillium radicum* sp. *nov.* Australian Journal of Soil Research
35 291-300.

Whitelaw MA, Harden TJ, Helyar KR 1999. Phosphate solubilisation in solution culture by the soil fungus *Penicillium radicum*. Soil Biology & Biochemistry 31 655-665.

Windels CE (1981) Growth of *Penicillium oxalicum* as a biological seed treatment on pea seed in soil. Phytopathology 71 929-933.

Contact details

Dr Steven Wakelin
Research Scientist
Microbiology and Plant Pathology
CSIRO Land and Water
PMB 2 Glen Osmond SA 5064
Phone 08 8303 8708
Fax 08 8303 8684
Email Steven.Wakelin@csiro.au

Delivery of soil biology services to Australian agriculture

Kathy Ophel Keller, Alan McKay, John Heap, Steve Barnett
South Australian Research and Development Institute (SARDI)

Introduction

Soilborne diseases are major drivers of crop rotation. Knowledge of disease risk at the rotation planning stage improves rotation and management decisions. This is the rationale behind the delivery of DNA-based assays which quantify soil-borne diseases. This service and an associated agronomist training course in management of root diseases were developed by Australian researchers at SARDI (South Australian Research and Development Institute) and CSIRO, and are available to Australian growers via Bayer Crop Science.

Services in the future that may be linked to this testing service are tests for soil health, sampling and disease management by zoning, based on precision agriculture technologies, and the training of consulting agronomists in the interpretation of these new technologies.

Root Disease Testing Service

In 1997 the launch of the Root Disease Testing Service by SARDI marked a world first in the delivery of a soil-based test able to identify and quantify a range of root pathogens. Now nine soil-borne diseases can be detected from a single soil sample. The amount of DNA present of target pathogens is measured and the potential risk of each disease is indicated.

The technology used in the service was developed in research and development projects over more than 10 years in three organisations - SARDI, CSIRO Entomology and the CRC for Soil and Land Management. Over this time research funding was received from Bayer Crop Science (and its predecessors Aventis Crop Science and Rhone-Poulenc), SAGIT (South Australian Grains Industry Trust Fund), RIRDC and GRDC.

There has been an enormous amount of technology evolution over this period. The current technology, developed jointly by SARDI and CSIRO Entomology, is based on DNA extraction which is robust from a range of soil types, and PCR technology which allows quantitative assessment of a range of nematode and fungal pathogens.

The development of commercial molecular diagnostics has involved a number of steps:

- prioritisation of target pathogens
- DNA sequencing of pathogens and related genera
- development of DNA sequences specific to the pathogens
- optimised DNA extraction from a broad range of soil types
- development of quantitative DNA tests
- calibration of DNA assay result to disease development using spiked samples and field samples
- development of sampling strategies
- investment in laboratory infrastructure to deliver throughput required

- delivery via commercial partner and agronomy network
- development of interpretative tools.

Current service

The current service (2003/04) delivers tests for take-all (*Gauemannomyces graminis* var. *tritici* and *G.g.* var. *avenae*), rhizoctonia (*Rhizoctonia solani* AG-8), root lesion nematodes (*Pratylenchus neglectus, P. thornei*,) CCN (*Heterodera avenae*), blackspot (*Mycosphaerella pinodes* and *Phoma medicagenis* var. *pinodella*) of peas, and crown rot disease (*Fusarium pseudograminearum* and *F. culmorum*).

The core technology has been extended in a research context to horticulture with DNA-based soil tests for root knot nematode and *Fusarium oxysporum* subsp. *lycopersici*, to detection of small-seeded weeds such as branched broomrape, and to monitoring aquatic sediments for organisms which are indicative of environmental impact.

Sampling strategies to detect root diseases

A critical issue in the success of disease prediction is the impact of soil sampling strategy on the test results. Across a paddock, there is more variation in disease than there is with soil factors such as pH or nutrients. The research of John Heap, funded by GRDC and SARDI, shows that where and how soil samples are gathered impacts on the accuracy of the final result.

Production zones for precision agriculture are created by overlaying data sets such as yield, electro-magnetic maps and satellite data. Initial research has demonstrated that there is often a relationship between these production zones and the level of soil-borne disease inoculum present. This means that sampling within paddock zones will help reduce the averaging affect of sampling across a whole paddock. This will allow growers to identify which parts of the paddock are at risk from various diseases and allow them to focus their disease management where it will provide the greatest return from their inputs.

Work on sampling has demonstrated that the size of the soil core collected (8-25 mm) at each sampling point is not crucial. However, mixing soil samples in the field and then sub-sampling for analysis reduced the accuracy of test results by 15 to 30%. Test accuracy increases if 30 to 50 soil cores are collected.

These results have now been implemented, in collaboration with Spurr Soil Probes, as the AccuCore sampling system. This system uses a 10 mm soil core and collects a total of 45 samples from across a paddock or paddock zone. Three samples are taken within the stubble row at 15 locations across the area and the total soil sample is analysed, eliminating the errors introduced by sub-sampling.

Understanding microbiology of suppressive soils

Long-term field trials at Avon SA, supported by CSIRO and GRDC, have demonstrated the benefits of long-term stubble retention. The trial began in 1979 and exhibits some highly desirable features, including suppression of soil-borne diseases, increased free-living nitrogen fixation and increased phosphate use.

SARDI researcher Steve Barnett, supported by GRDC funding, leads a project to identify microorganisms involved with disease suppression. He has identified three groups of soil bacteria: *Pantoea, Exiguobacterium* and *Microbacterium*, which suppress

not only rhizoctonia, but also take-all and Fusarium crown rot. These bacteria do not act directly in reducing pathogen levels in the soil, but reduce disease by a combination of reducing the amount of root infection and increasing the growth of infected plants. Other soil organisms, such as *Streptomyces, Trichoderma*, and fungal-feeding nematodes appear to be important in the suppression of these diseases at Avon.

This work will give us to a better understanding of which microorganisms are important for disease suppression. This may lead to development of tests for beneficial organisms to monitor or predict suppression, and management strategies to promote suppression. Also, the beneficial bacteria and fungi isolated from suppressive soils may be useful as inoculants to reduce the impact of disease.

Delivery of outcomes to industry

Soil biology outcomes are delivered from SARDI and collaborative research via a number of methods:

- commercialisation of the diagnostic services via the agricultural reseller network
- training of private, public and commercial agromonists via an accredited training program on root disease management
- manufacture and retailing of soil sampling tools via a commercial partner
- development of potential inoculants arising from fundamental work on disease suppression.

Predicta B™

The service was initially delivered by SARDI as the Root Disease Testing Service (RDTS) from 1998-1999. In November 2000 the commercial delivery of the core diagnostic technology was licensed to Aventis Crop Science (now Bayer Crop Science). A subsidiary company, C-Qentec Diagnostics, markets the service branded commercially as Predicta B™ in Australia.

Soil test kits are available from rural merchandise resellers. Test results are usually available within two weeks, and are returned to farmers via a network of trained agronomists who interpret the test report and assist in development of management strategies for the priority diseases.

Tests for new pathogens are being developed, including common root rot and stem nematode. In the future, tests for soil organisms which suppress disease may be added to the suite of results provided.

Agronomist training program

Agronomic staff from rural merchandise companies, as well as private and public consultant agronomists, are accredited by SARDI in a one-day course designed to teach fundamental principles of root disease management. To date this course has accredited 800 agronomists nationally to deliver the tests and increase agronomists' knowledge of root disease management. This training course can be expanded to include delivery of broader information on soil health.

AccuCoreTM

An optimised sampling tool, AccuCoreTM, was developed as an outcome of GRDC research and is manufactured under license from SARDI and retailed by Spurr Soil Probes. This product is marketed via field days and expos, and promoted at agronomy training courses.

Future possibilities

The development of predictive root disease tests by SARDI and CSIRO marked an important break through in root disease management. Now this service, and the training associated with it, is being expanded to provide better information and tools for sampling and integration with precision agriculture technologies. In the future, current fundamental research on disease suppression may lead to monitoring tools, management strategies and associated training to promote soil health more broadly.

Contact details

Dr Kathy Ophel Keller
Principal Research Scientist and leader, crop pathology
SARDI Waite Campus Glen Osmond SA 5064
Phone 08 8303 9368
Fax 08 8303 9393
Mobile 041 881 8657
Email Ophelkeller.Kathy@saugov.sa.gov.au

Managing soil-borne and stubble-borne cereal pathogens in the northern grains belt

Steven Simpfendorfer[A], John Kirkegaard[B], John Holland[A], Andrew Verrell[A], Rod Bambach[A], Kevin Moore[A]
[A]NSW Department of Primary Industries, Tamworth
[B]CSIRO Plant Industry, Canberra

Introduction

Winter cereal plants are challenged by a range of soil-borne and stubble-borne pathogens in the northern cropping zone which encompasses northern NSW and southern Queensland. These include fungal and nematode (*Pratylenchus thornei* and *P. neglectus*) pathogens of roots and crowns plus stubble-borne pathogens such as *Pyrenophora tritici-repentis* (yellow spot) and *Gibberella zeae* (Fusarium head blight) which infect above-ground portions of wheat plants. Disease management work at Tamworth is mainly focused on two fungal diseases, crown rot and common root rot. Crown rot caused by *Fusarium pseudograminearum* (*Fp*) is a major constraint to winter cereal production in Australia. Although it is generally more common in the northern cropping belt, it can occur throughout all mainland cereal growing areas and is estimated to cost the Australian grains industry $56 million per annum (Brennan, Murray 1998). Infection of winter cereals can occur through the crown, sub-crown internode, basal internode and/or lower leaf sheaths. This can occur at any growth stage from seedling emergence through to maturity (Purss 1969). Crown rot infection is characterised by a light honey-brown to dark brown discolouration of the base of infected tillers. The fungus survives in cereal and grass weed residues, while yield loss from the production of whiteheads is related to moisture stress post-flowering (Burgess et al 2001). Common root rot, caused by the fungus *Bipolaris sorokiniana* (*Bs*), is often found in association with crown rot and has been estimated alone to cost Australian growers $22 million per annum (Brennan, Murray 1998). Symptoms are a dark brown to black discolouration of whole or part of the sub-crown internode. Severely affected plants are stunted, have fewer tillers and produce smaller heads. Rotation to non-host break crops is essential to the successful management of both of these diseases.

Rotation to non-host pulse (chickpea, faba bean) oilseed (canola, mustard) or summer crops (sorghum, sunflower, mungbean, cotton) essentially reduces crown rot inoculum levels by starving the fungus of a suitable host which allows natural decline of cereal residues that harbour the pathogen to occur. The length of rotation needed to be effective in managing crown rot depends on the rate of decomposition of the infested residues (Summerell, Burgess 1989). Felton et al (1998) have demonstrated that chickpeas are effective in reducing the levels of crown rot when grown in rotation with wheat. However, the acreage of canola has expanded in the northern region as more adapted, shorter-season varieties are developed for the area. Canola-quality mustard is also being developed as a potential break crop for the area. Extensive research has been conducted on the possible impact of isothiocyanates (ITCs) released from brassica roots on the suppression of disease inoculum in a process termed ‘biofumigation’ (eg Angus et al 1994). *Fp* is also sensitive to the 2 phenylethyl (2PE) ITC, the principal ITC released by canola roots (Smith, Kirkegaard 2002). However, there have been no field studies investigating the potential for enhanced suppression of *Fp* as a result of

biofumigation by brassica crops or a general assessment of their effectiveness as break crops for crown rot in the northern cropping zone.

A series of three-year field experiments were conducted to investigate the impact of previous crops on the levels of *Fp* inoculum, the incidence and severity of disease, and the yield and quality of following durum and bread wheat crops. The experiments were especially interested in comparing the break crop benefits of brassicas with those of chickpea which is currently the most widely grown winter rotational crop in the region. A further experiment was conducted to look specifically at the effect of altering the carbon and nitrogen content on wheat stubble on the extent of residue breakdown. Treatments were applied as liquid spray amendments with their effect on stubble breakdown compared under a chickpea crop versus a fallow period.

Materials and methods

Rotation experiments

The effect of previous crop species (oilseed, legume and cereal) on the incidence and severity of crown rot and yield of wheat was investigated in two three-year no-till field experiments at Tamworth in northern NSW. The experiments were designed to compare the effectiveness of the brassica break crops canola and mustard with chickpea on the reduction of crown rot in subsequent wheat crops (Table 1).

Table 1. Rotation treatments used in the second year of experiments

Treatment	Description[A]	Tamworth1 2000	Tamworth2 2001
Mustard	Two root GSLs	99Y-1-1	99Y-1-1
Canola(H)	High root GSL	Mystic	Mystic
Canola(M)	Mod root GSL	Oscar	Oscar
Canola(L)	Low root GSL	Monty	Monty
Chickpea	Non-host	Gully	Gully
Wheat(T) (bread)	Host (6)	Sunco, Mulgara	Sunco
Wheat(S) (durum)	Host (1)	Wollaroi	Wollaroi
Barley	Host (1)	Grimmett	Grimmett

A Numbers in parenthesis represent crown rot resistance rating (1=poor, 9=good)
Glucosinolate levels (μmole/g freeze-dried root tissue) from Kirkegaard and Sarwar (1999) 2PE-GSL in canola cultivars: Monty 4.0, Oscar 10.0, Karoo 26.0, Mystic 23.5. Indian mustard cultivars: 99Y-1-1 PE-GSL 5.5, PR-GSL 3.0. (S) Susceptible; (T) Tolerant.

The experiments were further designed to examine if biofumigation was associated with improved effectiveness of break crops by including three canola varieties with varying levels of glucosinolates (GSL) in their roots and a mustard which contains two root GSLs. Responses to previous broadleaf and cereal crops were investigated in the third year in a crown rot-tolerant bread wheat (cv Sunco) and crown rot-susceptible durum wheat (cv Yallaroi). Disease severity assessments were based on visual symptoms while the incidence of infection by *Fp, Bs* and *Trichoderma* spp. was based on the culturing of surface-sterilised wheat crowns/sub-crown internodes onto ¼ PDA + 100 mg/L novobiocin. Cultures were incubated at 25°C under a combination of white and near-ultraviolet lights with a 12 hour photoperiod for seven days. Whiteheads were counted in 4 x 1m sections of row selected at random in each plot two or three times following their first appearance during the grain-filling stage. Whiteheads were expressed as a % of total head numbers in each treatment.

Stubble amendment experiment

The experiment was established in standing durum (cv Bellaroi) stubble grown in 2002. Main plots consisted of either a chickpea (cv FLIP 94-508C) crop or fallow through the 2003 growing season. Plots were split for stubble amendment treatments of nil (water), 5% sugar (as Coopers brewing sugar), 5% nitrogen (as urea @ 46% N) or combined 5% sugar + 5% nitrogen (S + N). There were four replicate plots of each treatment with the various stubble amendment solutions sprayed directly onto the residue at a rate of 350 mL per 10 × 2 m plot. The solutions were applied immediately after sowing of the chickpea crop and then on a fortnightly basis until full canopy closure of the chickpea crop (total of seven applications). Biomass of the remaining cereal stubble was assessed after harvest from two 0.33 m^2 quadrats cut from within each plot which were then dried at 80°C for 48 hours.

Results

All break crops (brassicas and chickpea) significantly reduced the severity of crown rot in both a susceptible (37-47% reduction) and tolerant wheat crop (21-51% reduction) compared with growing wheat on wheat or wheat on barley (Figure 1). Brassica crops were generally more effective than chickpea in reducing the severity of crown rot in the crown rot-susceptible durum wheat (by 23-26%) but the advantage was less evident in cv Sunco which has partial resistance to crown rot. The three canola varieties and mustard, which varied in their levels of root GSLs, did not significantly differ in their effect on crown rot severity levels in following wheat crops. Hence, there was no evidence to suggest that biofumigation associated with higher levels of ITCs released by brassicas reduced *Fp* inoculum levels and the severity of crown rot in following wheat crops (Figure 1).

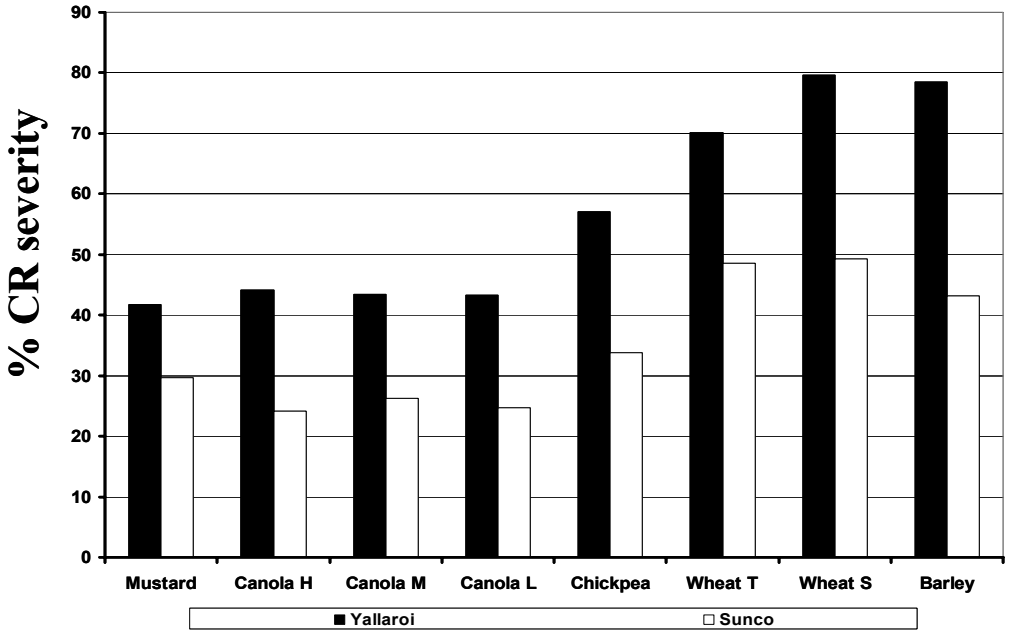

Figure 1. Effect of rotation on severity of crown rot infection (LSD 0.05=12.5).

The severity of crown rot infection following each rotation crop did not directly relate to the expression of whiteheads. Rotating to a non-host brassica or chickpea crop certainly resulted in a significant reduction in the formation of whiteheads compared with a cereal-wheat rotation (Figure 2). However, there was no difference in the

expression of whiteheads following the various break crops even though crown rot severity was significantly higher following chickpeas compared with the brassica crops.

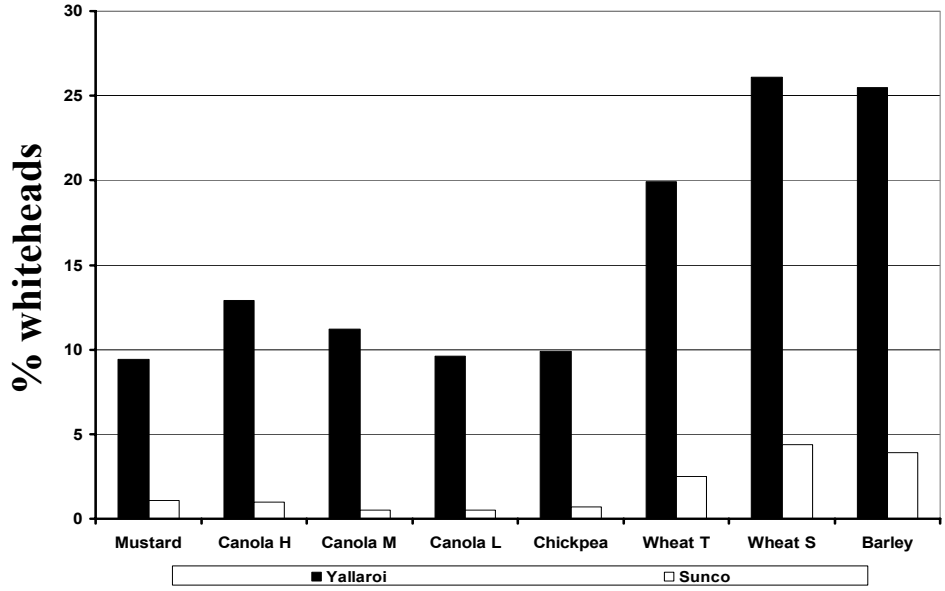

Figure 2. Effect of rotation on the formation of whiteheads (LSD 0.05=3.5).

Levels of *Trichoderma* spp. isolated from the crowns at harvest were consistently higher after brassicas than after chickpea or cereals in both the susceptible and tolerant wheat variety (Figure 3).

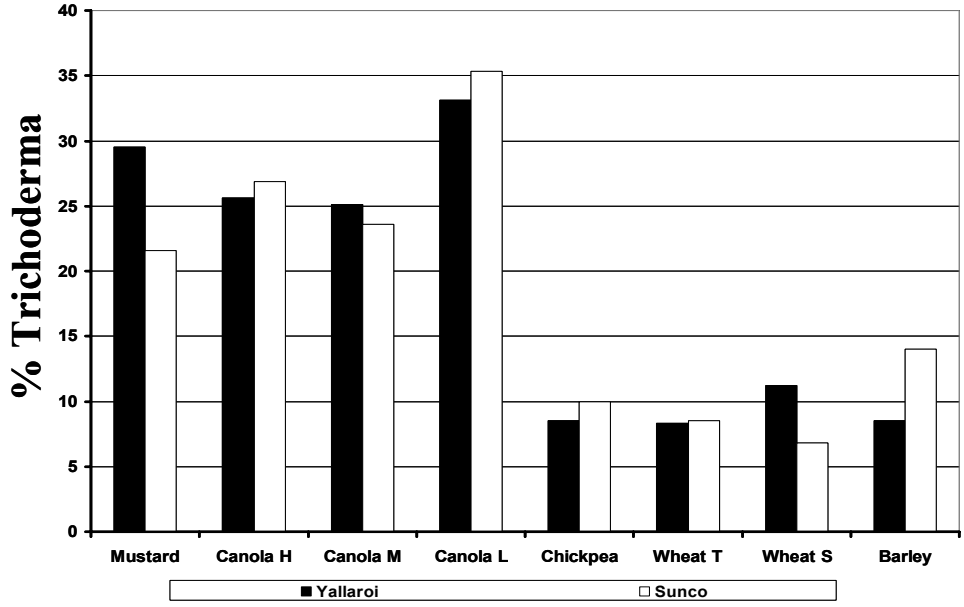

Figure 3. Effect of rotation on the *Trichoderma* spp (LSD 0.05=11.2).

The treatment of cereal stubble with solutions of sugar or nitrogen significantly increased the breakdown of residue compared with the untreated control (19-27% reduction, Figure 4). However, there was no difference between the effectiveness of these two amendments when applied separately. The combined treatment of sugar and nitrogen was the most effective treatment, reducing stubble loads by around 44%

compared with the untreated control, and resulted in significantly greater breakdown of the cereal residue than applying either sugar or nitrogen alone. Within treatments there was no significant difference between residue breakdown under a chickpea cover crop versus a fallow.

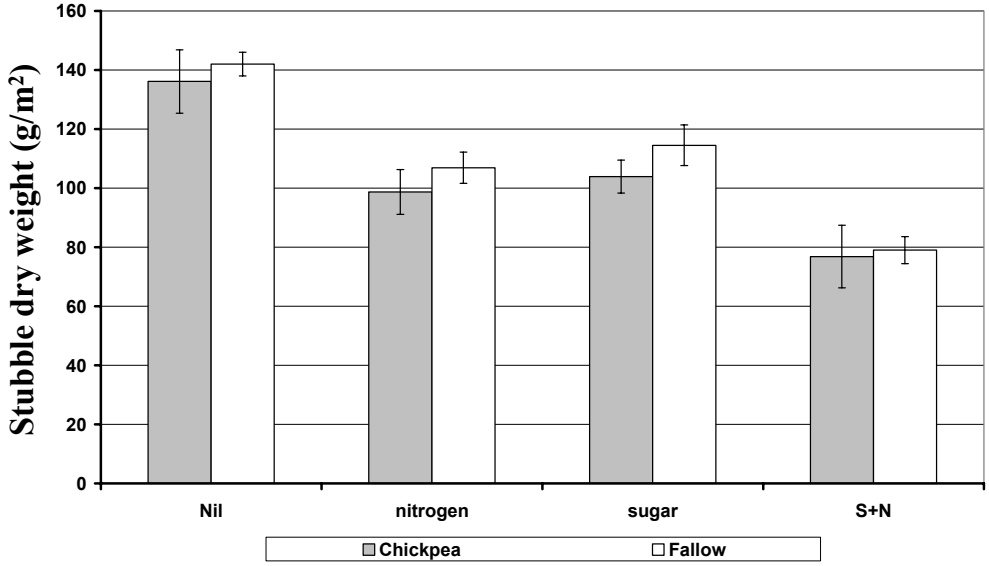

Figure 4. Effect of stubble amendments on the breakdown of durum wheat residue.

Discussion

Recent work conducted by Percy et al (2003) indicates that partially resistant varieties such as Sunco have a reduced rate of spread of the crown rot fungus through tissue than highly susceptible varieties such as the durum wheats. This is reflected in reduced severity (Figure 1) and whitehead formation (Figure 2) in the partially resistant variety (cv Sunco) compared with the susceptible variety (cv Yallaroi). This has generated debate as to whether this reduced rate of fungal spread carries through to a reduced build-up of crown rot inoculum by growing partially resistant varieties. In this study the severity of crown rot was slightly reduced (~10%) in a following susceptible variety by growing a tolerant wheat variety in the previous year compared with a susceptible wheat or barley crop (Figure 1). However, this difference was not evident when sowing a tolerant variety in the second year. The slight reduction in inoculum build-up obtained by growing partially resistant varieties does not appear sufficient to effectively control crown rot but may be useful in the integrated management of this disease.

This study demonstrated that barley is an extremely good host of *Fp* with high disease severity occurring in following wheat crops. All current barley varieties are susceptible to highly susceptible to crown rot but they do not tend to suffer extensive yield loss from this disease. However, yield losses of around 15-20% can occur in crown rot-infected barley crops in years conducive to disease expression (G. Wildermuth, personal communication). The earlier maturity of barley tends to assist it in avoiding moisture stress late in the season that exacerbates the formation of whiteheads in infected tillers, and the excellent tillering ability of barley allows for good yield compensation. However, growers need to be reminded to look for characteristic browning at the base of tillers infected with *Fp* to accurately determine their crown rot status rather than purely looking for the expression of whiteheads. This is even more important when

considering the role of barley in the rotation and its propensity to build-up crown rot inoculum levels.

The major finding from the rotation experiments was the occurrence of lower levels of crown rot severity following different classes of brassica break crops compared with chickpea. However, the greater crown rot severity following chickpeas by comparison with canola or mustard was not directly reflected in the formation of whiteheads. This could potentially relate to the different water use patterns of these break crops. Chickpeas tend to use less water during the season than brassicas and generally do not root as deep in the soil. Hence, wheat crops growing after chickpeas may experience reduced moisture stress through this water saving which decreases the production of whiteheads in infected tillers. This is a good reminder to growers of the need to think of crown rot in terms of both of its distinct phases of infection and yield loss through the formation of whiteheads. The formation of whiteheads is related to moisture stress post-flowering where *Fp* is believed to disrupt the vascular ('plumbing') system of the plant, preventing the movement of water from the soil into the heads. This results in the formation of a whitehead in infected tillers which have either shrivelled or no grain formation. Thus, under a wet finish, tillers can still be infected with *Fp* (ie still get inoculum build-up) but there is no moisture stress so the heads fill normally.

There was no evidence that the lower disease and higher yield following brassicas compared with chickpea was related to direct *Fp* inoculum suppression by biofumigation as no differences were evident between the three canola varieties. Suppression of *Fp* by biofumigation was unlikely because infection occurs largely from inoculum which survives predominately on above-ground residues. Soil-released ITCs would therefore be expected to have limited contact with the major inoculum source. Two possible reasons for the lower levels of crown rot infection following brassica break crops include more rapid breakdown of cereal residues under the brassicas, and microbial changes within the soil and residue more conducive to inoculum decline.

More rapid breakdown of cereal residues under the brassicas

The rate of residue breakdown is known to be directly related to microbial activity which depends on both moisture and temperature. Thus break crops with a denser canopy (eg brassicas, faba beans) are likely to provide a microclimate more conducive to the breakdown of cereal residues than crops such as chickpeas that generally have thinner canopies and do not close over until later in the season. This is evident in a further rotation trial at Tamworth where wheat stubble cover was measured following the various rotation crops prior to sowing in 2002. Percentage wheat stubble cover was greatest for wheat (85%), then chickpea (40%), canola (29%), faba beans (27%) and lowest following sorghum (15%). Sorghum is an excellent rotational crop for the management of crown rot as it allows for at least a two year break from susceptible winter cereals and enables grass weeds to be effectively controlled. Depending on row configuration, sorghum also facilitates good breakdown of winter cereal residue as it grows over summer when temperature and moisture are generally more conducive to microbial activity.

Microbial changes within the soil and residue more conducive to inoculum decline

The consistently higher levels of *Trichoderma* isolated from wheat following brassicas compared with that following chickpeas or cereals is an interesting observation from this study. *Trichoderma* are a group of fungi commonly associated with biological

control of pathogenic fungi. Recent work by CSIRO has shown that when grown in the laboratory *Trichoderma* are considerably less sensitive to the ITCs released by brassicas than other common soil/stubble borne wheat pathogens including *Fp* (Smith, Kirkegaard 2002). Work conducted by Wong et al (2002) under laboratory conditions has further shown that isolates of *Trichoderma harzianum* and *T. koningii* have the potential to significantly reduce the survival of *Fp* in wheat residue buried in soil. Survival of *Fp* in residue was reduced most after three months at 30°C in moist soil with displacement being considerably better in an acidic red duplex soil than an alkaline black soil. The role of *Trichoderma* spp. in the management of crown rot and their interaction with various rotational crops appears worthy of further investigation.

Irrespective of the mechanisms involved, these experiments demonstrate that canola and mustard provide an effective break crop for crown rot in northern NSW. Furthermore, brassicas would provide an excellent alternative rotation crop to chickpea in areas where adapted varieties are available as they appear to have an improved capacity to reduce the severity of crown rot in subsequent wheat crops.

Fp infection arises predominately from inoculum surviving on above-ground residues of previous cereal crops (Burgess et al 2001) so factors which influence the rate of stubble breakdown are likely to have a significant influence on the survival of *Fp* and the incidence of crown rot infection. The treatment of cereal stubble with a combined carbon (sugar) and nitrogen solution was shown to significantly increase the breakdown of the standing residue compared with an untreated control. Increasing microbial activity through supplementation of stubble with a readily useable carbon source combined with a more balanced carbon-to-nitrogen concentration may be useful in the integrated management of this stubble-borne disease.

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References

Angus JF, Gardner PA, Kirkegaard JA, Desmarchelier JM 1994. Biofumigation: Isothiocyanates released from Brassica roots inhibit the growth of the take-all fungus. Plant & Soil 162 107-112.

Brennan JP, Murray GM 1998. Economic importance of wheat diseases in Australia, NSW Agriculture, Wagga Wagga.

Burgess LW, Backhouse D, Summerell BA, Swan LJ 2001. Crown rot of wheat. In Fusarium: Paul E Nelson memorial symposium. Eds Summerell BA, Leslie JF, Backhouse D, Bryden WL, Burgess LW pp 271-294. American Phytopathological Society St Paul MN.

Felton WL, Marcellos H, Alston C, Martin RJ, Backhouse D, Burgess LW, Herridge DF 1998. Chickpea in wheat-based cropping systems of northern New South Wales. II. Influence on biomass, grain yield, and crown rot in the following wheat crop. Australian Journal of Agricultural Research 49 401- 408.

Kirkegaard JA, Sarwar M 1999. Glucosinolate profiles of Australian canola (*Brassica napus annua L.*) and Indian mustard (*Brassica juncea L.*) cultivars: Implications for biofumigation. Australian Journal of Agricultural Research 50 315-324.

Percy CD, Sutherland MW, Wildermuth GB 2003. Growth of *Fusarium pseudograminearum* in crowns of wheat differing in susceptibility to crown rot. In Abstracts, 9th International Fusarium workshop, Sydney p 29.

Purss GS 1966. Studies of varietal resistance to crown rot of wheat caused by *Fusarium graminearum* Schw. Queensland Journal of Agricultural and Animal Science 23 257-264.

Smith BJ, Kirkegaard JA 2002. In vitro inhibition of soil microorganisms by 2-phenylethyl isothiocyanate. Plant Pathology 51 585-593.

Summerell BA, Burgess LW 1989. Decomposition and chemical composition of cereal straw. Soil Biology & Biochemistry 21 551-559.

Wong PTW, Mead JA, Croft G 2002. Effect of temperature, moisture, soil type and *Trichoderma* species on the survival of *Fusarium pseudograminearum* in wheat straw. Australasian Plant Pathology 31 253-257.

Contact details

Dr Steven Simpfendorfer
Plant Pathologist Cereals
Tamworth Agricultural Institute
NSWDPI RMB 944 Calala Lane Tamworth NSW 2340
Phone 02 6763 1261
Fax 02 6763 1222
Email steven.simpfendorfer@dpi.nsw.gov.au

Enhancing beneficial root-zone processes by managing crop residue inputs

Darryl R. Nelson, Pauline M. Mele
Primary Industries Research Victoria, Rutherglen

Introduction

Cereal production is of great importance to the Australian economy, bringing in millions of dollars each year in export revenue. Improved management in Australian cereal production systems related to modified tillage practices, fertiliser regimes tailored to crop demand, legume rotations and improved plant varieties, have significantly increased production in wheat to over 1375 kg/ha on average per annum (Lawrie et al 2000). Despite this increase, cereal production is hampered by poor soil fertility and, specifically, subsoil constraints such as boron toxicity (Cartwright et al 1986) and aluminium toxicity (Marschner 1995, Hocking 2001).

Phytotoxic levels of aluminium in soils are commonly associated with acidic or acidifying soils that comprise about 90 million hectares of agricultural land in Australia (Scott et al 2001, Slattery et al 2001). Crop management on such soils involves the use of aluminium- tolerant cereals and the ameliorative strategy of liming (Scott et al 1999, Coventry et al 1997). Associated with low soil fertility is sub-optimal nutrient supply for cereal production systems. Although managed through fertiliser applications, long-term use of such strategies can be problematic. Legume residues are commonly used to increase soil nitrogen and general soil fertility and legume rotations have been associated with enhanced yields in subsequent cereal crops (Harris et al 2002, Lawrie et al 2000). Increases in soil fertility and plant yields have been attributed to soil microbial mineralisation processes (Pankhurst, Lynch 1994, Rowell 1994) such as nitrogen cycling (Barakah et al 1995, Vallini et al 1997). Effects of amendments on soil microorganisms have been largely measured in bulk soil away from the root zone, which is a critical site for exchange of nutrients between the plant and its surrounding soil environment. It is evident that different plants can influence the structure of microbial communities in the root zone (Wieland et al 2001), but it is not clear whether residue input influences microbially mediated processes in this critical nutrient exchange site.

Specific processes that are considered beneficial to plants include nitrogen fixation, both symbiotic (*Rhizobium* spp) and non-symbiotic (*Azospirillum* spp and *Beijerinckia* spp). Fixed nitrogen (in the form of ammonium) that is not utilised by plants immediately may be oxidised to nitrate via nitrification, which can in turn be reduced to nitrous oxide and atmospheric nitrogen, completing the nitrogen cycle. Traditionally, the measurement of nitrogen fixation involved acetylene reduction (Smith et al 1978). More recently, molecular techniques have been employed that target specific genes in the nitrogen cycle or the 16S rDNA of appropriate microorganisms. Functional genes are preferred because they can infer active populations carrying out relevant processes. For example, a section of the nitrogenase gene, *nif*H, is often targeted in analysis of nitrogen fixation (Lepo et al 1995, Widmer et al 1999, Poly et al 2001, Burke et al 2002).

In the present study, wheat rhizosphere soil was collected to investigate the functional properties of the microbial community in response to several common crops and lime.

Biolog® Microtiter plates that measure activity of soil microorganisms by utilisation of different carbon sources, were used to study the effect of lime and organic residues on microbial community structure. In addition, molecular techniques were used to investigate nitrogen cycling in the rhizosphere by targeting the *nif*H gene, essential for nitrogen fixation.

Materials and methods

Collection and preparation of crop residues and soil

All plant residues were collected from mature crops on the Rutherglen Research Farm. Wheat (*Triticum aestivum* cv Diamondbird), lupin (*Lupinus angustifolius* L cv Jindalee) and canola (*Brassica napus* cv Pinnacle) were collected prior to harvesting. The canola had been windrowed the previous day. Lucerne (*Medicago sativa* L cv Genesis) and pea (*Pisum sativum* cv Excel) were collected freshly cut prior to harvest. Residues were dried for 48 hours (40^{o}C) and milled through a 1.5 mm mesh. Soil, high in aluminium (~45 μM in solution) and low in pH (~4.3 in $CaCl_2$), was collected from a site in north-east Victoria, sieved (2 mm) and amended with the crop residues with and without lime (2.5 t/ha) to a constant value of 8.5 mg C/g (an approximate soil:amendment ratio of 50:1). The soil was contained in PVC pots (10.4 cm diameter and 30 cm depth) and soil moisture maintained at 75% field capacity with sterile water, applied weekly. Aluminium-tolerant *Triticum aestivum* var. Egret seeds, germinated in water agar (15 g/L), were planted (2 per pot, 10 mm depth) and plastic beads placed over the soil surface. The plants were monitored (Haun 1973) until the five-leaf physiological stage was reached, when the pots were opened and the plant roots exposed to 100 mm depth. Soil adhering to several seminal roots was collected using sterile forceps for DNA extraction (0.4 g) and Biolog® analysis (1 g). Plant shoots were collected for nitrogen determination.

Rhizosphere microbial community analysis using Biolog® Microtiter Ecoplates

Rhizosphere soil (1g) was serial diluted to 10^{-4} in saline solution and inoculated into Biolog® Microtiter Ecoplates. The plates were incubated in the dark (25^{o}C) for five days and the absorbance measured.

*Analysis of nitrogen fixing (*nif*H) Rhizosphere Gene Pools*

DNA was extracted using a Bio101 DNA Extraction Kit (Qiagen GmbH Germany) using a modified protocol (Yeates, Gillings 1998), and the functional gene *nif*H was amplified with the Polymerase Chain Reaction (PCR) using previously published primers (Poly et al 2001). The reverse primer was modified to include a 5' terminal phosphate group. Purified PCR products were prepared for Single Strand Conformation Polymorphism (SSCP) (Schwieger, Tebbe 1998) on a 0.6x polyacrylamide gel and stained using SYBR Gold.

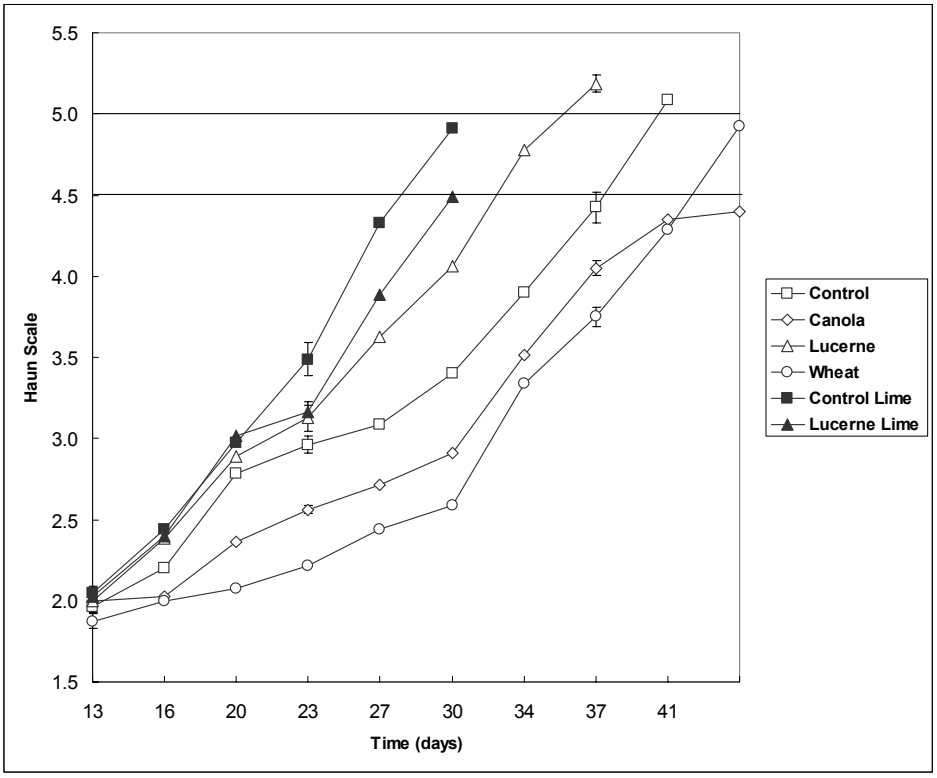
Results

Plant development and tissue analysis

Thirteen days after sowing, no significant differences in plant development were observed using the Haun scale. From 16 days onwards, both residue amendment and lime significantly affected plant growth (in most cases $P<0.001$), while the interaction between the two was also significant except for 20 days after sowing (Figure 1). Shoot nitrogen concentration was significantly lower in plants grown in soil with wheat and canola, compared with most other treatments. In plants grown in limed soil, nitrogen content was not significantly higher than the corresponding non-limed treatments in all

cases except for canola and wheat, where it increased. The amount of nitrogen in plant residues and the amount in wheat tissue at the end of the experiment correlated well in the non-limed treatments (R^2=0.88) but not in the limed treatments (R^2=0.61). However, in controls, nitrogen levels were generally higher than all treatments except lucerne and lucerne/lime.

Fig.1. Plant development measured using the Haun scale. Not all samples are displayed. Other treatments fell within the area between control/lime and wheat samples. Error bars represent the standard error.



Biolog® Microtiter Ecoplate Analysis

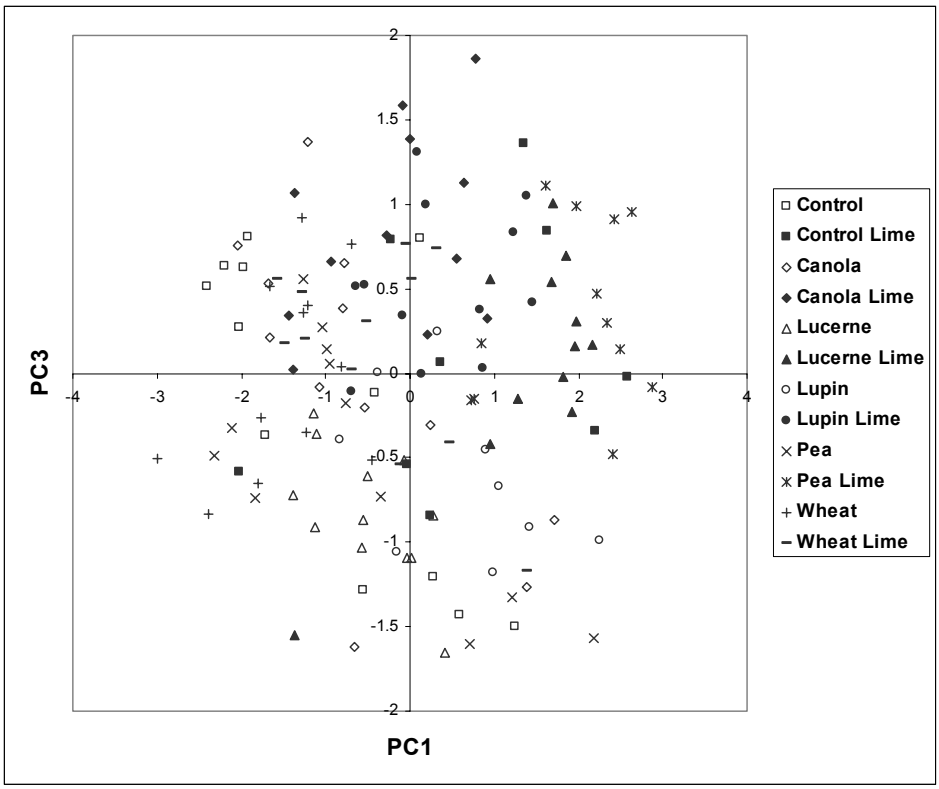
The first four principle components generated from the Biolog® data all revealed significant differences when subjected to ANOVA. Variation in PC1 (explaining 25.5% of total variation) was related mainly to treatment (lupin and wheat significantly different to most other treatments) and lime (pea/lime and lucerne/lime). PC2 (13.9%) variation was due to lime only, PC3 (8.1%) attributed to treatment (lucerne and lupin) and lime (canola/lime) and PC4 to treatment (mainly lupin and lucerne/lime). PC1 and PC4 additionally revealed significant interaction between treatment and lime effects. Principle Component scores separating the data most successfully are presented in Figure 2. Further analysis of Biolog® plates involved total utilisation of carbon sources in substrate groups. Of the six groups (amino acids, carbohydrates, carboxylic acids, phenolics, polymers and amines), four revealed significant changes in utilisation patterns (data not shown). Overall, carbohydrate, carboxylic acid and polymer utilisation was highest in pea/lime, and a significant lime effect was observed in phenolics utilisation.

Analysis of nif*H Gene Pools in rhizosphere samples*

As reflected by the levels of detectable fluorescence of the PCR products, the number of microorganisms containing *nif*H in limed soil appeared to increase (Figure 3). By

comparison, PCR amplicons from soil containing no lime were very faint. An analysis of the brightness of the bands using Scion Image (Scion Corporation USA) revealed significant differences attributed to treatment and lime effects. The addition of organic residues increased the amount of *nif*H in the rhizosphere in both limed (significant with canola, lucerne and pea residues compared with the control) and non-limed (canola significant compared with the control) soils. SSCP analysis revealed some variation between replicate pots in *nif*H distribution (Figure 4). Several bands recurred amongst the samples, especially the brightest band, which was more intense in canola, lucerne and pea samples compared with other treatments.

Figure 2. Principle Component Analysis of Biolog Plates. Plates were incubated at 25°C for 120 hours prior to reading.



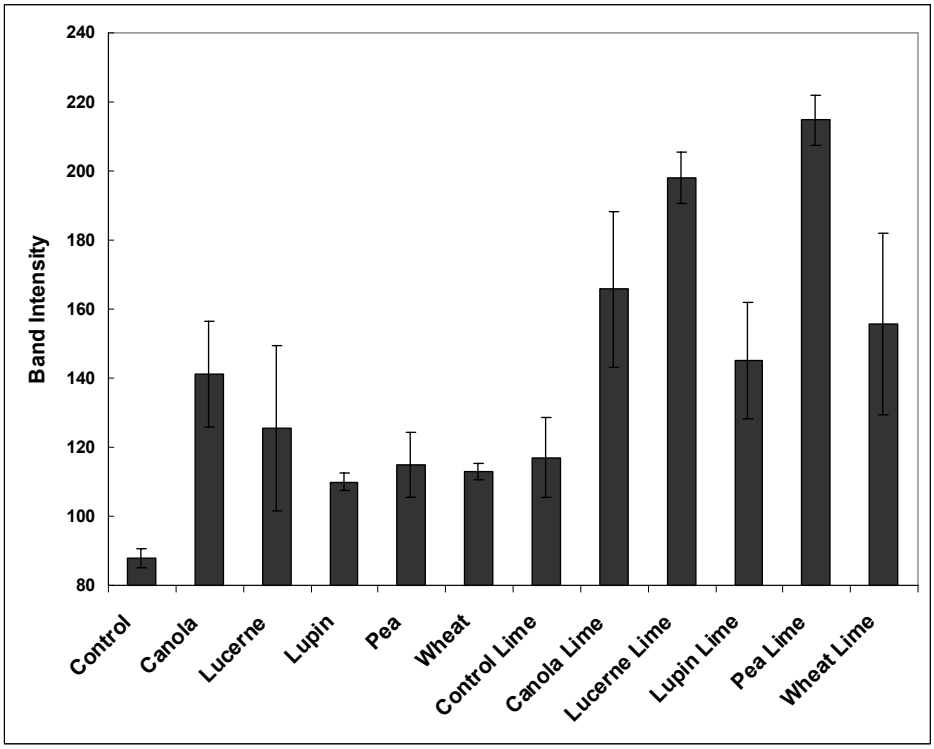
Discussion

Cereal crop producers have the capacity to influence soil microbiology in the root zone through residue management and lime application.

Biolog® Ecoplates are useful for detecting changes in microbial communities by identifying fingerprints based on carbon source utilisation patterns. The carbon sources consist of six groups that include many plant exudates and organic matter decomposition products that have proved useful in separating samples in previous studies (Hitzl et al 1997, Campbell et al 1997). Of the non-limed soils, those amended with lupin residues were significantly different to all other residue treatments, especially wheat, in both PCA and some substrate groupings. Apart from carbon and nitrogen, some other aspect of lupin residue may influence root zone microorganisms, such as calcium or manganese (Keyser, Munns 1979), and is currently being investigated. Interestingly, soil with lupin residues had the highest diversity of microbes compared with other non-limed soils, and wheat residues had significantly lower diversity (data

not shown). In limed soils, microbial populations in pea- and lucerne-amended soils significantly shifted compared with other treatments, also displaying the highest carbohydrate, carboxylic acid and polymer utilisation. The data presented here suggests significant changes in the rhizosphere microbial community when soil is ameliorated with lime and/or crop residues, and supports the suggestion that cereal crop producers can influence rhizosphere microbiology, and in turn, potentially influence plant growth.

Fig.3. *nif*H PCR product intensity. Standard errors are displayed.

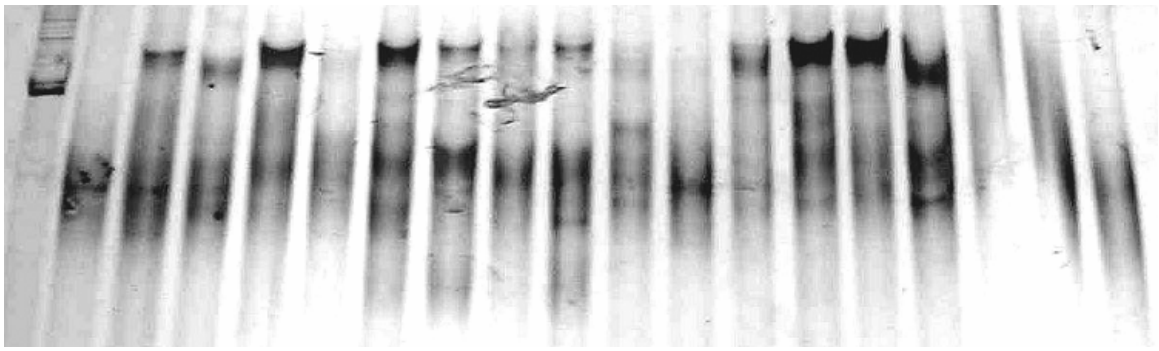


Nutrient 'lock-up', whereby nutrients for plants are in short supply due to microbial activity degrading organic matter, was a significant issue in these experiments. Plant growth was inhibited, and shoot nitrogen was lower in all cases where residues were added, except for lucerne. Nutrient 'lock-up' has been described in previous studies. Franzluebbers et al (1995) found that while mineralisable nitrogen was reduced in the short-term due to organic amendments, it increased in the long-term. In this experiment, residues were added one day prior to sowing, and incorporated throughout the pot in milled form, so the 'lock-up' was most likely exaggerated. The higher correlation in the non-limed treatments of residue nitrogen content vs. wheat shoot nitrogen suggested that nitrogen was the limiting factor in plant growth. In limed treatments, correlation was lower, suggesting an external input of nitrogen in those samples. This was probably provided by an increase in nitrogen fixers, which in most cases, was significantly higher in limed soils as opposed to the non-limed counterpart for each residue amendment.

SSCP is a relatively new molecular technique, first described by Orita et al (1989). The current method is based on that of Schwieger and Tebbe (1998), who first used the enzyme λ-exonuclease to digest one strand of double-stranded DNA. This method makes it possible to study complex microbial communities with many organisms containing the same gene. Residue and lime amendments significantly affected the communities of *nif*H-containing microorganisms in this study, although replicate

variation was a consideration. Currently, bands are being excised from the SSCP gels to determine the groups of nitrogen fixers that respond to residue and lime input.

Fig.4. SSCP analysis of *nif*H rhizosphere communities in limed soil. Three replicate samples are presented. Soils were amended with lime and canola (lanes 5-7), lucerne (lanes 8-10), lupin (lanes 11-13), pea (lanes 14-16) and wheat (lanes 17-19). Controls with no residue (lanes 2-4) and molecular weight markers (lane 1) are included.



This study has demonstrated that the rhizosphere can still be influenced by crop residue amendments and that liming is the most influential factor on rhizosphere nitrogen fixers, followed by crop residue amendments. In summary, it is suggested that cereal crop producers can manage soils and, in turn, influence rhizosphere microbiology by the use of crop residue and lime amendments.

References

Barakah FN, SH Salem, AM Heggo, MA Bin-Shiha 1995. Activities of rhizosphere microorganisms as affected by application of organic amendments in a calcareous loamy soil: 2. Nitrogen transformations. Arid Soil Research & Rehabilitation 9 467-480.

Burke DJ, EP Hamerlynck, D Hahn 2002. Interactions among plant species and microorganisms in salt marsh sediments. Applied & Environmental Microbiology 68(3) 1157-1164.

Campbell CD, SJ Grayston, DJ Hirst 1997. Use of rhizosphere carbon sources in sole carbon source tests to discriminate soil microbial communities. Journal of Microbiological Methods 30 33-41.

Cartwright B, BA Zarcinas, LA Spouncer 1986. Boron toxicity in South Australian barley crops. Australian Journal of Agricultural Research 37 351-359.

Coventry DR, WJ Slattery, VF Burnett, GW Ganning 1997. Liming and re-liming needs of acidic soils used for crop–pasture rotations. Australian Journal of Experimental Agriculture 37 577-581.

Franzluebbers AJ, FM Hons, DA Zuberer 1995. Soil organic carbon, microbial biomass and mineralisable carbon and nitrogen in sorghum. Soil Science Society of America Journal 59 460-466.

Harris RH, GJ Scammell, WJ Muller, JF Angus 2002. Crop productivity in relation to species of previous crops and management of previous pasture. Australian Journal of Agricultural Research 53 1271-1283.

Haun JR 1973. Visual quantification of wheat development. Agronomy Journal 65 116-119.

Hitzl W, A Rangger, S Sharma, H Insam 1997. Separation power of the 95 substrates of the BIOLOG system determined in various soils. FEMS Microbiology Ecology 22 167-174.

Hocking P 2001. Organic acids exuded from roots in phosphorus uptake and aluminium tolerance of plants in acid soils. Advances in Agronomy 74 63-97.

Keyser HH, DN Munns 1979. Effects of calcium, manganese and aluminium on growth of *Rhizobia* in acid media. Soil Science Society of America Journal 43(3) 500-503.

Lawrie JW, BW Murphy, IJ Packer, AJ Harte 2000. Soils and sustainable farming systems. In Soils: Their properties and management. Eds Charman PEV, Murphy BW pp 287-317. Oxford University Press, Melbourne.

Lepo JE, MK Chelius, DE Weber 1995. Characterisation of nitrogen-fixing bacterial rhizosphere communities using restriction fragment length polymorphisms of PCR-Amplified *nif*H. Proceedings of the Biotechnology risk assessment symposium Pensacola Beach, Florida, 6-8 June 1995 Eds Levin MA, Angle JS pp 408-422. University of Maryland Biotechnology Institute.

Marschner H 1995. Mineral nutrition of higher plants. Academic Press London.

Orita M, Iwahana H, Kanazawa H, Hayashi K, Sekiya T 1989. Detection of polymorphisms of human DNA by gel electrophoresis as single-strand conformation polymorphisms. Proceedings of the National Academy of Science USA 86 2766-2770.

Pankhurst CE, Lynch JM 1994. The role of the soil biota in sustainable agriculture. In Soil biota. Eds Pankhurst CE, Doube BM, Gupta VVSR, Grace PR pp3-9. CSIRO Melbourne.

Poly F, Jocteur Monrozier L, Bally R 2001. Improvement in the RFLP procedure for studying the diversity of *nif*H genes in communities of nitrogen fixers in soil. Research in Microbiology 152 95-103.

Poly F, Ranjard L, Nazaret S, Gourbière F, Jocteur Monrozier L 2001. Comparison of *nif*H gene pools in soils and soil microenvironments with contrasting properties. Applied & Environmental Microbiology 67(5) 2255-2262.

Rowell DL 1994. Soil science: Methods and applications. Longman Scientific and Technical, London.

Schwieger F, Tebbe C 1998. A new approach to utilise PCR-single-strand-conformation-polymorphism for 16s rRNA gene-based microbial community analysis. Applied & Environmental Microbiology 64(12) 4870-4876.

Scott BJ, Conyers MK, Poile GJ, Cullis BR 1999. Reacidification and reliming effects on soil properties and wheat yield. Australian Journal of Experimental Agriculture
39 849-856.

Scott BJ, Fisher JA, Cullis BR 2001. Aluminium tolerance and lime increase wheat yield on the acidic soils of central and southern New South Wales. Australian Journal of Experimental Agriculture 41 523-532.

Slattery JF, Coventry DA, Slattery WJ 2001. Rhizobial ecology as affected by the soil environment. Australian Journal of Experimental Agriculture 41 289-298.

Smith MS, Firestone MK, Tiedje JM 1978. The acetylene inhibition method for short-term measurement of soil denitrification and its evaluation using nitrogen-13. Soil Science Society of America Journal 42 611-615.

Vallini G, Pera A, Agnolucci M, Valdrighi MM 1997. Humic acids stimulate growth and activity of the in vitro tested axenic cultures of soil autotrophic nitrifying bacteria. Biology & Fertility of Soils 24 243-248.

Widmer F, Shaffer BT, Porteous LA, Seidler RJ 1999. Analysis of *nif*H gene pool complexity in soil and litter at a Douglas fir forest site in the Oregon Cascade Mountain Range. Applied & Environmental Microbiology 65(2) 374-380.

Wieland G, Neumann R, Backhaus H 2001. Variation of microbial communities in soil, rhizosphere, and rhizoplane in response to crop species, soil type, and crop development. Applied & Environmental Microbiology 67(12) 5849-5854.

Yeates C, Gillings M 1998. Rapid purification of DNA from soil for molecular biodiversity analysis. Letters in Applied Microbiology 27 49-53.

Contact details

Dr Darryl Nelson
Microbial Ecologist
Department of Primary Industries
RMB 1145, Chiltern Valley Road Rutherglen Victoria 3685
Phone 02 6030 4538
Fax 02 6030 4600
Email Darryl.Nelson@dpi.vic.gov.au

Poster
Soil health benchmarking in the macadamia industry

Justine Cox[1], Russ Stephenson[2], Pat O'Farrell[2], Jenny Cobon[2], Phil Moody[3], Lukas Van Zwieten[4], Paul O'Hare[2], Tony Pattison[2], Chris Searle[2]

[1]NSW Dept Primary Industries, Centre for Tropical Horticulture, Alstonville
[2]Qld Dept Primary Industries and Fisheries, Nambour, Brisbane, Mareeba
[3]Qld Dept Natural Resources, Mining and Energy, Brisbane
[4]NSW Dept Primary Industries, Wollongbar Agricultural Institute.

Introduction

The macadamia industry raised concern about the sustainability of the soil resource after a project assessing soil health in the subtropics of NSW quantified soil degradation under macadamia orchards. The industry was aware that the exposed soil floor necessary for surface harvesting of nuts may have led to soil loss over years of production. It also may have contributed to tree yield decline observed in orchards. The macadamia industry wanted to benchmark the soil condition at several growing regions with different soil types. The multidisciplinary research and extension team from NSW and Qld decided upon a suite of soil health indicators to characterise the soil's physical, chemical and biological attributes.

Methods

Orchards in Clunes (northern NSW), Gympie, Bundaberg and Atherton tablelands (Qld) were chosen to represent the major growing areas. Soil types were red Ferrosols (clays) at Clunes and Atherton, a Kurosol at Gympie and a Kandosol at Bundaberg (both sandy). The orchards were sampled in the row and inter-row, and compared with adjacent undisturbed natural vegetation (subtropical rainforest in Clunes and sclerophyll forest in Gympie, with no site available in Bundaberg). Four replicates of soil (at depths of 0-10 and 50-60 cm) and root samples were taken at each position. Measurements included bulk density, particle size analysis, aggregate stability (wet sieving), infiltration rate, gravimetric moisture content, effective cation exchange capacity (ECEC), soil pH, total carbon, total nitrogen, exchangeable cations, labile carbon, microbial biomass carbon, microbial activity (FDA hydrolysis), nematode community structure, root length density and proteoid root area.

Preliminary results

The microbial activity in the undisturbed area in the Atherton tablelands was three times higher than the row and inter-row. Activity in the row, inter-row and undisturbed area at Clunes were all similar. At Gympie activity in the inter-row was twice the level in the row and undisturbed area. Even though comparisons with undisturbed areas did not show trends, the activity in the inter-row was always higher than the row. This is likely to be due to the groundcover roots in the inter-row providing an adequate environment for microbes. The microbial biomass carbon results showed that in the ferrosols, there was a higher microbial population in the undisturbed areas. At Gympie the undisturbed and inter-row populations were similar. The microbes were more abundant in the inter-row than the row.

Total carbon, nitrogen and labile carbon values showed a similar pattern to the microbes. The ratio Cmic:Ctot was used to compare all sites regardless of soil type to show efficiency of organic matter use. Given the pool of carbon in each sample, the

microbial biomass was highest in the undisturbed areas. Possible explanations are that the increased diversity of carbon compounds in the natural systems can support a more diverse and abundant population of organisms, or that disturbance to the orchard soil has reduced the chances of microbial population build up and development.

The nematode analysis differentiated degraded and disturbed sites with reduced nematode diversity in the row. Bulk density was higher in the orchards than in the undisturbed areas and higher in the row than inter-row. Except for Clunes, undisturbed sites always had more macro- and micro- aggregates than row or inter-row areas. Overall, from a soil chemistry viewpoint, the rows were considered in good shape for pH, EC, exchangeable cations and ECEC. The biological indicators were therefore more able to pick up evidence of early soil degradation.

Contact details

Justine Cox
Soil Scientist
Centre for Tropical Horticulture
NSWDPI PO Box 72 Alstonville NSW 2477
Phone 02 6626 2434
Fax 02 6628 5209
Email justine.cox@dpi.nsw.gov.au

Poster
Relationship between arbuscular mycorrhiza fungi infectivity, soil characteristics and land use history

Irnanda Djuuna, Lyn Abbott, Guy Boggs, Kimberly van Niel
University of Western Australia

A geographical information system (GIS) is proposed as a suitable tool for mapping the relationship between land use and mycorrhizal infectivity. A farm in south-western Australia was sampled on a 150 m grid across 14 paddocks. The 291 composite soil samples were collected for soil testing and for mycorrhizal infectivity. The ArcView and ARC/INFO packages were used to identify relationships between land use, soil tests and mycorrhizal infectivity. GIS was used to analyse the relationships between variables. Both soil characteristics and mycorrhizal infectivity varied with paddock history. The crop (or pasture) one year before sampling and fertiliser use were the most important factors directly related to variability in mycorrhiza infectivity. The crop (or pasture) two years before sampling also had a significant effect on the level of mycorrhizal infectivity. There was no correlation between infectivity of AM fungi and soil phosphorus or between microbial biomass and AM fungi infectivity.

Further data analysis will be conducted to compare soil management, paddock history and soil characteristics. The shapes of any relationships and multiple variable interactions may be more important in field studies than correlations at a point, and this is being investigated.

Contact details

Irnanda Djuuna
Postgraduate Student
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia Crawley WA 6009
Phone 08 6488 3595
Fax 08 6488 1050
Email djuuni@cyllene.uwa.edu.au

Poster
Temporal dynamics and critical periods of plant-specific microbial functions in southern Australian cropping regions

VVSR Gupta, David Roget
CSIRO Land and Water, Glen Osmond SA

Production by both crops and pastures is supported and enhanced by soil biological processes. Australian soils are generally low in biologically available organic carbon so most biological activity is concentrated near decomposing crop residues in a thin layer of surface soil and in the rhizosphere. Plants are the major source of available carbon for biological activity, so soil biodiversity and biological activity depend on the quality and quantity of carbon inputs from plants through root exudation and above- and below-ground plant residues, and on plant-induced changes in soil physical and chemical properties. Since plant type has a critical role both directly and indirectly through changing the soil environment and in modifying biological communities, a plant-specific approach is better suited for maximising benefits from biological functions in these environments.

The composition and metabolic capabilities of the soil microbial and faunal communities underlie the occurrence and rates of many soil processes such as disease suppression, nutrient mineralisation and leaching losses. In most dryland cropping regions, moisture availability plays a critical role in determining the activity of both microflora and soil fauna. In southern Australia, temporal patchiness in the favourable soil and environmental conditions dictate the functional role of various groups of biota and the true significance of plant-specific biological functions for crop productivity and soil health.

We need to know more about the temporal dynamics of mineralisation--immobilisation processes to synchronise nutrient availability to plant needs and restrict losses through leaching. We need to know the optimum soil and environmental conditions for larger soil fauna which are present in measurable populations in the dryland Mallee region but rarely play a significant role in ecosystem functions. Similarly identification of critical periods for the various interactions between pathogen-soil biota-plant interactions is needed for promoting the natural disease suppressiveness in different southern Australian cropping regions. This paper discusses some examples of temporal dynamics of specific microbial functions related to plant nutrition and health.

Contact details

Dr VVSR Gupta
Senior Research Scientist
CSIRO Land & Water
PMB 2 Glen Osmond SA 5064
Phone 08 8303 8579
Fax 08 8303 8550
Email Gupta.Vadakattu@csiro.au

Poster
Effect of stubble burning and seasonality on microbial processes and nutrient cycling

Frances Hoyle, Daniel Murphy
University of Western Australia

In this poster, we examine results from a long-term stubble management trial, where stubble has either been retained or burnt. The experimental site, established in 1987 (G. Reithmuller WADA) was located on a red-brown earth, a heavy clay loam, at Merredin and was used to determine the effect of stubble management on microbial activity, soil organic matter-carbon turnover and related nutrient cycling. The site has been continuously cropped since establishment in a wheat:legume rotation, and stubble either retained or burnt.

In 2003, wheat cultivar 'Wyalkatchem' was sown at 110 kg/ha after adjustment for seed size and assuming a field emergence of 70%, to achieve a target density of 200 plants per m^2. The trial was sown on June 7 in a randomised block design with various row spacing treatments, and replicated six times. For the purpose of this investigation, three replicate plots were sampled from both the stubble retained or burnt treatments sown on 180 mm row spacings. All treatments were sown with a basal fertiliser of 150 kg/ha Agras (17.5% N) on narrow points with press wheels.

Initial screening of a range of parameters was conducted prior to sowing to characterise the background soil fertility and biological activity of the site. Physical soil and stubble parameters included bulk density, soil moisture and water holding capacity. Analyses for chemical status of the soil indicated there were no measurable constraints to yield at this site.

Microbial activity was found to be greater where stubble was retained, particularly at soil temperatures above 10°C. Stubble retention also resulted in a higher mass of microorganisms in the surface (0-5 cm) layer of the soil. Microbial nitrogen was estimated at 89 kg N/ha in stubble retained and 67 kg N/ha in burnt treatments. This means that there is equivalent to 192 kg/ha (stubble retained) and 146 kg/ha (stubble burnt) of urea contained within the soil microorganisms, a significant source of potentially plant available nitrogen.

Changes in the mass of microorganisms and their activity are commonly reflected in changes to the soil supply rate of both carbon and nitrogen. In this experiment, the stubble retained treatments released more soil nitrogen than the burnt treatments. Clearly these results illustrate that stubble retention promoted both a larger but also more active microbial community compared with burning. Grain yield (estimated by hand harvest) at this site reached 3.07 t/ha for stubble retained and 2.28 t/ha for burnt treatments. Grain protein was measured at 9.51% (retained) and 10.96% (burnt). This indicates a nitrogen uptake of approximately 292 and 250 kg/ha. The results obtained in this study suggest the relationship between stubble retention and grain yield observed can in part be associated with changes in the amount of nitrogen supplied from the microorganisms.

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Contact details

Frances Hoyle
Research Officer
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia Crawley WA 6009
Phone 08 6488 7345
Fax 08 6488 1050
Email fhoyle@agric.uwa.edu.au

Poster
Effect of brown and green manure residue incorporation on the mass of microorganisms and associated release of plant available nitrogen

Daniel Murphy, Frances Hoyle, Nui Milton
University of Western Australia

In this poster, we examine results from a trial at Avondale Research Station, WA where oat and pea crops were incorporated as brown/green manures in spring 2001 and plots 10 metres wide and 20 metres long were subsequently sown to a wheat in 2002. Several spring manipulation treatments were imposed within each block (split block design) at flowering, the exception being the control plots which were taken to harvest in 2001. Residue incorporation treatments consisted of different levels of soil disturbance designed to simulate different rates of nutrient release from the crop residues. In 2002, wheat cultivar 'Carnamah' was sown at 75 kg/ha to achieve a target density of 120 plants per m^2. The trial was sown plot on plot, on June 13 in a randomised split block design, and replicated three times. All treatments were sown with a basal fertiliser of 115 kg/ha DAPSZC at seeding and topdressed on July 22 with 75 kg/ha urea (46% N). Crops were direct-seeded (knife points) in eight row plots, two metres wide and 20 metres long. Soil (0-50 cm) was collected on a number of occasions during the 2002 wheat phase to quantify the impact of residue incorporation on the mass of microorganisms and soil inorganic nitrogen profiles.

For both oat and pea residue treatments, subsequent wheat grain yields were greater in plots that had received brown or green manure compared with harvested plots (ie where the oat and peas in 2001 were grown to maturity and not incorporated as residues). Grain protein and nitrogen uptake in grain (measured by grain protein % x grain yield) were also greater in residue treatments than under harvested plots, indicating that a total of between 68-77 kg of additional N/ha was assimilated in residue treatments.

The amount of microorganisms in soil varied throughout the season with a general decline through the season. These variations are related to wet and dry cycles in the soil, and the availability of easily decomposable food substrates such as decomposing residues and plant root exudates. The amount of microorganisms was generally greater under field pea residue treatments. However, no differences in the mass of microorganisms associated with incorporation treatments were observed in this soil.

There were high inorganic nitrogen levels in the soil following brown/green manures. Values were higher after peas compared with oats which is related to differences in the quality of plant residues (ie the higher carbon:nitrogen ratio of the oats) and the subsequent release of plant-available nitrogen. With oats the degree of incorporation of the residue within the soil did not affect inorganic nitrogen release. However, increased incorporation of pea residues resulted in larger amounts of inorganic nitrogen release. This occurs because there is greater surface area contact between residue and soil microorganisms causing faster residue decomposition. This results in greater nutrient release but also faster decline in residue and soil organic matter levels.

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Dr Daniel Murphy
Research Fellow
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia, Crawley, WA 6009
Phone 08 6488 7083
Fax 08 6488 1050
Email dmurphy@agric.uwa.edu.au
Websites www.clr.uwa.edu.au, www.soilhealth.com

Poster
Mapping biological soil nitrogen supply using mid infrared technology

Daniel Murphy, Nui Milton
University of Western Australia

Introduction

Nitrogen is the primary nutrient limiting crop production in farming systems throughout the world. In our agricultural soils 40-80% of crop nitrogen requirements are met through microorganisms. They breakdown residues and organic matter to release plant available nitrogen (ie biological soil nitrogen supply). The remaining nitrogen requirement is met through fertiliser applications. To improve nitrogen fertiliser management it is important to know the timing and location of biological soil nitrogen supply, so that fertiliser nitrogen is applied only when and where necessary.

Splitting applications of fertiliser nitrogen at strategic plant growth stages is becoming common. This maximises the opportunity for crop uptake at the right time and minimises the risk of nutrient leaching. Growers can also spatially adjust fertiliser rates in the field using information gained from yield mapping, through knowledge of best/worst performing areas of a paddock, and by soil type. The next extension of this approach is to utilise spatial soil maps that tell us about the soil's capacity for biological nitrogen supply together with information on the chemical and physical fertility of the soil. This allows soil constraints limiting crop production to be identified (see paper by Murphy et al, this proceedings). For example where biological soil nitrogen supply is high, less fertiliser nitrogen may be needed to achieve optimal yields. Alternatively where biological soil nitrogen supply is low greater reliance on fertiliser nitrogen is required for adequate crop growth. This would have an economic and environmental (minimising leaching) benefit to growers.

How do we know what the capacity of the soil is for biological supply of nitrogen?

In the laboratory we incubated soil and measured potentially mineralisable nitrogen as an index of biological soil nitrogen supply. This method takes one week to complete and is thus both too costly and slow for use as a decision support tool for fertiliser application rates. However, this may all change in the future. Our current work is exploring the possibility of using mid infrared technology to develop calibration curves for a range of soil biological, chemical and physical soil properties (eg biological soil nitrogen supply, organic matter, cation exchange capacity, clay content). The advantage of the mid infrared technology is that, once calibrated, soil samples can be collected from the field and scanned rapidly (two minutes per sample) to provide predictions for a number of soil properties. This process considerably reduces analytical costs, so growers could afford to have more soil samples analysed, enabling spatial maps to be generated (see Figure 4 in Murphy et al, this proceedings) or deeper soil layers to be assessed.

Mid infrared is not as accurate as measuring each soil property by standard analytical techniques but does have a place in the development of soil spatial maps for the purpose of zoning fields to allow for variable management strategies and to identify soil constraints currently restricting crop production. The application of this technology is

demonstrated in this poster where there was good agreement between measured and mid infrared predicted values of biological soil nitrogen supply and key soil chemical and physical properties.

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Contact details

Dr Daniel Murphy
Research Fellow
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia Crawley WA 6009
Phone 08 6488 7083
Fax 08 6488 1050
Email dmurphy@agric.uwa.edu.au
Websites www.clr.uwa.edu.au, www.soilhealth.com

Poster
Effects of fumigation and soil amendments on nematode-feeding groups in cereal growing soils

Jackie Nobbs[A], Daniel Murphy [B], Frances Hoyle[B], Nui Milton[B], Sharyn Taylor[A]
[A] SARDI GPO Box 397 Adelaide SA 5001
[B] The University of Western Australia

Introduction
Little is known of the impact of broadacre management practices on the different nematode-feeding groups in Western Australian soils. This research investigated the response of nematode functional groups and the soil microbial biomass (ie mass of living microbes in soil) to commercial products designed to either attack pathogenic nematodes or enhance overall soil biological fertility.

Materials and methods
Trials were located in the northern (low rainfall, Latham) and southern (high rainfall, Jerramungup) grain-growing regions of Western Australia. Site locations were chosen based on an initial screen of sites for high levels of *Pratylenchus neglectus* using the PredictaB ^{TM} DNA disease diagnostic service.

Six treatments were examined:

- control
- methyl bromide (at a rate of 1 kg/10m^2) pumped across the soil surface under plastic covering that was left for 24 hours before removal
- a talc-based formulation containing 100 million spores per gram of *Trichoderma harzianum*, *T. lignorum*, *Gliocladium virens* and *Bacillus subtilis* applied as a solution with 1 kg talc/ha.
- beneficial mix of microorganisms applied with a food source after being incubated for 24 hours (commercial formulation of undisclosed composition)
- predatory fungi formulation consisting of a 5 kg talc applied as a solution containing 1 x 10 exp^9 CFU per gram of *Arthrobotrys oligospora, A. conoidus, Paecilomyces fumosoroseus, P. lilacinus,* and *Verticillium chlamydosporium*
- food source developed to support microbial populations and obtained as a commercial formulation with an undisclosed microbial food preparation.

Findings
The soil treatments investigated at these sites were considered to be ineffective for broadacre farming. Changes to nematode composition and microbial biomass were small and did not result in a difference in plant biomass production. Improvements in the effectiveness of the soil treatments may occur when applied as seed dressings; this is currently being evaluated

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Contact details

Dr Daniel Murphy
Research Fellow
Centre for Land Rehabilitation
School of Earth and Geographical Sciences
Faculty of Natural and Agricultural Sciences
The University of Western Australia Crawley WA 6009
Phone 08 6488 7083
Fax 08 6488 1050
Email dmurphy@agric.uwa.edu.au
Websites www.clr.uwa.edu.au, www.soilhealth.com

Poster
Soil biology in the paddock

Ian Packer[A], Jennifer Johnson[B], Richard Langley[C]
[A] NSW Department of Infrastructure, Planning and Natural Resources, Cowra
[B] University of Sydney, Leeds Place, Orange
[C] Homestead View', Greenethorpe

Introduction
Studies of conservation farming practices have shown significant improvements in soil physical properties but there is a heavy reliance on inputs such as artificial fertilisers, herbicides and fungicides to maintain productivity. Limited research data and anecdotal observation by landholders show it is possible to address these problems and gain economic and environmental advantages by using practices which improve and maintain soil health or quality. This paper presents preliminary data on the change in soil health observed and measured by a central NSW landholder farming on a fragile red Chromosol soil.

Methods
Soil samples were collected from three sites.

- Langley paddock: direct-drilled with stubble from 1989-1999 and then no tilled with controlled traffic 1999 to present.
- Neighbour paddock: conventionally farmed for the past 10 years.
- Roadside: area adjacent to Langley paddock which has been undisturbed and ungrazed for the last 15 years.

Fifteen samples were collected from each area for

- bulk density using a 7.5 cm X 7.5 cm core
- a bulked sub sample to measure soil organic carbon and total nitrogen levels using the LECO method
- 5 cm probe samples for plating on a potato dextrose agar (PDA) solid medium.

Recognisable Taxonomic Unit (RTU) assessment was carried out for the PDA plates. Unfortunately expertise was not available to classify the RTU units.

Results
Bulk density was lowest in the Langley paddock and highest in the Neighbour paddock. Carbon density was highest in the Roadside site. Several RTU species were present in the Langley paddock and Roadside site, but not in the Neighbour paddock. However, RTU #1 was highly abundant in the Langley and Neighbour paddocks compared with the Roadside site. However RTU#1 species vigour was greater in the Langley paddock than the Neighbour paddock.

Conclusion
Hopefully this preliminary data can attract further funding for studies to classify the biological activity. The change is visually evident in colour and friability. Soil health benefits for the farmer are reduced herbicides for weed control, no obvious soil disease problems, more soil moisture for sowing and finishing crops, reduced soil acidification, significantly reduced nitrogen input at sowing and no erosion problems. These benefits have an economic and productivity reward, particularly gross margins. Seeing real soil

health in a real paddock done by a real farmer is a future research challenge and direction to logically explain these obvious soil changes.

Contact details

Ian Packer
Senior Natural Resource Officer
DIPNR PO Box 510 Cowra NSW 2794
Phone 02 6341 9321
Fax 02 6342 2565
Email Ian.Packer@dipnr.nsw.gov.au

Poster
Soil health card

Greg Reid
NSW Department of Primary industries, Wollongbar

The soil health card is a suite of do it yourself tests that investigate the soil/plant environment. The development of the card was driven by the desire of landowners to know more about their soils than the nutrient levels available in laboratory reports.

Members of Tuckombil Landcare, and staff of Wollongbar TAFE and Wollongbar Agricultural Institute worked with local farmers to develop a card that had broad application and was cheap and easy to use.

The card explains when and where to test soil and recommends sampling patterns for different situations. It provides instructions on how to make simple equipment to measure different aspects of soil health, and how to measure ten criteria: ground cover, compaction, infiltration, diversity of soil life, root development, soil structure, aggregate stability, earthworms, acidity and leaf colour. Farmers record results on a record sheet which immediately grades the results of each test, so that soil health patterns are seen easily. A trouble-shooting guide links observed soil problems to possible causes.

Demand for the card has been so strong that the card is in its third printing less than eighteen months after launching. A survey of users has found that almost all report having learnt something new about their soil and discovering unsuspected variations in the soil across their fields. Most report that the card has influenced discissions about soil management.

The Northern Rivers Soil Health Card is a valuable template that can be easily adapted to different regions or to the specialised needs of particular agricultural industries.

You can obtain the card on the internet at www.tuckombillandcare.org.au or by contacting Greg Reid.

Contact details

Greg Reid
Soils Advisory Officer
Wollongbar Agricultural Institute
NSWDPI 1243 Bruxner Highway Wollongbar NSW 2477
Phone 02 6626 1357
Fax 02 6628 1744
Email greg.reid@dpi.nsw.gov.au

Poster
The Australian Soil Club

Jen Slater and Lyn Abbott
University of Western Australia

The Australian Soil Club (ASC) began as a collaboration between the Kondinin Group and Lyn Abbott from The University of Western Australia. It is a national network of land managers and others interested in increasing their knowledge of soils and sustainable management practices. The objective is to provide members with access to the latest information and research into the physical, chemical and biological aspects of soil. The Australian Soil Club Newsletter is produced for members on a bi-monthly basis. In addition, it is intended to hold workshops and seminars according to the members' interests and to develop a website.

There are currently 250 members of the Australian Soil Club across Australia. Topics of interest identified by members include: soil testing, straw and stubble management, soil compaction and hard setting soils, nutrient management and recognition of deficiencies, soil moisture issues and management, liming, organic fertilisers, acidity, salinity, soil electrical conductivity, and increasing organic matter. Authors for the ASC newsletter articles have been from a range of education and research centres such as NSW Department of Primary Industries, The University of Western Australia and the Centre for Excellence in Natural Resource Management in Albany, Western Australia.

The first seminar held by the ASC was arranged by an ASC member in Young, NSW, and was attended fifty members of the ASC and other interested people on the issue of soil biological fertility. Other such events can be organised.

The Club is now affiliated with the Kojonup Soils Centre in Western Australia (which has been established by farmers with the objective of promoting knowledge of soil). We will be appointing State coordinators of the ASC to assist with organisation of events and recruiting authors to provide articles on any aspect of soils as identified by members. If you are interested in participating, please contact Lyn Abbott.

Contact details

Professor Lyn Abbott
Head, School of Earth and Geographical Sciences
The University of Western Australia
35 Stirling Highway Crawley WA 6009
Phone 08 6488 2499
Fax 08 6488 1050
Email labbott@cyllene.uwa.edu.au