# SUSTAINABLE MANAGEMENT OF SOIL ORGANIC MATTER

# **Sustainable Management of Soil Organic Matter**

### Edited by

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## **Preface**

With the rapidly emerging interest from many sectors of society in sustainable development, there is a realization that an understanding of soil management is of fundamental importance to the debate. Before the latter part of the 20th century, soil was seen as a matrix from which food could be produced and on which wastes could be disposed or buildings constructed. Soils research focused primarily on how an understanding of relevant processes could be applied to optimize food production. Although soil protection has, for many decades, been an issue with which scientists and land managers have been concerned, it has focused primarily on the need to prevent soil loss through erosion rather than to enhance soil quality through management per se. During the 1980s and 1990s, a change in attitude to soil began to take place. Concern about environmental issues such as climate change, and the physical and chemical degradation of soils was accompanied by an appreciation of the need to protect soils in their own right in order to underpin efforts to develop society in a way which is sustainable over the long term. Many countries are now developing statutory soil protection policies in an analogous way to those applied to air and water. There is a substantial body of research (some of which is reported in this book) that has defined the importance of understanding soil quality as an issue. Thus a practice of land use that is sustainable needs not only to preserve soil materials but also to maintain or enhance various attributes of its quality. The organic matter content of soils and the components of the organic matter itself play a vital role in defining this quality.

The aim of this book is to examine the role that organic matter plays in soils with a view to understanding the ways in which this contributes xx Preface

to the various functions of soils. The book is based upon papers offered to the annual meeting of the British Society of Soil Science entitled 'The Sustainable Management Of Soil Organic Matter' held in Edinburgh in September 1999. The meeting was host to around 200 delegates from more than 20 different countries. Authors were invited to submit papers for publication, which were then selected on the basis of peer review. The structure of the book follows that of the conference, with keynote papers followed by shorter more focused papers in five main sections;

- · Organic matter and sustainability
- Modelling soil organic matter dynamics
- Soil organic matter: the roles of residue quality in C sequestration and N supply
- The role of soil organic matter and manures in sustainable nutrient cycling
- Implications of soil biodiversity for sustainable organic matter management.

After the death of Walter Russell, the British Society of Soil Science decided to honour his contribution to soil science by the creation of a Memorial Lecture, to be given at the Society's Annual Meeting in alternate years. The first of these lectures was by Professor Dennis Greenland at the Society's 50th anniversary meeting in Newcastle in September 1997. The second lecture was given by Professor James Tiedje at the Edinburgh conference in 1999. His intriguing insight into the soil as a habitat of microbial diversity and his vision for the application of new technologies to expand the frontiers of soil science is presented in the penultimate chapter.

We wish to thank all of those involved in helping to organize the conference and prepare the resulting publication. In particular, we are indebted to Sally Burgess, David Green, Jane Lund, John Parker, Rachel Thorman and Adrian Tams for support during the conference. We also wish to thank numerous anonymous referees for their help with the revisions of papers, and Sandra Chalmers, Frances Haldane, Aileen Stewart and Pat Carnegie for help with the preparation of this publication.

R.M. Rees B.C. Ball C.D. Campbell C.A. Watson

### Organic Matter - the Sustenance of Soil

The functioning of soils is profoundly influenced by their organic matter content. The abilities of a soil to supply nutrients, store water, release greenhouse gases, modify pollutants, resist physical degradation and produce crops within a sustainably managed framework are all strongly affected by the quality and quantity of the organic matter that it contains. These attributes of organic matter lead it to have a major influence on the quality of soil material itself. As societies throughout the world begin to realize the potential value of the soil resource in contributing to sustainable farming practices, then the need to understand the role that organic matter plays in contributing to soil quality has become more important.

The paradigm of sustainability is one that most scientists subscribe to. However, concepts of sustainable soil management differ widely, and a wide variety of approaches are advocated that all aim to resolve a common problem. As we enter the 21st century, pressure on the world's ecosystems to provide for human needs is at an unprecedented level. It was estimated by Oldeman (1994) that by 1990, some 562 million hectares (38% of the world's cropland) had been degraded by poor agricultural practices. Although some degradation was relatively minor, it was recognized as being sufficient to impair at least some attribute of the soil's function and, in some cases, to lead to complete crop failure. During the 1990s, further damage has occurred, with the annual degradation of 5–6 million hectares, and current trends are not encouraging (UNEP, 1997).

Losses in the organic matter content of soils during the last 100 years have been substantial, and have been associated with changing patterns of land use that are driven by population increases. The process of cultivation of native soils is nearly always associated with a loss of organic carbon, as previously protected organic matter is oxidized following exposure to the atmosphere (Davidson and Ackerman, 1993; Gregorich *et al.*, 1998). The current global store of organic carbon in agricultural soils is thought to be ~168 Pg (Paustian *et al.*, 1997). Estimates suggest that this has declined from ~209–222 Pg, resulting in a loss of 41–54 Pg (Houghton and Skole, 1990; IPCC, 1996). It is likely that these losses were not evenly distributed across the globe, with disproportionately large losses from upland, organic and wetland soils. Losses of soil organic matter are also associated with land use change other than direct conversion to agriculture, such as deforestation and biomass burning (IPCC, 1996).

Given the importance of soil organic matter in contributing to essential soil functions, these losses are clearly of concern. In some circumstances, organic matter loss will almost certainly have contributed to catastrophic soil damage through soil erosion and loss in productive capacity. However, given the inevitability of some organic matter loss following cultivation, the question arises of whether it is possible to manage systems sustainably on a lower organic matter capital, or whether the losses that have occurred so far are part of a continuing trend towards loss of fertility and productive potential.

One of the key functions of organic matter in soils is that of nutrient supply, and an indication of the pressures that are being placed on cultivated soils to produce crops is provided by nutrient budgeting approaches. The balance between the input and output of nutrients within a given area provides a quantifiable indicator of sustainability. Some analyses carried out in parts of Africa show that nutrients are being depleted at an alarming rate (Smaling et al., 1996). Nutrient budgets can be used at differing scales and, although associated with a high degree of spatial heterogeneity, they can be valuable in identifying regional trends. In Nigeria, Smaling et al. (1996) found that the difference between the input and output of N leads to an average net annual loss of 27 kg ha<sup>-1</sup>, in soils that in many cases are already nutrient poor. This compares with an average nitrogen surplus in Germany of 47 kg ha<sup>-1</sup>. Although the African situation is clearly not sustainable, the solutions need to be broader than limited attempts at providing additional fertilizer inputs. Policies of integrated fertility management are seen as the most useful approach to overcoming such problems in which increased nutrient supply is provided within a framework of improved land and soil management policies. These must include organic matter management to increase soil nutrient capital and improve the sustainability of production (Smaling, 1998; Scoones and Toulmin, 1999).

In temperate regions, nutrient budgets may be more balanced, as a result of high external inputs of both inorganic and organic nutrients, but can often result in high inputs and high losses (Rosswall and Paustian, 1984). Long-term field trials at Rothamsted in the South East of England have established that the organic matter and nitrogen capital of cultivated soils can increase as a result of long-term cultivations (Powlson et al., 1986; Johnston, 1997). The increased accumulation of organic matter resulting from increased fertilizer applications and the consequent production of more plant material potentially can cause difficulties. Shen et al. (1989) found that soils that had received an annual addition of 144 kg N ha<sup>-1</sup> over 137 years contained more organic matter than those receiving no fertilizer additions. However, this was associated with significantly higher mineralization rates, and potentially higher losses. Positive nutrient balances do not necessarily equate therefore with improved sustainability, as environmental degradation may be linked with nutrient loss in some circumstances. Positive nutrient balances very often will depend on external inputs of nutrients, and are often coupled with external energy inputs to a system that further weakens its sustainability. Pimentel and Heichel (1991) found that the energy output/input ratio of maize grown in Mexico using only human labour was 12.9, with a crop yield of 1944 kg ha<sup>-1</sup>. In Minnesota, a field under conventional management supported grain yields of 6500 kg ha<sup>-1</sup>; however, the intensive management meant that the energy output/input ratio was only 3.3. Such systems again must ultimately be unsustainable as they currently depend upon the substantial input of finite non-renewable energy reserves in, for example, fuel and fertilizers. Although technologies requiring reduced inputs are available (Vanlauwe et al., 1996) government policies towards agriculture and the production of commodities need modification (e.g. extensification) if large-scale improvements in energy balances are to be achieved.

## Soil Organic Matter and Sustainability

There is much debate about the meaning of the term sustainability; however, a definition that has proved to be useful is that offered by the Bruntland Report (WCED, 1987) and is one that simultaneously 'Meets the needs and aspirations of the present without compromising the ability of future generations to meet their own needs'. These needs will, amongst other things, require an increased crop production per unit area of land to meet the demands of an increasing population, and therefore a need, as a consequence, to develop a better understanding of the processes that underlie such production.

One of the important contributions of organic matter to sustainability compared with other soil properties is that it influences many soil functions

(Swift and Woomer, 1993). The effects of organic matter in soils interact to influence the biological, chemical and physical properties of the soil material itself. In an attempt to quantify the role of soils in contributing to sustainable land use, the concept of soil quality has proved useful (Carter, Chapter 1). This involves assessing the ability of a soil to undertake a particular function. Thus the quality of a soil which is assessed for the purpose of storing water may be different from the quality of the same soil when assessed for the purpose of growing wheat. Soil organic matter strongly influences the quality of soils when assessed according to these criteria. However, it can be argued that one of the most important features of a soil, and the organic matter that it contains, is the ability to act as a living system. For this reason, Doran and Safley (1997) suggested that the term 'soil health' could usefully be employed to define 'the capacity of a soil to function as a living system within ecosystem and landuse boundaries, to sustain biological productivity, to promote the quality of air and water environments to sustain biological activity, and maintain plant, animal and human health'. Pankhurst (1997) suggested that soil 'health' can be distinguished from soil 'quality' on the grounds that it includes a measure of time and must involve an assessment of biological activity.

In order for the concept of soil health or quality to be useful, these concepts must be based on defined and measurable soil properties. Doran and Safely (1997) argue that the latter should be based on parameters that reflect the dynamics of the physical, chemical and biological functions of soil. The definition of threshold values of organic matter below which the soil cannot be managed sustainably is difficult using this approach. Different soil types contain widely different amounts of organic matter reflecting differences in soil genesis (among other things) at a given site. Loveland et al. (Chapter 1.1) carried out an extensive review of the literature that relates organic C to soil physical properties. They found that there was little consistent evidence to define critical thresholds of organic matter below which physical properties change significantly. The inevitable loss of organic matter following cultivation of native soils need not necessarily lead to permanent loss of function. Equilibrium between inputs and outputs can be achieved at a lower level of productivity leading to sustainable soil management, despite changes in land use.

It has been suggested that total soil organic matter may not be a good indicator of soil quality (Carter *et al.*, 1999), particularly as much of the total pool contains relatively inert physically and chemically stabilized fractions. Microbial biomass carbon represents one of the more labile pools and one that makes a critical contribution to nutrient flows, organic matter turnover and the structural stability of soil aggregates. The biomass is also highly responsive over short periods to changes in land management, and measurement of this pool can provide advanced warning of longer term changes to the overall organic matter fraction (Powlson *et al.*, 1987).

Janzen *et al.* (1998) describe a conceptual model in which organic matter accumulates during the process of soil formation and then reaches a steady state at which primary production equals loss of organic C by respiration. Cultivation of the soil results in a rapid but relatively short-lived loss of organic C followed by a new steady state. Further modification to the soils organic matter status may be introduced by deliberate measures taken to sequester carbon (Smith *et al.*, Chapter 4.13).

A new criterion in the sustainable management of organic matter comes from the need to reduce the build up of atmospheric carbon dioxide. The size of the potential sink that may be provided by soils for atmospheric carbon is now of considerable interest to scientists and policy makers. It is argued by Paustian (1997) that this provides a guide to the upper limits of carbon that potentially can be sequestered by agricultural soils. In many cases, this is quite modest, but there are large regional variations. It is probably in temperate regions where sophisticated techniques of agricultural management can be introduced rapidly (as a result of infrastructure and good extension services) where opportunities for further sequestration are greatest (Smith et al., Chapter 4.13). Powlson et al. (1998) reported that afforestation of 30% of the current agricultural land in Europe could lead to an increase of 43 Tg C year<sup>-1</sup> in the storage of carbon in soil, equivalent to 3.8% of anthropogenic emissions. If accumulation in woody biomass is included, the percentage of emissions consumed rises to 15.3%. Soil organic matter storage is also modified by the nature of tillage operations. Some interest has been shown recently in observations that deeper tillage operations resulting from the use of more modern machinery may be contributing to increased organic matter storage at depth (Soane and Ball, 1998). However, in some cases, it seems that tillage may simply lead to a redistribution of organic carbon without leading to an increase in total storage (Yang and Wander, 1999).

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# Organic Matter and Sustainability

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#### Introduction

For several decades, there has been a concern about soil organic matter and sustainability of the soil resource in intensive farming systems. Underlying these concerns is the recognition that soil organic matter plays an important, yet often poorly understood, role in sustaining soil functions. In the UK, several studies (MAFF, 1970; HMSO, 1994, 1996) have emphasized the need to understand better how soil organic matter functions in soil and how management of soil organic matter can be optimized to achieve sustainable farming systems (Box 1.1). Although difficulties still remain in

**Box 1.1.** Major concerns with regard to soil organic matter and sustainability as given in UK studies.

Improved methods to estimate soil organic matter.

Monitor soil organic matter concentrations over time.

Establish minimum soil organic matter concentrations for different soils below which adverse effects may occur.

Protect soil as a limited resource and as an essential part of life-support systems.

Ensure land management maintains soil functions by preventing irreversible declines in soil organic matter.

Soils should be given same priority in environmental protection as air or water.

Future surveys of land should provide a measure of soil biological quality.

Based on UK studies (MAFF, 1970; HMSO, 1994, 1996).

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defining 'sustainability', there is a consensus that soil organic matter has a significant role to play in the sustainability of farming systems (Swift and Woomer, 1993) and that it is an important indicator of soil quality and productivity (Larson and Pierce, 1994). Costanza *et al.* (1997) identified 17 key ecosystem services on a global scale, eight of which include soil as a critical component and most of which involve organic matter.

The objectives of this chapter are briefly to introduce some approaches used to characterize sustainability in agroecosystems and their implications for soil organic matter management; to address the concept of 'soil quality' as a framework to evaluate soil functions and its possible role in soil organic matter quality evaluation; and to examine the interrelationships between organic matter and other soil properties that regulate and control the functional purpose of organic matter in soil.

### Soil Organic Matter and Sustainable Land Management

Soils are part of a larger environmental system called 'land', which reflects the natural integration of soil, water, climate, landscape and vegetation characteristics (FAO, 1976; Carter et al., 1997). There are many and varied approaches used to define and assess sustainability (Farshad and Zinck, 1993). Smyth and Dumanski (1995) proposed five components that integrate environmental and socioeconomic principles that must be satisfied to attain sustainable land management (Box 1.2). Generally, these components are based on indicators of performance over time, with each indicator given a 'threshold' or critical value, which is associated with a significant decline or adverse change in land use sustainability. Gomez et al. (1996) illustrated the use of the above approach, based on threshold levels using soil organic matter as an indicator of natural resource protection, to evaluate the sustainability of several farms in the Philippines.

The sustainability components given in Box 1.2 can be reduced to three major areas: natural resource protection (including protection of adjacent ecosystems), economic viability (including productivity and security) and social acceptability (including natural use and aesthetic

Box 1.2. Components of sustainable land management (after Smyth and Dumanski, 1995).

Productivity - maintain and enhance production

Security - reduce level of production risks

Protection - protect the quality of natural resources and prevent soil degradation and conserve soil resource

Viability – maintain economic viability

Acceptability - be socially acceptable

quality). Although all components must be satisfied to meet the goal of sustainable land use, it is recognized that sustainability has a time scale and that sustainability for any specific land use is relative rather than indefinite (Smyth and Dumanski, 1995), and in most cases can only be evaluated from long-term experimentation (Reeves, 1997). Furthermore, the time scale may differ for each component. Even though loss of sustainability in any one component would classify the land use system as 'unsustainable', in reality a tension emerges as short-term economic viability can be sustained by inputs to the system even when the natural resource base has exceeded its sustainability time scale. This latter scenario has serious implications for soil organic matter and the sustainability of intensive farming systems, as most economic models utilized to describe sustainability in agriculture often disregard or marginalize the natural resource component (Farshad and Zinck, 1993).

Sustainability can also be approached using ecological models. Ecosystem components (e.g. plant biomass, consumer and decomposer organisms, and soil) are connected by ecosystem processes (e.g. production and decomposition), that in turn are regulated by driving variables such as climate, soil type (e.g. texture and mineralogy) and landscape. In intensive agroecosystems, the main driving variable, or regulator, is the farmer or society. In contrast to natural ecosystems, agroecosystems are 'open' and complex with major exports of primary production (i.e. harvest), and inputs of nutrients and energy, and have additional economical and sociological components with their own driving variables (e.g. demands of society for agricultural goods). In a sense, agroecosystems, in contrast to natural ecosystems, are 'super-systems' where management decisions are often dominated and controlled by the socioeconomic components. The dominance of the socioeconomic components may justify continuous outputs from a system, even when the ecological components of the system are failing, or excessive inputs (e.g. fertilizer) above that of the natural storage capacity of the system lead to pollution of adjacent ecosystems. Similarly to the observations stated above for sustainable land management, ecology and economics can be in conflict, leading to a decrease in soil organic matter and consequently an adverse affect on organic matter-dependent soil functions.

### **Concept of Soil Quality**

Soil quality involves placing value on a soil in relation to a specific use (Larson and Pierce, 1994; Carter *et al.*, 1997). The seminal idea involves categorizing the 'fitness of a soil for a specific use', but this basic concept is now enlarged to address those soil functions that allow the soil to accept, store and recycle water, nutrients and energy. The evaluation of soil quality

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is problematic. Unlike water and air where 'fitness of use' can be related directly to a single function (i.e. human consumption), soil has multiple functions and is only related indirectly to human welfare (Oliver, 1997). Furthermore, a significant part of soil quality (i.e. inherent quality) that involves some important soil properties and attributes (e.g. particle size distribution and mineralogy) are not readily subject to human manipulation and are relatively static. This aspect of soil quality has long been addressed by land resource and soil survey inventories. Other properties and attributes of soil quality (i.e. dynamic quality) that include, for example, organic matter and structural properties (e.g. porosity, permeability and aggregation), can be subject to relatively rapid change and responsive to soil management.

An ecological framework used to evaluate soil quality is given in Table 1.1. The framework is based on the following logical sequence: purpose, function, processes, properties (including critical values), indicators and methods (including standardization).

### Soil Organic Matter Quality Evaluation

The soil quality framework given in Table 1.1 can also be applied to evaluate those soil functions specifically related to soil organic matter. Generally, the limiting steps in the above sequential framework for soil organic matter evaluation are the setting of 'critical values', the choosing of suitable attributes or 'indicators', and 'standardization'.

Table 1.1.	Sequential framework to evaluate soil quality for specific purpose or fitness of use
(after Carter	et al., 1997).

Sequence steps	Sequential framework	Questions implied by the framework
1.	Purpose	What will the soil be used for?
2.	Functions	What specific role is being asked of the soil?
3.	Processes	What key soil processes support each function?
4.	Properties/attributes	What are the critical soil properties for each process? What are their critical or 'threshold' values?
5.	Indicators/surrogates/ pedotransfer function	When the attribute is difficult to measure or not available, which indirect or related property or properties can be used in its place?
6.	Methodology Standardization	What methods are available to measure the attribute? Technical rules and protocols for soil sampling, handling, storage, analysis and interpretation of data.

### Soil organic matter and soil functions and processes

At the agroecosystem, or 'field' scale, organic matter influences many readily measurable soil functions or processes (Schnitzer, 1991). Organic matter is both a source and a sink for plant nutrients, and provides an energy substrate for soil organisms. Soil macro- and microaggregation, that aid the infiltration of air and water, are promoted and stabilized by soil organic matter (Tisdall, 1996). Organic matter promotes water retention and influences the efficacy and fate of applied pesticides (Gregorich *et al.*, 1994, 1997). It also influences certain soil physical processes such as compactibility (Soane, 1990), friability (Watts and Dexter, 1998) and the range of soil 'available' water for plant growth (Kay, 1998). Overall, the positive interrelationship between soil organic matter and soil aggregation has important benefits on both water and air infiltration, soil erodibility and conservation of organic matter and nutrients (Feller and Beare, 1997).

### Soil organic matter properties, attributes and constituents

Gregorich et al. (1994) indicated that soil organic matter should be viewed as a set of fractions rather than a single entity. These fractions are descriptive of the 'quality' of soil organic matter. Important fractions of organic matter are the light fraction, macroorganic matter (i.e. particulate carbon), microbial biomass carbon, mineralizable carbon, carbohydrates and enzymes. These fractions have biological significance as they are involved in several soil functions and processes such as aggregation and formation of soil structure, and nutrient cycling and storage. Chemical characterization of organic matter, that provides information on chemical structure and functional groups, is also useful to evaluate the influence of land use changes on organic matter (Monreal et al., 1995; Mahieu et al., 1999). However, the utility of such measurements in soil quality evaluation is not so clear.

Soil biota are important soil constituents that are usually strongly associated with or, in some cases (i.e. microbial biomass carbon), components of soil organic matter. Measurements of soil biota abundance, diversity or activity are considered potential indicators of soil quality (Gregorich *et al.*, 1997). The microbial biomass is the main agent that supports the soil function and associated processes involved with the storing and cycling of nutrients and energy (Carter *et al.*, 1999). Mycorrhizal fungi play an important role in sustainable plant productivity and in the formation and maintenance of soil structural stability (Tisdall, 1996; Gregorich *et al.*, 1997), while soil fauna are major determinants of soil processes influencing nutrient cycling, aggregate formation and permeability of soil (Lavelle *et al.*, 1997).

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### Selecting indicators to assess soil organic matter change

Measurement of total organic matter concentration in soil at any one time is relatively straightforward; however, in many cases it is difficult to measure significant change in soil organic matter concentration, even if soil samples are obtained over time. Small, but important changes can be difficult to detect against the relatively large background concentration of soil organic matter.

To resolve this dilemma, some of the above-described soil organic matter fractions can be chosen that are more sensitive to change than total organic matter. For example, soil microbial biomass within specific limits can serve as an indicator of organic matter change (Carter et al., 1999). Other labile soil organic matter fractions, such as macroorganic matter and the light fraction, are highly responsive to changes in carbon inputs to the soil and will provide a measurable change prior to any such change in the total organic matter (Gregorich and Janzen, 1996). Campbell *et al.* (1997) and Bolinder et al. (1999) illustrated the sensitivity of several soil organic matter fractions to changes in carbon inputs. However, the validity of this approach to indicate the direction of soil organic matter change is restricted under conditions where climate impedes adequate carbon inputs (Mele and Carter, 1993) or suppresses the rate of decomposition (Janzen et al., 1998). In these latter situations, the change is characterized by a temporary increase in labile carbon and does not indicate a concomitant change in total soil organic matter.

### Critical values of soil organic matter

Critical values involve establishing a critical concentration or range for the property in question that is needed to ensure that dependent soil processes and function are not restricted or adversely affected (Larson and Pierce, 1994). Analogous to the 'critical loads concept' used to assess the influence of various pollutants in the environment (HMSO, 1996), the concept of 'critical values' is used commonly in soil pollution studies (Nortcliff, 1997).

Little attempt has been made to characterize critical values for soil organic matter. Limiting values or thresholds for fractions and components of soil organic matter are generally not known (Carter *et al.*, 1999). The non-uniform and multi-functional nature of organic matter ensures that the task of selecting a specific organic matter-mediated function and attempting to set a critical value would be problematic. However, some studies have been successful in this regard for total organic matter. For example, Feller *et al.* (1996) linked critical values of soil organic matter for both soil fertility and erodibility in tropical 'kaolinitic' (e.g. Alfisols, Ultisols and

Oxisols) and 'smectitic' (e.g. Vertisols) soils. A critical threshold of soil organic matter, based on a linear equation utilizing soil silt and clay content, was useful in predicting the sustained fertility and productivity of a collection of tropical kaolinitic soils (Feller and Beare, 1997), while a critical value of organic matter could be established that was associated with decreased runoff water turbidimetry for a Vertisol (Feller *et al.*, 1996).

Generally, critical values for total soil organic matter would be soil specific, developed using a range rather than a set value, and probably based on the most limiting soil function and process.

### Standardization of soil organic matter measurements

Standardization deals with the development and applications of technical rules, specification and protocols with regard to a measuring method (Nortcliff, 1997). At present, there is a need to develop acceptable standard sampling and measurement protocols to monitor and evaluate change in soil organic matter. In many cases, estimates of organic matter status and change are still based on carbon concentration rather than mass, and comparisons between treatments derived from unequal soil depth, densities or soil mass. Ellert and Bettany (1995) emphasize the importance of assessing management-induced changes in soil organic matter on an 'equivalent mass' or a per unit area basis. Such procedures provide some degree of standardization for soil sampling among soil management comparisons, but their interpretation may be problematic in situations where soil erosion has been dominant, or in ecological studies comparing differences in vegetation types.

In addition to sampling protocols, there is no well-accepted operational definition of soil organic matter (Agricultural Soils Working Group, 1999). Does a measure of soil organic matter include plant litter, crop residues or root material? For a wide range of study sites in eastern Canada in mainly Luvisols, Cambisols and Podzols, estimated annual straw and root carbon inputs of cereal crops (Bolinder *et al.*, 1997) ranged from 2 to 5% of the total (0–60 cm depth) soil carbon (Carter *et al.*, 1998). This range would probably be higher for grasses and legumes. Thus, accommodating crop-derived carbon in soil organic matter estimates has implications for soil handling protocols after sampling, such as sieve size.

## Regulation of Organic Matter Functioning in Soil

In agroecosystems, factors that regulate organic matter functioning in soil are related to organic matter additions or inputs that influence particulate or macroorganic matter, and the relationships between organic matter and 16 M.R. Carter

soil aggregates. The capacity of a soil to store organic matter is related to the association of organic matter with clay and clay plus silt (2–20  $\mu$ m diameter) particles, soil microaggregates (20–250  $\mu$ m diameter) and macroaggregates (> 250  $\mu$ m diameter), and the accumulation of sand-sized (> 50  $\mu$ m) macroorganic matter (Christensen, 1996; Tisdall, 1996; Feller and Beare, 1997; Hassink, 1997).

# Carbon inputs, macroorganic matter and organic matter functions in soil

Climate and soil type can significantly influence the accumulation and storage of soil organic matter due the interactions of temperature and moisture on plant productivity and the ability of the soil's mineral components to retain organic matter (Carter, 1996). Holding constant parent material and topography, the potential for soil organic matter accumulation increases with increasing precipitation and decreasing temperature (Cole *et al.*, 1993). Moist, warm or hot climates favour rapid soil organic matter decomposition relative to wet, cool climates.

Variable topography increases the potential for soil erosion which, in turn, may also influence the accumulation and storage of soil organic matter (Gregorich *et al.*, 1998). Although mineralization causes a direct loss of soil organic matter, soil erosion can affect organic matter concentration via removal, dilution and deposition processes.

Within any one soil type, increasing carbon inputs via agricultural management is the key to increasing soil organic matter quantity (Jenkinson, 1990). Where the initial concentration of organic matter is low, i.e. below the capacity of a specific soil to store organic matter, it has been observed that soil organic matter concentrations increase linearly with increasing input levels, although the slope of the line depends on climate, soil type and soil management (Parton et al., 1996). The latter variable influences carbon inputs mainly in the following three ways: increasing primary production (e.g. perennial crops, plant nutrition and organic amendments, Angers and Carter, 1996; Reeves, 1997; Janzen et al., 1998); increasing the proportion of primary production returned to, or retained by the soil (e.g. crop residue retention and placement); and influencing both microbe- and plantinduced changes in soil structure that can suppress the rate of decomposition through enhancing soil aggregation (Hassink et al., 1997; Angers and Caron, 1998). Under certain climates, placement of residues at depth can allow a relative increase in stored organic matter (Carter et al., 1998), while surface accumulation of crop residue and plant material can occur under conditions of desiccation or waterlogging. In addition to carbon inputs, vegetative differences and quality of crop residue can also influence the quantity of soil organic matter (Juma, 1993; Parton et al., 1996).

Addition of organic matter to the soil, in the form of crop residues or organic amendments, increases the concentration of 'free, low-density' macroorganic matter which can range from 10 to 40% of total soil organic matter (Carter *et al.*, 1998; Kay, 1998). This form of organic matter functions in improving the mechanical properties of soil, specifically in reducing soil compactibility (Soane, 1990), and enhancing the range of soil 'available' water for plant growth (Kay, 1998) and soil friability (Watts and Dexter, 1998). On the basis of this important role in organic matter functioning, it would be advantageous to establish critical ranges for macroorganic matter as it is sensitive to soil management, and easily measured and quantified.

# Soil aggregation, aggregated organic carbon and organic matter functions in soil

Soil type characteristics (e.g. mineralogy and particle size distribution) regulate the capacity of a soil to preserve organic matter and control soil aggregation. The interrelationship between soil aggregation and soil organic matter constrains both decomposition (e.g. separate carbon substrate from decomposer organisms) and predation (e.g. separate microbes from predators) processes and subsequently conserves and stabilizes soil organic matter (Juma, 1993).

Arable soils contain less organic matter than adjacent grassland soils, but the amount of organic carbon associated with the clay plus silt (i.e. 2–20 µm diameter) can be similar (Hassink, 1997). More than 80% of the organic carbon in temperate soils can be associated with < 20 µm diameter organomineral particles (Christensen, 1996). Once the clay plus silt is saturated with organic matter, additional organic matter would be found mainly in the sand-sized macroorganic matter fraction. Thus, grassland and forest soils that can contain relatively high concentrations of organic matter generally have more sand-sized organic matter than arable soils (Carter et al., 1998). In soils with low concentrations of sand fraction carbon, > 90% of the soil organic matter can be found in the clay plus silt particles (Christensen, 1996). Thus, silt plus clay carbon, being an inherent measure of the capacity of any one soil type to store organic matter (Hassink, 1997), provides a basic measure of aggregated organic carbon. In addition, silt plus clay carbon is easily estimated and quantified.

In most soils, the development of aggregation distributes organic matter into different sized aggregates with an increasing susceptibility to decomposition as follows: within clay plus silt particles, within microaggregates (i.e. intra-microaggregate), within macroaggregates but external to microaggregates (i.e. includes the light fraction, macroorganic and microbial biomass carbon) and 'free' macroorganic matter (Carter, 1996;

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Christensen, 1996). Table 1.2 illustrates the change in turnover time for soil organic matter fractions and organic matter in aggregates based on isotopic studies. The aggregation process itself is a means to both conserve organic matter and allow the stored organic matter to function as a reservoir of plant nutrients and energy. In addition, the role of organic matter in the aggregation process has major implications for the functioning of soil in regulating air and water infiltration. This interaction has allowed organic matter to be identified as an indicator associated with the processes of soil permeability and erodibility (Feller and Beare, 1997).

## **Conclusions**

The tension between natural resource and economic sustainability in agroecosystems, which has important consequences for conservation of soil organic matter, underlines the need to develop quantitative estimates of the value of the functions and services provided by organic matter. The soil quality framework provides a logical procedure to evaluate soil organic matter quality along the sequence of 'function' to 'methodology', although at present the question of 'critical values' and 'standardization' are limiting steps in this evaluation.

Organic matter inputs and the soil aggregation process are important factors in the maintenance and regulation of organic matter functioning in soil. Both the sand-sized macroorganic matter and silt plus clay carbon are useful and easily measured indicators.

**Table 1.2.** Estimates of turnover time for soil organic matter in different fractions and in soil aggregates (after Carter, 1996; Gregorich and Janzen, 1996; Collins *et al.*, 1997; Monreal *et al.*, 1997).

Type of organic matter	Estimated turnover time (years)	
Organic matter in fractions		
Litter, crop residue	0.5–2	
Microbial biomass	0.1-0.4	
Macroorganic matter	1–8	
Light fraction	1–15	
Organic matter in aggregates		
Non-aggregated soil	1–7	
Macroaggregates <sup>a</sup> (> 250 μm diameter)	1–23	
Microaggregates (20–250 μm diameter)	3–80	
Silt plus clay (< 20 µm diameter)	5–1000	

<sup>&</sup>lt;sup>a</sup>Organic matter in macroaggregates but external to microaggregates (i.e. inter-aggregate).

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# Critical Levels of Soil Organic 1.1 Matter: the Evidence for England and Wales

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## Introduction

There is a widespread perception that the amount of organic matter (SOM) or organic carbon (SOC) in soils needs to be kept above a minimum level in order to prevent, or at least minimize, irreversible decline in a range of soil properties. Soil structure, ease of cultivation, improved water retention, better seedbed establishment, reduced erosion risk, more effective use of nutrients (especially N and P) have all been commented on (see, for example, Carter and Stewart, 1996), although it is known that, in certain soils, other factors such as the 'free' iron content and exchangeable sodium content may have a marked effect on aspects of soil structural behaviour. Greenland et al. (1975) proposed a 'rule of thumb' that soils in England and Wales should be regarded as structurally unstable if the SOC content fell below 2%; in conventional terms, this equals 3.4% SOM. This value seems to have become a reference point or benchmark in the soil science community. The UK Royal Commission on Environmental Pollution, in its report on 'Sustainable Use of Soil' (RCEP, 1996), commented on the undesirability of allowing SOM to decline too far, although avoiding a specific recommendation as to any limiting values. The RCEP report thus raised some fundamental questions in relation to SOM in the soils of England and Wales and the long-term, sustainable use of UK soils for agricultural production, against a background of the need to manage soils in an environmentally sensitive manner. We sought to address three of these questions: what is the quantitative evidence for critical levels of SOM in relation to specific soil properties; if such critical levels can be 24 P.J. Loveland et al.

substantiated, how do they differ between soil types and management practices; and what is the magnitude of any decline in soil properties if these critical levels are not maintained?

## The Evidence

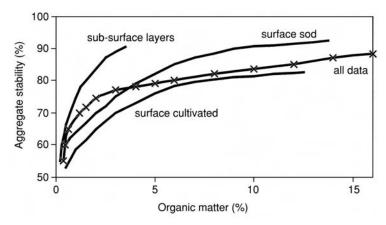
There are thousands of papers, reports, etc. which assert that a change in SOM status is associated with an improvement or deterioration in the behaviour of agricultural soils. However, assertion is not enough, nor should it be enough – against a background of increasing regulation based on quantitative approaches – to say that soil scientists know this to be true. We were seeking robust, numerical evidence to support these assertions for soil types relevant to the UK, e.g. equations of state, regression relationships and graphical demonstration of relationships, supported by properly designed experiments with acceptable statistical treatment. We were also seeking work in which the authors were careful in the use of terms such as SOM, SOC, 'humus', and the like, and not using these interchangeably without adequate explanation. We examined about 1500 such papers (back to ~1938), whose abstracts led us to believe that the paper contained quantitative evidence relevant, although not exclusively so, to temperate agriculture and soils types in the UK.

The vast majority of the papers examined, despite the claims to the contrary, did *not* contain quantitative data which met our criteria for robustness, good experimental design or careful use of terminology (often all of these).

Of those papers which did contain 'hard' evidence, the majority were concerned with aggregate stability. Apart from the difficulties about SOM versus SOC, etc., the literature suffers from different definitions of aggregate size, and a plethora of methods for determining aggregate strength or stability, with almost no inter-method or inter-laboratory comparisons. This is clearly an area which would benefit enormously from some standardization. There is an increasing move in the more modern literature to quote aggregate size in multiples of 250  $\mu$ m, although many papers quote aggregate size in terms of the *mean weight diameter* (MWD), which does not give aggregate size directly.

Probably the most comprehensive investigation of the relationship between *total* SOM and aggregate stability is still that of Kemper and Koch (1966), who examined 519 samples of topsoils and sub-soils from the western USA and Canada. They derived a number of curves showing that aggregate stability declined increasingly sharply as SOM contents fell below ~4% (Fig. 1.1.1). It is interesting to note that the rate of decline is steepest when all the data are combined (and does seem to give some credence to the 2% threshold, albeit for SOM, not SOC), but is less marked when the

Critical Levels of SOM 25



**Fig. 1.1.1.** The relationship between aggregate stability and soil organic matter content in 519 soil samples from the western USA and Canada (redrawn from Kemper and Koch. 1966).

soils are split by land use and depth. The authors also investigated the influence of 'free' Fe oxides and exchangeable Na on aggregate stability. The effects of these components was not large, as demonstrated in the relationships found:

```
% water
                     49.7 + 13.7 \log(OM\%) + 0.61 (Clav\%) - 0.0045
                     (Clay\%)^2 + 9.0 (Fe_2O_3\%) - 1.6 (Fe_2O_3\%)^2 -
stable
                     0.28 \text{ (ES\%)} - 0.06 \text{ (ES\%)}^2 \text{ [31\% variance]}
aggregates
(all soils)
                     explained]
% water
                           40.8 + 17.6 \log(OM\%) + 0.73 (Clay\%) -
                           0.0045 \text{ (Clay\%)}^2 + 3.2 \text{ (Fe}_2O_3\%) \text{ [}44\% \text{ of}
stable aggregates
(arable topsoils)
                           variance explained]
% water
                           45.1 + 22.6 \log(OM\%) + 0.28 (Clay\%) -
stable aggregates
                           0.0021 \text{ (Clay\%)}^2 + 1.55 \text{ (Fe}_2O_3\%) [38\% \text{ of}]
(grass topsoils)
                           variance explained]
```

It is interesting to note that the explanation of the variance in aggregate stability was slightly greater for arable soils, i.e. those with smaller amounts of SOC, than in the permanent grass soils. This is not something which has been noted often. Kemper and Koch (1966) invoke a number of possible mechanisms to explain the different behaviour of their three soil groups, including qualitative differences in SOM/SOC related to land use history, the kinds, amounts and distribution of clay particles, and whether the soils had been 'disrupted', i.e. ploughed, or not. However, they were unable to differentiate clearly between these effects.

Douglas and Goss (1982), however, found that the amounts of organically bound Fe and Al (although generally < 0.2%) contributed

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significantly to the aggregate stability of Hamble, Lawford and Denchworth soils in southern England, but not to that of the calcareous Andover soil (a Rendzina). The latter is not surprising as the stabilizing effect of high calcium values on soil structure is well known.

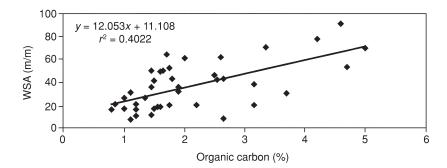
In an attempt to derive a general classification of the structural stability of soils in England and Wales, Greenland *et al.* (1975) examined 180 soil samples from a very wide range of soils. They summarized their work as:

- Soils < 2% organic carbon: unstable
- Soils 2–2.5% organic carbon: moderately stable
- Soils > 2.5% organic carbon: stable

This classification illustrates a little considered point in all such studies, in that it represents a relatively narrow range of values for the critical limits, but the methods of determination of SOC in common use (mostly based on acid dichromate oxidation of the carbon) are *relatively* imprecise (a variation of 0.1% in a determination of 2% SOC is not uncommon).

The description of soil structural relationships by power functions, as in Kemper and Koch (1966), is not common. Much more typical is that found by Stengel *et al.* (1984) for a wide range of British soils (Fig. 1.1.2), in texture classes ranging from sandy to clayey, and with and without free calcium carbonate present.

A study of some New Zealand soils by Haynes and Swift (1990) illustrated a point originally made by Emerson (1954), that the physical behaviour of soils is strongly influenced by the initial treatment of, for example, the aggregates. Figure 1.1.3 shows that the behaviour of the field moist aggregates is best described by a linear relationship, whereas the behaviour of the air-dried aggregates is best described by a power function. This again highlights the contrast between different studies; most find that the behaviour of air-dried aggregates is best described by a linear relationship



**Fig. 1.1.2.** Relationship between soil organic carbon (%) and water-stable aggregates (WSA mass/mass) in samples from Ashley, Hamble, Wotherstone, Fincham, Hanslope, Wicken, Denchworth, Lawford, Newchurch, Newport and Andover soil (redrawn from Stengel *et al.*, 1984).

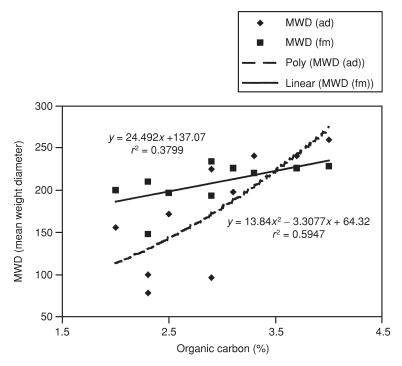
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and that of moist aggregates by a power function, whilst others find the opposite. We have been unable to establish clearly why this should be so.

Not all work has been carried out solely on aggregate stability. An equally important soil property is bulk density (Db). Williams (1971) studied 189 British soils, and found the following relationships:

Db (arable topsoils) = 1.42-0.78(total N%) [38% variance explained] Db (grass topsoils) = 1.37-0.76(organic carbon %) [69% variance explained]

The identification of total nitrogen as the most important factor in the arable topsoils is surprising, given that it is usually strongly related to total SOC. Total N may be more of a reflection of fertilizer regimes in arable soils than in grassland soils. There is no obvious mechanism by which inorganic N should affect Db in this way, unless it is through stimulation of mechanisms such as fungal growth, the hyphae of which often have been implicated in soil structural development (Tisdall, 1991). More recently, Cannell *et al.* (1994) found a very strong linear relationship between SOM



**Fig. 1.1.3.** Relationships between organic carbon (%) and aggregate stability expressed as mean weight diameter (MWD) of air-dried (ad) and field-moist (fm) aggregates for some soils from New Zealand (drawn from data given by Haynes and Swift, 1990).

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and Db in a cultivation experiment extending over 18 years and a range of tillage practices:

Db = 
$$-0.102(SOM\%) + 2.07 (r^2 = 0.93; P < 0.001)$$

This reinforces a major point in these investigations. With the exception of the study by Kemper and Koch (1966), there is little or no evidence for sudden or increasingly marked change in soil properties in relation to SOM or SOC. The relationships are generally linear, and do not indicate a catastrophic change; or the possibility of identifying clearly whether there is a critical value of SOM or SOC for a specific soil type, whether or not in relation to a specific land use.

Investigations by Hollis *et al.* (1977) of the relationships between SOC and volumetric water content (THV5) at –5 kPa tension in topsoils of soils from the West Midlands of England gave the relationship below for all land uses:

THV5 = 
$$23.88 + 7.85$$
 (OC%) –  $0.43$  (OC%)<sup>2</sup> [ $n = 77$ ; 73.5% variance explained]

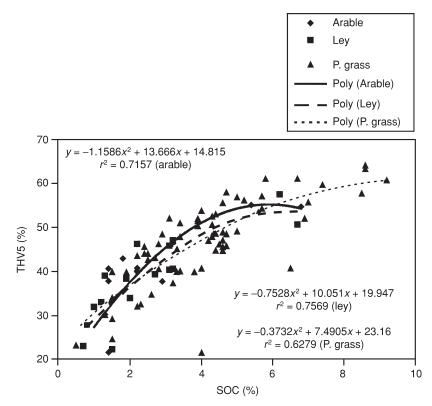
Expansion of this data set (n = 99), and its subsequent stratification according to land use, is illustrated in Fig. 1.1.4. The main point is that expansion of the data set does not greatly affect the form of the relationship, although this clearly differs slightly between land uses. However, the differences between the land use groups are clearly small and are not great enough to suggest significantly different soil behaviour, or a marked difference in the contribution to volumetric water content from SOC.

We have investigated the contribution of SOC and other soil factors, for a larger number of topsoils, to the soil volumetric water content at different tensions. The data are summarized in Table 1.1.1.

It can be seen that SOC explains ~13% of the variance at low tensions, falling to 1% at wilting point. Examination of similar data for sub-soils shows that SOC contributes < 2% to the variance of volumetric water content at all tensions. Stratification of the topsoil data set in terms of clay content and major agricultural land uses is summarized in Table 1.1.2.

This table illustrates the complex nature of these relationships, even where relatively large amounts of data are available for analysis. It also highlights a further difficulty. As already said, SOM or SOC do not represent a single entity, but a complex mixture which changes in time and space. Clay content also represents a very wide range of mineralogy, which influences soil water release and retention in complex ways. There are additional factors which, although we cannot demonstrate their effects numerically, will also contribute to soil behaviour. Fe and Al have been mentioned, but calcium carbonate content, the composition of the exchangeable cation population and their interactions are others – many of which we have insufficient data for.

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**Fig. 1.1.4.** The relationship between volumetric water content at –5 kPa tension (THV5), and soil organic carbon content (SOC%) for West Midland topsoils under three agricultural land uses (expanded from Hollis *et al.*, 1977 using unpublished SSLRC data).

In terms of the long-term sustainability of soil, it is important to consider the magnitude of change in SOC over time. Data from the National Soil Inventory (McGrath and Loveland, 1992) and subsequent re-investigation at the same sites (DETR, 1999, p. 212), has shown that there is a significant change in the distribution of sites in the organic carbon groups for both arable/ley grass sites and for permanent grass sites (Table 1.1.3). The absolute magnitude of the change in the mean value of OC in the two data sets is 0.5% (arable/ley grass) and 0.8% (permanent grass) during ~15 years. Thus, whatever one might derive from the evidence in terms of critical levels of SOC/SOM, the need to consider change in these levels is likely to arise at decadal intervals.

The work reviewed thus far is in terms of *total* SOM or SOC. Tisdall and Oades (1982) summarized a considerable amount of earlier work, which had shown that 'active' or 'fresh' components of SOM were at least, or sometimes more, important in controlling soil physical behaviour than

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Table 1.1.1.	Contribution of different factors to volumetric soil water content (V%) of A horizons
of mineral soil	s at different tensions (in kPa).

	%Variance explained					
Factor	Clay (< 2 μm)	+ Silt (2-60 μm)	+ Organic carbon	No. of samples		
V% (-5 kPa)	44.8	52.4	65.7	652		
V% (-10 kPa)	51.4	60.5	69.3	597		
V% (-40 kPa)	60.8	70.2	73.8	597		
V% (-200 kPa)	64.6	70.8	72.4	516		
V% (-1500 kPa)	72.0	74.3	75.3	652		

**Table 1.1.2.** Relationship between soil volumetric water content at -5 kPa tension (V%) and other soil properties (soil organic carbon = SOC%; clay content = C%; silt content = Z%) in A horizons of mineral soils in England and Wales stratified by clay content and land use (SSLRC data, unpublished).

Land use	Clay content	Equation explaining greatest proportion of the variance in V%	% Variance explained	No. of observations
Arable	> 35%	V% = 32.46 + 0.30C	39	45
	Between 18 and 35%	V% = 23.43 + 0.26C + 0.11Z + 1.28SOC	36	109
	< 18%	V% = 18.58 + 0.35C + 0.12Z + 2.28SOC	56	101
Ley	> 35%	V% = 9.20 + 0.51C + 0.32Z	59	14
grassland	Between 18 and 35%	V% = 33.34 + 2.32SOC	25	44
	< 18%	V% = 20.71 + 0.21Z + 3.12SOC	56	48
Permanent	> 35%	V% = 45.8 + 0.77SOC	20	55
grass	Between 18 and 35%	V% = 32.12 + 0.10Z + 2.13SOC	31	125
	< 18%	V% = 25.68 + 0.08Z + 0.49C + 1.30SOC	27	58

total SOM, some or much of which could be 'inert'. In summary, they concluded that:

- only part of SOM stabilizes aggregates;
- above a certain amount of SOM, there is no further stabilizing effect;
- SOM itself is not a major binding agent;
- disposition of SOM can be more important than amount; and
- particle arrangement is more important in some soils than the absolute amount of SOM.

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	Arable/ley gr	ass $(n = 899)$	Permanent grass (n = 767	
% Organic carbon	1980	1994	1980	1995
< 2	317	350	43	27
2–4	387	444	313	404
4–6	121	66	231	246
> 6	74	39	180	90
$\chi^2$ value with 3 df	31.2		45	5.6
	P < 0.0001		P < 0.0001	

**Table 1.1.3.** Number of National Soil Inventory samples in soil organic carbon classes (% in < 2 mm oven-dry soil) in different years.

There are relatively few data for 'active' carbon for soils in England and Wales. Griffiths and Jones (1965) and Griffiths and Burns (1972) found strong evidence that microbially produced polysaccharides contributed considerably to soil strength, whilst Reid and Goss (1980) attributed similar increases to 'root exudates'.

None of this work found a relationship between the increase in soil strength and the initial amount of total SOC, or the ratio between total SOC and the amount of polysaccharide/exudate. More recently, Ball *et al.* (1996) have investigated the effects of differences in soil carbohydrate chemistry in relation to soil structure, but the data are insufficient to draw firm conclusions in a national context.

## Conclusions

From the forgoing analysis, we have concluded that:

- there is little *consistent* evidence that there are critical thresholds of SOC above or below which soil physical properties change significantly;
- there is a growing body of evidence that the 'active' fraction of SOM is
  a more important factor in controlling change in soil properties than
  total SOM, but there are insufficient data for the amounts and kind of
  'active' SOM for soils in the UK to enable firm conclusions about the
  magnitude of its affect on soil behaviour;
- there are insufficient data to explore thoroughly the relationships and interactions between other factors which affect soil physical behaviour;
- it would be inappropriate to set thresholds for critical levels of SOM or SOC for particular soils; and
- a single value for a critical level of SOM cannot be set for the soils of England and Wales on the basis of the data available.

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# FT-IR Studies on Soil Organic Matter from Long-term Field Experiments

1.2

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#### Introduction

Soil organic matter (SOM) is affected by the type of land use which, in turn, determines the organic matter turnover, and the water, and nutrient cycling. Körschens and Müller (1996) found significant correlations between management practice and crop yield as well as organic carbon  $(C_{org})$  and total nitrogen  $(N_t)$  content in the soil of a 100-year-old fertilization experiment. In addition to the SOM content, the SOM composition can be affected by the type of land use and soil management practices. In this chapter, SOM composition is defined as the variety and quantity of functional groups (e.g. carboxyl and hydroxyl groups). These functional groups play an important role in the sorption of solutes in the soil, such as plant nutrients or contaminants (Stevenson et al., 1982). Fourier transform infrared (FT-IR) spectroscopy can be used for the characterization of soil organic matter (Stevenson, 1982). Celi et al. (1997) showed that the content of carboxyl groups of SOM can be determined by FT-IR spectroscopy via integrating the area below the absorption band of the C = O group at 1700 cm<sup>-1</sup>. For a long-term field experiment located on a sandy soil, the contents of the carboxyl and hydroxyl groups in the sodium pyrophosphate extracts of soils from plots fertilized with cattle manure were found to be higher than in those from plots that received straw + mineral nitrogen (Ellerbrock et al., 1999a). The hot water-extractable SOM from a sandy site was found to be not significantly influenced in composition by the type of fertilization (Ellerbrock et al., 1999b). This was assumed to be caused by the relatively low application rate of organic fertilizer (32 dt ha<sup>-1</sup> year<sup>-1</sup>). In FT-IR Studies on SOM 35

order to test our hypothesis that higher organic fertilizer application rates will affect the composition of hot water-extractable SOM, we included a long-term field experiment on a sandy site which had received 1.6 times more organic fertilizer than the one at Müncheberg. Stevenson (1982) speculated that the effect of SOM on soil properties may be smaller in a loamy than in a sandy soil. Soil properties, such as structure, water storage capacity or cation exchange capacity (CEC), may therefore be affected more by fertilization via changes in SOM in a sandy soil than in a loam. Therefore, an additional objective of this study was to investigate whether differences in SOM composition due to fertilization could be found in soils with higher clay contents.

#### Materials and Methods

## Long-term field experiments

A detailed description of the field experiments can be found in Körschens *et al.* (1990). Some site and soil characteristics are listed in Table 1.2.1. The fertilizer treatments were as follows: no organic manure and no nitrogen fertilizer (control), mineral nitrogen (N), cattle manure with no mineral nitrogen (FYM) and cattle manure with mineral nitrogen (FYM + N). Table 1.2.2 gives the mean annual input rates of organic matter (excluding stubble and roots) and nitrogen fertilizer for the different sites. Soil samples were taken at 0–25 cm depth using an auger. The soil samples from each plot were air dried, and sieved to pass 2 mm.

# Chemical analyses and spectroscopy

Organic carbon content ( $C_{\rm org}$ ) was analysed by elemental analysis (LECO, CNS 2000) according to DIN ISO 10694 (1994). The cation-exchange capacity was determined according to DIN 19684 part 8 (1977). SOM was extracted in two ways:

- 1. Hot water: 10 g of the soil were mixed with  $0.1~\text{dm}^3$  of distilled water and heated under reflux for 1 h according to Körschens et al. (1990). The solid residue was separated by centrifugation followed by a membrane filtration (0.45  $\mu$ m, Nytran from Schleicher&Schüll®). Ions were removed from solution by dialysis, then the solution was freeze dried.
- 2. Sodium pyrophosphate solution: 10 g of soil were mixed with  $0.1 \,\mathrm{dm^3}$  of  $0.1 \,\mathrm{M\,Na_4P_2O_7}$  solution and shaken for 6 h at room temperature (Hayes, 1985). The solid residue was separated by centrifugation. The remaining solution was adjusted with 1 M HCl to pH = 2, to precipitate the SOM. After 12 h, the precipitation was completed and the mixture was

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Location	Established in	Clay (g kg <sup>-1</sup> )	Silt (g kg <sup>-1</sup> )	Sand (g kg <sup>-1</sup> )	Soil type (FAO, 1990)	pH range
Bad Lauchstädt						
Loamy soil	1902	211	677	212	Haplic Chernozem	5.3-6.5
Müncheberg						
Sandy soil	1963	50	210	740	Podzoluvisol to Arenosol	5.4–5.8
Groß Kreutz						
Sandy soil	1967	50	220	730	Albic Luvisol/ Luvisol Arenosol	5.0-6.5

**Table 1.2.1.** Site and soil characteristics of the long-term field experiments.

**Table 1.2.2.** Mean annual input rates of farmyard manure dry matter (FYM<sub>dm</sub>) and nitrogen (N) (kg ha<sup>-1</sup> year<sup>-1</sup>) at Bad Lauchstädt, Müncheberg and Groß Kreutz (Körschens *et al.*, 1990).

	Bad Lau	chstädt	Müncheberg		Groß Kreutz	
Fertilizer combination	FYM <sub>dm</sub>	N	FYM <sub>dm</sub>	N	FYM <sub>dm</sub>	N
Control	0	0	0	0	0	0
N	0	155	0	150	0	200
FYM	75	385	32	64	50	100
FYM + N	75	520	32	214	50	300

centrifuged. The precipitate was washed free of salt with distilled water and freeze dried.

FT-IR spectra were obtained using a BioRad®, FTS 135. A KBr technique (2 mg sample/150 mg KBr) (Celi *et al.*, 1997) was used to measure the absorption spectra of SOM extracts.

## Results and Discussion

In accordance with Körschens and Müller (1996), the  $C_{\rm org}$  content of the studied loamy and sandy soil increases with fertilizer input. The highest organic carbon content was found for the FYM + N fertilization and the lowest for the soil of the control plot for both experiments (Table 1.2.3). The difference in the  $C_{\rm org}$  contents due to the fertilization regime is lower for the sandy than for the loamy soil. This can be explained by the fact that the fertilizer application rate in the sandy soil is half that in the loamy soil. The clay content of the soils may have an additional effect on the  $C_{\rm org}$ 

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	Bad La	uchstädt	Münch	neberg	Groß l	Kreutz
Fertilizer	$C_{org}^{}}$	CEC	$C_{org}$	CEC	$C_{org}$	CEC
Control	16.6	209.6	4.3	32.5	4.2	33.9
N	19.3	208.7	4.69	32.0	5.2	36.4
FYM	24.1	219.4	4.63	31.5	5.5	40.0
FYM + N	27.0	223.8	4.9	35.6	6.8	46.2

**Table 1.2.3.** C<sub>org</sub> content (g kg<sup>-1</sup>) and CEC (mmol<sub>c</sub> kg<sup>-1</sup>) of the different plots.

content (Table 1.2.3). According to Stevenson (1982), the ability of SOM to form organomineral complexes increases with the clay content. These complexes protect the organic material against mineralization resulting in higher  $C_{\rm org}$  contents. Therefore, the  $C_{\rm org}$  content of the studied soils corresponds to the fertilizer application rate as well as the clay content. For the sandy soil at Müncheberg, the fertilizer regime had no significant effect on  $C_{\rm org}$  content. This may be as a result of the relatively low fertilizer application rate at this site (Table 1.2.2).

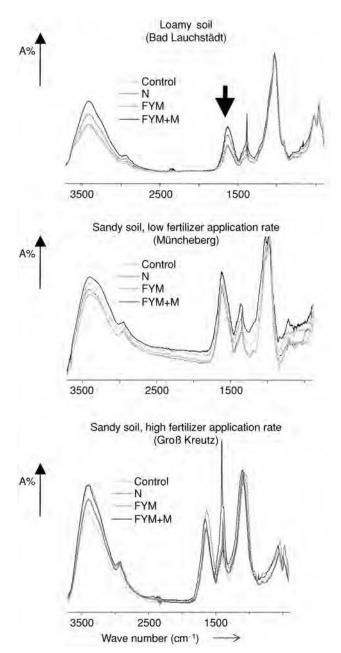
The CEC values of the loamy soil are 5–6 times higher than those of the sandy soils. This is due to the higher clay and organic carbon content of the loamy soil (Tables 1.2.1 and 1.2.3) because CEC depends on clay and SOM content. The CEC for the loamy soil samples increases in the sequence: control < N < FYM < FYM + N. For the sandy soil from Groß Kreutz, the CEC increases in the same sequence, but the differences in CEC due to the type of fertilization are most distinct for the samples from the loamy soil. This finding is also correlated with the differences in  $C_{\rm org}$  content.

Figure 1.2.1 shows the FT-IR spectra of the hot water extracts of the soils from the studied long-term field experiments. The spectra are normalized on the C–O–C band at  $1000~\rm cm^{-1}$ , which shows the same absorption intensity for all samples. All spectra show an overall scheme, but there are differences in the absorption intensity especially of the C = O band at 1710 and  $1690~\rm cm^{-1}$ , and the O–H band at 3500 cm<sup>-1</sup>. As Celi et al. (1998) show that the absorption intensity of the C = O group correlates with the CEC of the samples, we focus the discussion on this absorption band. A comparison of the FT-IR spectra of the hot water extracts of the soil samples from the loamy soil experiment (Fig. 1.2.1) show that the intensity of the C = O bands (representing carboxyl groups) decreases in the sequence: FYM + N > FYM > N > control. For the samples from the sandy soils with different fertilizer application rates, the sequence was FYM > FYM + N > N > control.

<sup>&</sup>lt;sup>a</sup>M. Körschens, Bad Lauchstädt, 1999, personal communication.

The analytical error was  $\pm$  0.5 g kg<sup>-1</sup> for C<sub>org</sub> and  $\pm$  5 mmol<sub>c</sub> kg<sup>-1</sup> for CEC.

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**Fig. 1.2.1.** FT-IR spectra of the hot water extracts of different fertilized soils from the long-term field experiments located at Bad Lauchstädt, Müncheberg and Groß Kreutz, after correction with the mineral phase.

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However, differences in absorption intensity are not as distinct as in the samples with the low application rate. We hypothesize that, because of the lower input of organic fertilizer at Müncheberg (32 dt ha<sup>-1</sup> year<sup>-1</sup>), mineralization prevents a significant increase in  $C_{\rm org}$  content in the fertilized soils at this site compared with the sandy site at Groß Kreutz with a higher FYM<sub>dm</sub> application rate (50 dt ha<sup>-1</sup> year<sup>-1</sup>). The absorption intensity of the C = O band in the FT-IR spectra of the sodium pyrophosphate extracts indicates a similar effect of the type of fertilization on SOM composition as shown for the spectra of the hot water extracts.

The absorption intensity at 1710 and 1690 cm<sup>-1</sup> correlates with the CEC of the SOM extracts. A high absorption intensity indicates a high CEC value. As the SOM extracts of the samples from the FYM + N plots show the highest absorption intensities at 1710 and 1690 cm<sup>-1</sup>, we expected the highest CEC values for these samples. This corresponds to the finding that the soil samples of the FYM + N plot of all long-term field experiments show the highest CEC (Table 1.2.3) due to differences in SOM composition. However, the information from the spectra is only partially representative for the CEC of the soil samples as the extracts represent certain parts of SOM, only.

However, the differences in the absorption intensities of the hot water extracts are higher for the loamy soil than for the sandy soils. A possible explanation is that at the loamy soil site a higher amount of farmyard manure (Table 1.2.2) was applied than at sandy soil sites. This results in a higher C<sub>org</sub> content in the loamy soil (Table 1.2.3) and may correspondingly lead to a higher content of carboxyl groups also in the hot water extracts. It may also be possible that the hot water-extractable SOM is protected against mineralization by the higher clay content at the loamy soil (Stevenson, 1982). However, for the sodium pyrophosphate extracts, the reverse is found. This may be caused by the higher clay content in the loamy soil. SOM seems to be protected against extraction due to complexation with clay minerals.

# Conclusions

The fertilization regime influences the  $C_{\rm org}$  content, CEC and SOM composition of all studied long-term field experiments. Relatively low input of organic fertilizer, 32 dt year<sup>-1</sup> for the sandy soil at Müncheberg, results in non-detectable differences in  $C_{\rm org}$  content and CEC between the different fertilized soils. The characterization of SOM by FT-IR spectra of sodium pyrophosphate and hot water extracts shows that the composition of SOM from the loamy and sandy soils was affected by the type of fertilization and the amount of organic fertilizer. Comparing the long-term field experiments from a loamy and a sandy soil, we conclude that the

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input of organic fertilizer in the sandy soil has an effect on SOM quality comparable with that in the loamy soil. Consequently, the composition of the extracted SOM depends on the type of fertilization and is here independent of the texture. However, the differences between fertilizer treatments in the pyrophosphate and hot water extracts were relatively smaller in the loamy soil compared with the sandy soils. The higher clay content in the loamy soil may protect hot water-extractable SOM against mineralization and sodium pyrophosphate-extractable SOM against extraction. The results also suggest that for soils with higher clay contents, improved SOM extraction methods are required.

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# Modelling Soil Organic Matter Dynamics – Global Challenges

2

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#### Introduction

Soil organic matter (SOM) modelling is gaining recognition as a key part of efforts better to understand, and manage, the terrestrial carbon cycle. The influence of the terrestrial carbon cycle on greenhouse gas emissions and uptake, and the potential feedbacks on long-term climatic change, have become front page news. Major questions with far-reaching societal impacts include: what is the current role of soils as a source/sink for increased atmospheric CO<sub>2</sub>?; will climate change increase CO<sub>2</sub> emissions from soil, resulting in a positive feedback to drive global warming?; and can soils be managed effectively to mitigate increased greenhouse gas loading? Answers to these questions require the application of models to quantify and predict SOM dynamics as a function of environmental factors and human management. Consequently, SOM modelling has played a prominent role in much of the recent research on climate change and the global carbon cycle (e.g. King et al., 1997; Field and Fung, 1999; Schimel et al., 2000).

The requirements for an accurate accounting of greenhouse gas emissions and sinks is central to the UN Framework Convention on Climate Change (FCCC) which requires national reporting of all major sources and sinks, including soils. Modelling will play a key role in integrating the variability and dynamics of soils, climate, topography and land use (as they affect SOM) to provide national estimates of soil C stock changes (e.g. Eve *et al.*, 2000; Tate *et al.*, 2000). SOM models have been used to estimate the potential for soils to sequester carbon and thus mitigate

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 $CO_2$  increases at national and regional levels (Dumanski *et al.*, 1997; Smith *et al.*, 1997a). If soil sinks are included under the Kyoto protocol, methods to quantify and verify changes in soil C at local, regional and national levels will be required. The potential for national and international trading of carbon emission offsets, making soil carbon a commodity, will necessitate that carbon changes be quantified to known levels of uncertainty. SOM modelling can play a central role in such quantification systems, but it will require a new level of integration with field measurements and other data on environmental and management factors.

Several of the SOM models currently in use today were first developed in the late 1970s and early 1980s (e.g. Jenkinson and Rayner, 1977; Parton et al., 1983). Since then, many new models have been developed (see reviews by Jenkinson, 1990; Paustian, 1994; McGill, 1996) and older models have continued to evolve on a more or less continuous basis. Despite their diversity, most of these models share some basic assumptions which include the representation of SOM as multiple pools (or as a quality spectrum) with differing inherent decomposition rates, governed by first-order rate constants modified by climatic and edaphic (e.g. soil physical attributes) reduction factors. Most of these models were conceived originally to describe processes at the ecosystem or field scale. New approaches continue to be explored, and some of the major recent trends in SOM modelling and their application to environmental problems will be discussed below.

#### Collaborative Networks

A significant recent development in SOM modelling has been the increased level of formal and informal collaborations between different modelling groups and between modellers and experimental scientists. Whether this has occurred as a result of the greater international collaboration spurred by global change research, or simply as a result of increasing ease of communication and the maturation of the science, is perhaps not important. Whatever the case, such collaboration has facilitated the comparison and evaluation of different models and approaches, such as a recent workshop in which a number of SOM models from around the world were brought together and evaluated using a common set of long-term experimental data from a variety of climatic, soil and land use conditions (Powlson et al., 1996; Smith et al., 1997b). Such comparisons help to clearly identify the difference in structure and assumptions of existing models (Paustian, 1994; McGill, 1996) and their relative strengths and weaknesses for a particular set of circumstances. These developments mirror similar recent efforts to conduct cross-model evaluations for water quality modelling, trace gas modelling and general circulation climate modelling, to name a few examples.

Long-term field experiments have been a mainstay for the development and evaluation of soil organic matter models (Powlson, 1996). Since soil organic matter levels change relatively slowly against a large and variable background level, long-term experiments with well-documented management histories are well suited to elicit and detect changes. Many SOM models originally were developed and tested using such experiments, but often for a restricted set of ecosystem, soil or climate conditions. However, with the increased use of models for regional, national and even global applications, testing of models to determine their generality (or lack thereof) over the entire domain of their application becomes crucial. This is particularly true given that these models are used increasingly for predictive purposes to guide policy-making and management decisions. The recent compilation of many of the long-term experiment data sets for the US and Canada (Paul et al., 1997) and Europe (Powlson et al., 1998) have helped to address this need by providing data, in standardized formats, which can be used to test and validate models for broad geographic regions and multiple types of land use, management, soil and climate conditions. A formal network of global metadata, describing both long-term experimental data and SOM models, SOMNET, has been established to facilitate collaboration between modellers and data holders (Smith et al., 1997c). Further expansion of such networks and compilation of other networked field data, such as long-term ecosystem C flux estimates, will play an important role in continued improvements in our models.

# Measurement and Modelling Integration

A recurrent theme in discussions about the state-of-the-art of SOM modelling is the disparity between modelled pools and analytical SOM fractions measured in the laboratory. Most models represent the heterogeneity of SOM by defining several pools, typically 3–5, each with a characteristic specific decay rate or mean residence time, usually assuming first-order kinetics (i.e. constant proportional mass loss per unit time) (see reviews by Paustian, 1994; McGill, 1996). As fresh residues and SOM fractions decompose, a portion of the residual products is more resistant to further decay, which is represented by transfers to more slowly decomposing pools, in a decomposition cascade (see Swift *et al.*, 1979). Alternatively, specific decomposition rate can be made a function of a continuous SOM quality spectrum, which varies according to characteristics of the plant residues and the duration of decay (e.g. Bosatta and Agren, 1996). In either case, the representation of the model pools (or quality spectrum) is primarily conceptual in nature.

The sum of model pools is equal to the total organic carbon, a measurable and unambiguous quantity. However, individual pools generally are

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only loosely associated with measurable quantities obtained through existing chemical and/or physical fractionations performed in the laboratory. Consequently, it is not possible to falsify the internal dynamics of SOM models with conceptual pool definitions via a direct comparison to measured pool changes. Thus, a closer linkage between theoretical and analytical representations of SOM heterogeneity can be advanced through revising model definitions to coincide with measurable quantities or by devising more functional laboratory fractionation procedures, or both. The phrase 'modelling the measurable or measuring the modellable' has been coined as representing the two approaches towards a closer reconciliation between theoretical and experimental work on SOM (Christensen, 1996; Elliott *et al.*, 1996).

Various attempts have been made to correlate laboratory fractions with model pool definitions and to devise more general guidelines for initializing the soil organic matter pools as defined in existing models. For example, Motavalli et al. (1994) compared laboratory measurements of C mineralization with simulations by the Century model (Parton et al., 1994) for several tropical soils. When the active and slow pools in the model were initialized using laboratory determinations of microbial + soluble C for the active pool and light fraction for the slow pool, C mineralization was consistently underestimated, although all fractions were highly significantly correlated to C mineralization in a regression analysis. In contrast, using the standard procedure for internally initializing model pools, by running the model to equilibrium with estimated climate and primary productivity driving variables, provided the best fit to measured C mineralization. Similarly, Magid et al. (1997) tested the DAISY model using field measurements of litter decomposition, soil microbial biomass and particulate organic matter (POM), and endeavoured to 'measure the modellable' 'by relating the analytical fractions and residue quality indices to model pools. They concluded 'there is no firm relationship between the standard set of measurable quality parameters of the added plant materials and an adequate parameterization of the model.' Standard measures of residue quality such as lignin/N ratios and water-soluble/insoluble fractions were not able to account for the initial N dynamics during decomposition.

An alternative approach to modelling the measurable has been proposed by Arah (2000) and others (see Gaunt *et al.*, Chapter 2.6) using analytically defined pools and measurement of <sup>13</sup>C and <sup>15</sup>N stable isotope tracers to derive parameters in the model. The approach considers all possible transformations between measured C and N pools and devises a system of equations using observed changes in total C and N and <sup>13</sup>C and <sup>15</sup>N for each fraction to solve all model unknowns. Necessary requirements of such an approach are that the analytical fractions are distinct and together account for the total carbon inventory. It must also be parsimonious (no more than 4–5 pools each for C and N) such that

the model is sufficiently constrained to solve for parameter values (Arah, 2000).

Collins *et al.* (2000) used C mineralization data, total C and acid hydrolysis to estimate amounts and turnover rates for a three-pool model (representing active, slow and passive fractions) for five long-term experiments in central USA. They demonstrated that the amount of crop-derived carbon, determined from field sampling and <sup>13</sup>C natural abundance methods, was well correlated with the mean residence time of the slow pool determined from laboratory fractionations. The relationship was consistent across soil depth increments (up to 1 m) within sites, but differed distinctly between forest- and prairie-derived soils.

For both approaches, measuring the modellable or modelling the measurable, the derivation and testing of SOM models has benefited substantially from the increased application of isotopic methods, particularly the use of the stable isotopes <sup>13</sup>C and <sup>15</sup>N and radioactive <sup>14</sup>C. Isotope dynamics can be coded explicitly into models, including appropriate isotope discrimination coefficients. In particular, the use of natural abundance <sup>13</sup>C has been valuable in quantifying SOM dynamics following land use change or changes in agricultural management (e.g. Balesdent et al. 1987; Six et al., 1998; Collins et al., 2000). The method relies on the difference in <sup>13</sup>C natural abundance between plants with different photosynthetic pathways (usually contrasting  $C_3$  versus  $C_4$  plants) so that the relative mix of SOM derived from a particular vegetation can be quantified. Thus, where land use changes have occurred at a known point in time, for example C<sub>3</sub> forest conversion to C<sub>4</sub> cropland, the rate of loss of the original forest-derived SOM and the contribution of new crop-derived SOM can be inferred from changes in the <sup>13</sup>C signature of the soil. The approach can be extended to make inferences about the dynamics of SOM isolates as well as whole soil. Since land use or management changes involving shifts in major vegetation types are relatively widespread and occur in nearly all types of climatic and soil conditions, the approach is well suited for testing models for general use and regional applications. However, the inferences based on stable isotope data introduce additional sources of variability and error that need to be accounted for (Veldkamp and Weitz, 1994), and accurate measures of pre-disturbance soils and the amount and type of post-disturbance plant residue inputs are required.

Radiocarbon dating of SOM and tracer methods utilizing the <sup>14</sup>C enrichment from above-ground bomb testing have been utilized in conjunction with simulation modelling of SOM dynamics (e.g. Jenkinson *et al.*, 1987; Trumbore *et al.*, 1995), although to a lesser extent than <sup>13</sup>C, due in part to the greater expense and more limited availability of <sup>14</sup>C analysis facilities. Radiocarbon dating of SOM and SOM fractions has had a major role in the definition of recalcitrant pools in SOM models (Falloon and Smith, 2000). Initialization of the inert pool fraction in the Roth-C model is

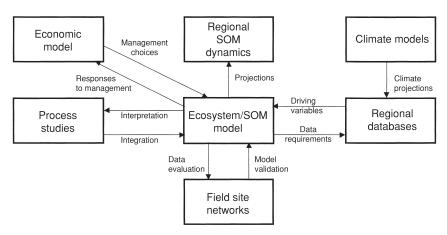
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based explicitly on <sup>14</sup>C carbon dating, and default estimation procedures have been published by Falloon *et al.* (1998a). The inclusion of a small (~5–10% of total C), totally inert, and very old SOM fraction in the Roth-C model was based on the need to reconcile the radiocarbon age of the soil with rates of SOM turnover inferred from bomb-C measurements. In analysing <sup>14</sup>C dynamics for a tropical and temperate soil, Trumbore (1993) concluded that the temperate soil was best represented by a relatively even distribution of organic matter in active, slow and passive pools while the tropical soil was best represented as comprised of mostly actively cycling carbon with a small, very recalcitrant pool.

Seeking a closer correspondence between theoretical and measurable fractions will continue to be a major thrust of SOM research. It should be realized that both theoretical and measurement-based depictions of SOM components are, in essence, 'models', i.e. abstractions and simplifications of reality. As new analytical procedures are developed that give functionally meaningful results, with consistent and repeatable patterns, they will doubtless influence the definition of modelled pools. Similarly, the expanded use of isotopic tracers and the availability of a richer set of field experiments will allow more robust and constrained testing of conceptually based models.

# **Regional Applications**

Climate change issues, specifically questions about: (i) the role of soil carbon as a potential part of the 'missing' terrestrial carbon sink; (ii) climate change impact studies; (iii) the need to quantify the emissions/sinks of CO<sub>2</sub> from soils as part of national greenhouse gas inventories; and (iv) interest in the potential of soil C sequestration as a greenhouse gas mitigation option have helped spur the development and application of SOM modelling systems at regional and global levels (e.g. Burke et al., 1991; Donigian et al., 1994; King et al., 1997; Falloon et al., 1998b; Paustian et al., 2000; Schimel et al., 2000). The approaches used are similar in most cases and rely on the linkage of simulation models with geographically distributed databases, typically maintained in geographic information systems (GIS) which contain model-driving variables and initial conditions (i.e. inputs) and help to manage and display model output (Fig. 2.1). Typically, model-driving variables (i.e. climate, soils, vegetation and land use) define a unit area of land, or polygon, having a unique combination of driving variables to which the model is applied. Approaches vary in the spatial and temporal resolution employed, the degree to which sub-polygon distributions of soil, vegetation and land use are represented, the source of the input data and the model being used. Such approaches have enabled information and understanding of SOM, originally derived at the field level, to be scaled up



**Fig. 2.1.** Structure for regional modelling and integrated assessment of SOM dynamics. Ecosystem/SOM models, based on fundamental process studies, are driven by spatially distributed databases of driving variables and validated by regional networks of long-term experimental sites. For climate change impact assessment, climate models provide projected climatic driving variables for the region. Economic decisions influence the type and distribution of management systems, and the ecosystem responses to changes in management and climate provide a feedback to economic and management decision models.

to encompass geographic and political units which are relevant to decision and policy makers.

However, significant challenges remain, particularly considering the potential impact of decisions based on these types of applications. A number of authors (e.g. Elliott and Cole, 1989; Paustian et al., 1995; Powlson, 1996) have emphasized the importance of linking such applications to regionally distributed experimental databases for model validation purposes, as described earlier in this chapter. As of yet, there are few studies that have examined the influence of spatial and temporal scale on model predictions at the regional scale. Depending on the properties of the driving variables and the model functions that utilize them, aggregation errors may be more or less severe, depending on the linearity of their scaling properties (Paustian et al., 1997). Similarly, there is a need to develop and apply formalized methods for uncertainty analysis of regionalized SOM models, including differentiating between uncertainty associated with input data and uncertainty in model predictions. Decision-makers utilizing the information generated by regional modelling systems will accept uncertainty as long as they have an idea as to the degree of uncertainty and that the estimates are unbiased.

In intensively managed ecosystems, such as agriculture, management decisions by farmers have a profound influence on the structure and functioning of the ecosystem, including the amount and dynamics of soil 50 K. Paustian

organic matter (Antle, 1996). Management practices are to a large degree driven by economic factors. Thus, the feedbacks and interactions between biophysical and economic components that govern the larger humandominated ecosystem are fundamental to predicting potential changes in soil C amounts and dynamics. This is particularly true for assessing the potential outcome of greenhouse gas mitigation policies in which changes in soil C and other soil-derived greenhouse gases will have a direct effect on farm income. However, it is equally true for assessing the effects on soil C of climate change or other changes in policies or economics to which farmers respond. Understanding regional soil organic matter dynamics, as driven by economically based management decisions, will require a whole-ecosystem approach where the interactions between SOM, crop yields, economic returns and subsequent changes in management feedback to determine SOM and crop responses (Fig. 2.1). Integrating SOM models into such systems will provide valuable tools to guide policy and decision-making on many of the pressing environmental issues facing society.

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# Soil Organic Matter Sustainability and Agricultural Management – Predictions at the Regional Level

2.1

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#### Introduction

In Europe, the potential for carbon sequestration from various changes in land management has previously been estimated using data from the Global Change and Terrestrial Ecosystems Soil Organic Matter Network (GCTE-SOMNET; Smith et al., 1996). Linear relationships between management practices and yearly changes in soil organic carbon were developed and used to estimate changes in the total carbon stock of European soils (Smith et al., 1997a,b, 1998, 2000a,b). To refine these semi-quantitative estimates, the local soil type, meteorological conditions and land use must also be taken into account. We previously have used the Rothamsted carbon model (Roth-C) linked to geographical information systems (GIS) to estimate the potential effects of afforestation on soil carbon stocks in central Hungary (Falloon et al., 1998). Further developments have involved a combined modelling approach. Our new approach is based on the CENTURY model frame and allows either the Roth-C or CENTURY soil organic matter (SOM) decomposition model to be used (Falloon et al., 1999, 2000). This allows an equal comparison of the models. The GIS-linked system integrates land use, soil and weather data with knowledge of land use history, net primary production, local agricultural practices and best estimates of the current soil organic carbon (SOC) stock.

In this chapter, we describe how these developments can be used to estimate SOM sustainability and carbon sequestration at the regional level using a dynamic simulation model linked to spatially explicit data, and show results of carbon sequestration potential estimated with a simple regression-based approach. We demonstrate the system in use for an area of central Hungary.

#### Methods

Our case study area (24,804 km²) in central Hungary and GIS data sets have been described in detail elsewhere (Falloon *et al.*, 1998, 2000). Here, we describe the use of the IGATE system (Fig 2.1.1; Falloon *et al.*, 2000) to compare C sequestration estimates using the Roth-C (Coleman and Jenkinson, 1996) and CENTURY (Parton *et al.*, 1988) models with estimates using simple linear regressions (Smith *et al.*, 1997a,b, 1998, 2000a,b).

The CENTURY model was validated and calibrated using 17 data sets from the GCTE-SOMNET database (Smith *et al.*, 1996) and the IBP woodlands data set (DeAngelis *et al.*, 1981). To compare the models at the regional scale, we initialized the models by running for 3000 years under native vegetation (forest: Marton *et al.*, 1989) and then 500 years of current land use. We then ran a demonstration scenario of afforestation of all arable land to (i) show the maximum possible increase in SOC stocks; and (ii) highlight the largest differences between model estimates. We used the regressions of Smith *et al.* (1997a,b, 1998, 1999a,b) to estimate changes in regional SOC stocks under the six scenarios given in Table 2.1.1, all applied to all arable land. We calculated annual CO<sub>2</sub> emissions offsets using 1990 national data from Marland *et al.* (1999), and scaled this to our

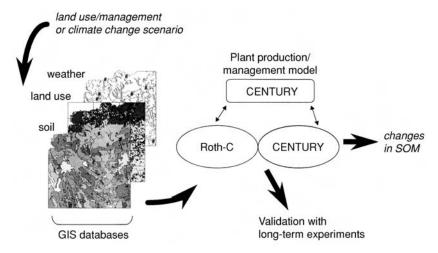


Fig. 2.1.1. IGATE system.

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_	Regression		Roth-C model		CENTURY model	
Scenario	SOC change, Tg	% annual emissions offset	SOC change, Tg	% annual emissions offset	SOC change, Tg	% annual emissions offset
Afforestation	101.8	21.66	37.00	8.00	55.00	12.00
No till	44.77	9.52	_	_	_	_
Organic manures <sup>a</sup>	20.00	4.26	_	_	_	_
Sewage sludge <sup>b</sup>	131.25	27.93	_	_	_	_
Cereal straw <sup>c</sup>	46.44	9.88	_	_	_	_
Ley-arable <sup>d</sup>	62.55	13.31	_	_	_	_

**Table 2.1.1.** Changes in SOC stocks and CO<sub>2</sub> emissions offsets for the area of central Hungary.

All scenarios applied to all arable land. Annual application rates:  $^a10 \text{ t ha}^{-1}$ ;  $^b2.59 \text{ t ha}^{-1}$ ;  $^c5.07 \text{ t ha}^{-1}$ ;  $^d30\%$  ley arable rotation.

study area. We used 1990  $CO_2$  emissions since 1990 is the Kyoto protocol baseline.

Finally, we investigated the sustainability of SOC stocks for a typical arable polygon of the case study area in our GIS database using the CENTURY model. We ran the model for 3000 years under native forest, then 350 years extensive arable agriculture and 100 years (1860–1960) intensive arable agriculture with the inorganic N fertilization rate set at 1960 levels (the earliest statistic from FAOSTAT, 1999). From 1960 to the present day, the model was run under intensive arable agriculture with annual N application rates set from FAOSTAT, and finally for a 100 year scenario of intensive arable agriculture with N application rates at the current level.

#### **Results and Discussion**

There was good agreement between measured and simulated SOC levels at the validation sites ( $r^2 = 0.83$ ). The results of the model comparison and the regression estimates for the various scenarios are given in Table 2.1.1. For our demonstration scenario of afforestation of all arable land, SOC changes predicted using the regression method are significantly greater than those predicted using the Roth-C or CENTURY models, and the CENTURY model predicts a greater C sequestration estimate than Roth-C. Although some of these scenarios are unrealistic and require refinement for the study area, the magnitude of  $CO_2$  emissions offsets indicated is encouraging.

Figure 2.1.2 shows the dynamics of SOC predicted by the CENTURY model for our investigation of SOC stock sustainability. Under native forest

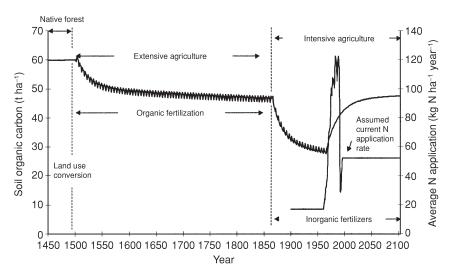


Fig. 2.1.2. Historic-future SOC dynamics using fertilizer statistics (CENTURY model).

vegetation, the SOC content of the soil reached a high, steady state. Upon conversion to extensive arable agriculture, there was a steady loss of SOC. On conversion to a more intensive arable system, with a low level of N application, there was a rapid loss of SOC, which recovered following the high N application rates during the 1970s. Despite the drop in N application rates after 1990, assuming that current N application rates continue for 100 years, the model predicted a recovery and stabilization of SOC stocks. We assumed that current N application rates continue into the future since no projected figures were available. We remain uncertain regarding the sustainability of current SOC, due to the limited information on historical land use and management.

In conclusion, we have shown that different methods for estimating changes in regional SOC stocks can produce quite different results. There may be several reasons for the differences between SOC stock estimates obtained using the regressions CENTURY and Roth-C. The regression-based estimates are generalizations of data from European long-term experiments, on specific soil types and with specific cropping regimes. Reasons for differences in carbon changes resulting from the actual management scenarios investigated here have been discussed in detail by Smith *et al.* (1997a,b, 1998, 2000a,b). The long-term data sets used in the regression-based approach may not be specific to central Hungarian soil, climatic and management conditions. The dynamic modelling approaches do, however, account for local soil type, weather and management conditions, and hence we would expect different (and more site-specific) results. We would also expect different results from the two models (CENTURY and Roth-C), since they have different structures, pool sizes and turnover

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times. More information is needed on historical land use and practices to allow estimates of the sustainability of current agricultural practices with regard to SOC stocks.

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## Modelling of Carbon Dynamics 2.2 in a Rural Area of Central Germany

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#### Introduction

Soil organic matter (SOM) is a very important component of agroecosystems. Biological, chemical and physical soil properties are strongly correlated with the carbon content of the soil, which is the usual indicator for SOM. It is very important to distinguish between the inert and the decomposable part of SOM. Turnover processes of this decomposable SOM lead to a more or less continuous flowing stream of nitrogen and other nutrients in soil. SOM changes are very slow and poorly indicated by measurements. Modelling may give faster results about the directions and amounts of soil organic matter change in the future. This chapter presents the results of a study in the Chernozem region of central Germany on the consequences of land use change for organic matter cycling and SOM dynamics.

#### **Model and Database**

The CANDY system integrates a simulation model and a database of parameters, measurement values, initial values, weather data and management data. The user interface not only provides the function for simulation control but also tools for parameter estimation, risk analysis and recommendation for fertilizer application. The simulation model works on a daily time step with modules for calculation of soil temperature, moisture content, the processes of the soil carbon–nitrogen cycle and pesticide

dynamics. The model is integrated in a geographic information systems (GIS)/database environment in order to obtain inputs on weather, soil physics and agricultural management on a regional scale. A more detailed description of the construction and evaluation results of the CANDY model has been published (Franko 1996; Franko *et al.*, 1997).

A landscape unit is considered as a composition of homogeneous patches that are independent from each other with regard to the processes in the soil. The construction of the homogeneous patches is a result of an overlay of a soil map with a map of the farm fields and a map of precipitation patterns. It is very useful to use identical shapes for farm fields and precipitation patterns in order to reduce the number of patches to a reasonable level. The soil map and the farm field map as well as the map of climate structure have a database background with detailed information.

The soil mapping units are characterized by a list of soil profiles, weighted by their part in the whole area of the item. A soil profile description contains a list of horizons with their depths. The soil physical parameters of density, texture, field capacity and wilting point are assigned to each horizon.

Climatic data include air temperature, global radiation and precipitation provided by a station situated nearby. A precipitation index has been calculated for every farm field from long-term averages of regional precipitation distribution expressing the relationship of precipitation intensity between a given point in the area and the position of the climate station. During the simulation runs, this index is used in order to adapt the local precipitation data.

Farm field management is described by a date, an action, a specification and an intensity. Actions are cropping, tillage, application of fertilizer, manure or pesticides, and irrigation. Cropping data have to include the vield and/or the nitrogen uptake of the crop at harvest.

#### Investigation Area and Simulation Scenario

The study was performed on an area of 3850 ha of arable land on Haplic Chernozem with loess as parent material. The water-holding capacity of the upper 100 cm is ~310 mm, with 190 mm available to plants. Clay content varies between 18 and 20%. Long-term averages of air temperature and annual precipitation are 8.7°C and 520–560 mm, respectively.

Land use data for every farm field were collected for a time period from 1981 to 1996. With the re-unification of Germany, there were changes in the agriculture of Eastern Germany concerning setaside of land, reduction of animal breeding, crop rotations and yields because of improved agrochemicals and technology. An important question is how this change in land use will affect the SOM dynamics. The land use change in the

investigation area is characterized by a decrease of area for root crops from 24 to 13% which is now used for oilseed rape, sunflower and as setaside land. Two percent of the former cereal area (now 60%) is now used for legume crops. The yield has increased for winter wheat from 0.64 kg m<sup>-2</sup> in the 1980s to 0.73 kg m<sup>-2</sup> in the 1990s and for sugar beet from 3.5 to  $5.2 \text{ kg m}^{-2}$ .

The simulated scenario starts in August 1980. Observed values for the required initial conditions of the model were not available. The initial content of SOM was calculated using site-specific statistical information about yields and animal concentration before 1981 assuming an even application of the organic inputs and that the organic matter content had reached a steady state. Parameters and algorithms of the necessary calculations have been published (Franko, 1997). Initial soil water content was assumed to be 60% of field capacity, and soil mineral nitrogen has been assigned according to internal standard parameters of the model that are related to the different soil units.

#### **Evaluation of Soil Organic Matter Level**

It is known from long-term experiments that land use is only responsible for changes in the decomposable part of SOM whereas the inert SOM is dependent of the site conditions. Therefore, the evaluation is restricted to the decomposable SOM pool. Recently published results of Körschens (1999) show that the effect of SOM on yield is limited to a certain level and that in different long-term experiments a SOM range of ~0.14–0.51% of decomposable carbon provides the best conditions for high-quality yields.

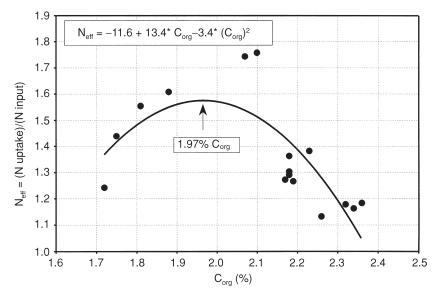
Could higher inputs of organic material to the soil and the subsequent increase in the carbon storage in the soil be of benefit for the global carbon cycle? A simple calculation shows that there are better solutions for the reduction of carbon dioxide emissions. Under the conditions of Chernozem in central Germany, an additional straw input of 400 g m<sup>-2</sup> per year will eventually lead to an increase in the SOM carbon of ~1.2 kg m<sup>-2</sup> with a maximum rate of 60 g m<sup>-2</sup> year<sup>-1</sup>. This rate decreases with time because of the remineralization of the newly formed SOM. It takes ~100 years to reach the final level. Afterwards, no further increase will occur. At this steady state, the total input is equal to the mineralization. However, the same amount of straw can be used as an energy source in order to replace fossil fuels. The energy equivalent of 400 g of straw is ~5.6 MJ (Brenndörfer et al., 1994) - an amount equivalent to the energy gain from ~120 g of carbon in fossil fuels. This is twice as high than the maximum accumulation rate in SOM. Because of its regenerative character, the consumption of the straw as an energy source will relieve the global carbon balance per square

metre of ~12 kg within 100 years, which is ten times better then the sequestration of carbon in soil.

The range of optimal content of decomposable carbon, published by Körschens (1999), shows a tendency for higher values in soils with a lower turnover activity. This leads to the assumption that the turnover rate of SOM could be a good indicator for SOM management. This is also reasonable because an increase in the SOM level increases the nitrogen mineralization and could also increase the nitrogen surplus. Results from the long-term experiment at Bad Lauchstädt (Körschens *et al.*, 1994) suggest that the optimal SOM level for nitrate recovery is 1.97% Corg (Fig. 2.2.1), which is equivalent to a decomposable carbon content of 0.42% and an annual carbon turnover of ~90 g m<sup>-2</sup>. Using these values as a benchmark, possible ranges for the evaluation of the SOM level are presented in Table 2.2.1.

#### **Results and Discussion**

The steady increase in yields – especially after 1989 when agricultural technology improved very rapidly – leads to a continuous increase in SOM with some fluctuations due to yield decreases in years with very low precipitation



**Fig. 2.2.1.** Efficiency of the nitrogen usage ( $N_{eff}$ ) as the relationship between N uptake and N input from mineral fertilizer and manure for 16 different treatments of the Bad Lauchstädt long-term experiment in term of dependence on the corresponding content of soil organic carbon ( $C_{org}$ ). Data after Körschens *et al.* (1994).

(Fig. 2.2.2). Using the amount of decomposable SOM as an indicator, > 90% of the area is now at an optimal SOM level, and < 5% is classified as 'low' or 'high'. It is obvious that the system has not reached a steady state. Despite some decreasing trends, most of the area shows a carbon accumulation. Because of the very slow reaction time of SOM, it would be more informative to evaluate the carbon input rates in order to predict the future prospects. Taking the input rates of carbon reproducing SOM ( $C_{rep}$ ) from 1993 to 1996 as an indicator, a large area has to be classified as 'very high' in terms of supply of SOM (Fig. 2.2.3).

Despite the temporary very high contents of mineral nitrogen in the soil (>  $20 \text{ g m}^{-2}$ ), the percolation water leaving the rooting zone meets the quality conditions for drinking water. The total nitrogen export (leaching plus gaseous losses) amounts to  $4.4 \text{ g m}^{-2} \text{ year}^{-1}$  and is lower than the nitrogen import from the air which is assumed to be  $\sim 6 \text{ g m}^{-2} \text{ year}^{-1}$ .

If the trend of carbon accumulation continues, a reduced nitrogen efficiency is to be expected, with subsequent increasing export of nitrogen

Table 2.2.1.	Classification of the SOM content (SOM level) in terms of soil organic carbon (C <sub>org</sub> ),
decomposable	e carbon (C <sub>dec</sub> ) and soil organic carbon reproduction rates (C <sub>rep</sub> ) relative to the
efficiency of th	ne nitrogen input (N efficiency) for a Haplic Chernozem in central Germany.

SOM level	N efficiency	C <sub>org</sub> (%)	C <sub>dec</sub> (kg m <sup>-2</sup> )	C <sub>rep</sub> (g m <sup>-2</sup> year <sup>-1</sup> )
Very low	< 1.50	< 1.80	< 1.45	< 60
Low	1.50-1.58	1.80-1.91	1.45-1.95	60-80
Normal	> 1.58	1.91-2.03	1.95-2.45	80-100
High	1.58-1.50	2.03-2.15	2.45-2.95	100-120
Very high	< 1.50	> 2.15	> 2.95	> 120

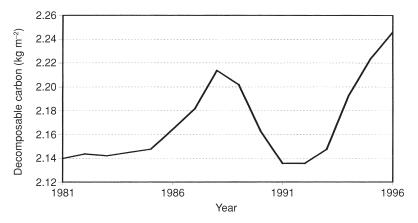


Fig. 2.2.2. Regional average of decomposable carbon in soil.

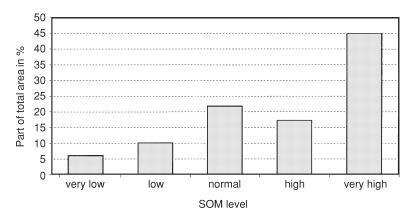


Fig. 2.2.3. Regional classification of the SOM level according to the average carbon input.

into the environment. In order to avoid this risk and to keep the system at an optimal SOM level, it is necessary to find new solutions for organic material management that guarantee an equal distribution of the required carbon combined with an energetic usage of the surplus.

#### Conclusions

Model outputs show an increase in carbon storage in the soil of the investigated area due to higher inputs of organic material. Under the current conditions, the nitrogen export into the environment is lower than the nitrogen import from the atmosphere. Results of the long-term experiment at Bad Lauchstädt indicate an optimal rate of SOM turnover of ~90 g m<sup>-2</sup>. Using this as the indicator for the evaluation of the carbon dynamics in the investigation area, a surplus of organic matter can be found that could cause an increase in nitrate exports into the environment. Surplus of organic matter could be used better for the replacement of fossil fuels in order to reduce the carbon dioxide output to the atmosphere.

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# Modelling Variation in C and N Loss and Effects on N Mineralization After Grassland Ploughing Over a Catchment

2.3

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#### Introduction

Ploughing grassland has a great impact on organic carbon (Corg) and nitrogen (N<sub>t</sub>) turnover, and related budgets vary considerably. Expressed as relative total N content, its decline after grassland ploughing can be approximated by a unique exponential decay function. As a generalization, 50-60% of initial nitrogen will remain under long-term arable land use (Voroney et al., 1981; Johnston et al., 1994) for loamy grassland soils. Difficulties arise for regional quantitative analysis, since long-term observations are missing, and initial values are often unknown on the landscape scale. 'Snap-shots' of the current status for soil organic matter (SOM) combine spatial and temporal variation reflecting physicochemical properties of soils and differences in management. Variable soil physical and hydrological properties may have caused enrichment and differences in the chemistry of SOM. One question is whether equilibrium C and N contents of arable soils will be reached within a period meaningful for farm management (a decade or a generation). Models, which describe the underlying dynamics as a function of soil, climate and management variables, may be able to explain the observed variability. As a first step, we attempt here to simulate SOM dynamics following ploughing up of grassland using a standard arable model (SUNDIAL) with modifications only to the soil carbon parameters as measured. These early predictions will be compared with SOM N fractions and N mineralization measurements.

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#### **Materials and Methods**

Soil samples of the A horizon were collected on several arable sites in two German catchments. They represent a series characterized by variable time elapsed since grassland ploughing (GP, 1–33 years), soil texture (sandy loam–clay loam) and SOM (h2–h4) class. Arable (A) sites were assumed to be under cultivation for at least 115 years, setting the long-term equilibrium content (Y[A]).  $C_{org}$  and  $N_t$  (Kjeldahl) were measured by dry combustion. Assuming a ploughing depth of 0.25 m and a uniform dry bulk density of the soil (1.5 g cm<sup>-3</sup>), the storage of C and N in the GP soils ranged from 200 to 90 t C ha<sup>-1</sup> and 18 to 7 t N ha<sup>-1</sup>, actually representing two different SOM classes. The decomposable and recalcitrant fractions of potentially mineralizable N ( $N_0 = N_{\rm dpm} + N_{\rm rpm}$ ) were determined by incubation (Richter *et al.*, 1989). A sub-sample was used to estimate carbon and nitrogen in the microbial biomass ( $C_{\rm mic}$ ,  $N_{\rm mic}$ ) by chloroform fumigation extraction (CFE; Brookes *et al.*, 1985; Vance *et al.*, 1987).

#### Modelling

Two approaches were used to explain the dynamics of SOM fractions after grassland ploughing in the sub-samples of sandy and clay loam. A static model (Equation 1) was used to quantify the total amount of C, N and N<sub>0</sub> in SOM derived from grassland (Y[G]) at any time ( $t_{GPA}$ , years) under arable cultivation before reaching an equilibrium content (Y[A]). The half-life,  $t_{1/2}$ , is estimated from the rate constant k (Equation 2).

$$Y = Y[A] + Y[G] \times \exp(-k \times t_{GPA})$$
 (1)

$$t_{1/2} = \ln 2/k \tag{2}$$

The dynamic model SUNDIAL was chosen, which is based on the Rothamsted carbon model (Roth-C; Jenkinson and Coleman, 1994) extended to include N turnover (Bradbury *et al.*, 1993). As major inputs, the model considers C and N from fertilizer (mineral and organic) and crops (stubble + straw and debris), which are partitioned between mineral ( $CO_2$ ,  $NH_4$  and  $NO_3$ ) and organic pools (humus and microbial biomass) in the soil. Mineralization and immobilization of N due to decomposition of crop residues, biomass and humus are calculated from the C/N ratio of the decomposing material on a weekly basis. For the simulations of C and N turnover after grassland ploughing, humus C and fraction of microbial biomass C ( $C_{mic}/C_{act}$ ) were initialized according to measurements in single samples of loam soils (Widmer, 1993; Table 2.3.1). The fraction of  $C_{mic}$  was changed in the range of low (0.028; arable) to high (0.046) to account for different mixing ratios (ploughing depth, sod depth and litter incorporation). Absolute values for  $C_{mic}$  were high compared with other locations

Management/soil type	Humus C (t ha <sup>-1</sup> )	C <sub>mic</sub> /C <sub>act</sub> fraction	C/N ratio
A/sand	25	0.028	12–17
A/loam	36	0.028	11
A/clay	44	0.028	10
GP/loam	90	0.032	10
GP/humic loam	204	0.012	11.4
Grass/loam	94	0.035	9.4

**Table 2.3.1.** Parameters selected for model initialization.

(Lavahun *et al.*, 1996), but the relative difference of microbial C in grassland and arable soil was similar (20%).

Management scenarios for the simulation were based on a standard 3-year rotation, consisting of winter oilseed rape, winter wheat and winter barley (RWB), with and without residue (straw; Y/N) incorporation. Two different yield potentials were considered for the cereals (miN/maX), which were correlated with a difference in N fertilizer addition (+50 kg N ha<sup>-1</sup>). Generally, a trend was accounted for in N fertilization rising between 1965 and 1985 in cereals, starting at 120 kg N ha<sup>-1</sup> and reaching a plateau of 180 and 230 kg N ha<sup>-1</sup>, respectively. Simulations were also performed for 'continuous arable' (A) to check the effect of fertilization on SOM equilibria.

#### **Results and Discussion**

#### Measurements

Soils from areas of high water table had about twice the amount of SOM; clay soils were too variable to fit an empirical relationship (Fig. 2.3.1). For  $C_{\rm org}$  and  $N_{\rm t}$  measured in humic soils, the hypothetical equilibrium content will not be reached until 115 years of arable cultivation. Some decline can be derived from the measured  $N_{\rm t}$  contents in clay soils if one identifies two different soil types (Humic Gley and Gley). For the sub-sample of loess soils (sandy loam), typically low in SOM (h2–h3), exponential decay functions could be parameterized (Table 2.3.2) as shown for total nitrogen (Fig. 2.3.1). We assumed that equilibrium was reached after 115 years. Quantifying the half-life of either fraction under arable conditions (Table 2.3.2) shows that  $N_{\rm t}$  reaches its equilibrium level faster than  $C_{\rm org}$ . The drop in carbon content is ~10% larger compared with nitrogen, in spite of returning carbon-rich residues, such as straw, with a wide C/N ratio and a comparatively slow turnover rate. Overall, mineralization of the grass sod goes along with a narrowing C/N ratio. Potentially mineralizable nitrogen,

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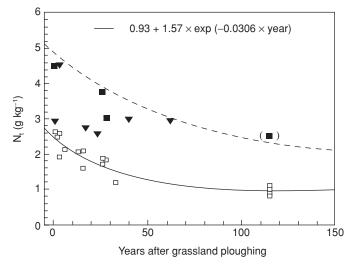


Fig. 2.3.1. Spatial and temporal variation of total N in the A horizon after ploughing grassland within two catchments; □ loam soil (h2-h3), ■ humic loam soil (< h3) and ▼ clay soils (h4).

**Table 2.3.2.** Mean parameters ( $\pm$  SE) and approximate half-life,  $t_{1/2}$ , after grassland ploughing in loam soils (h2–h3); exponential decay function:  $Y = Y/AI + Y/GI \times EXP(-k \times t_{GPA})$ .

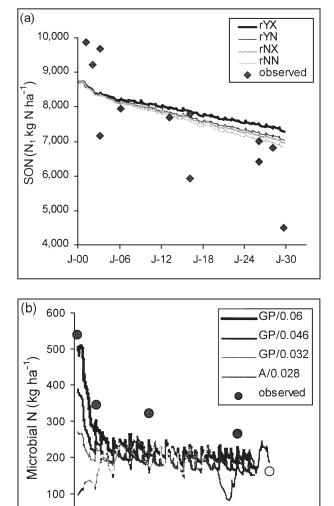
Parameter					
variable	Y[A]	Y[G]	k (year <sup>-1</sup> )	r²	$t_{1/2}$ (years)
C <sub>org</sub>	0.85	1.72	0.0217	0.887	32
(g 100 g <sup>-1</sup> )	(0.05)	(0.11)	(0.004)		
$N_t$	0.93	1.57	0.0306	0.884	22
$(g kg^{-1})$	(0.11)	(0.11)	(0.006)		
$N_0$	125	192	0.071	0.771	10
$(mg kg^{-1})$	(16)	(25)	(0.020)		

 $N_0$ , which may be derived from microbial N, decreased by 35% and had a half-life of ~10 years. This is longer than predicted for  $N_{mic}$  in the SUNDIAL model (Bradbury *et al.*, 1993).

#### **Simulations**

The SUNDIAL simulation results presented here include the active organic matter fractions, humus and biomass, and net mineralization. Nitrogen in the active humus fraction initially is underestimated by  $\sim 1000$  kg N ha<sup>-1</sup> (Fig. 2.3.2a; humus N + N<sub>mic</sub>). Depending on the C/N ratio, this corresponds to  $\sim 10$ –15 t C ha<sup>-1</sup>, which can be attributed to the

grass sod being ploughed in. In permanent grassland, Garwood *et al.* (1977) found 22.8 t  $ha^{-1}$  of macroorganic matter in the top 15 cm which corresponds to 13 t C  $ha^{-1}$ . The rate of N loss in the first 30 years of arable cultivation is underestimated in the simulation by > 1000 kg N  $ha^{-1}$ , corresponding to 40 kg N  $ha^{-1}$  annually. The differences in arable management



**Fig. 2.3.2.** Comparison of simulated and observed course of N in the soil after grassland ploughing (GP): (a) total N (humus +  $N_{mic}$ ) related to management as rape rotation, residues returned (Y/N) and yield potential (maX/miN) and (b)  $N_{mic}$  (G/GP •; A  $\odot$ ) related to model parameterization.

F-19

J-25

N-31

0-12

0 <del>|</del>

M-06

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(straw incorporation; yield potential) accounts for a variation in  $N_t$  of ~500 kg N ha<sup>-1</sup>, about half the spread observed in the catchment values.

The fraction of N<sub>mic</sub> in the soil is quickly decreasing after ploughing grassland by ~200 kg N ha-1 to a value fairly stable over the next 30 years (Fig. 2.3.2b). Increasing the fraction of microbial carbon ( $C_{mic}/C_{org}$ ) in the simulation to values measured in soils could not explain the absolute height of microbial N observed for grassland,  $N_{mic}(G)$ . To fit the value  $N_{mic}(G)$ , the C<sub>mic</sub> fraction needed to be increased to 0.06, which is 70% higher than before. The difference between model predictions and observations may be caused by the distribution assumed for SOM and biomass. In arable soils, it is assumed that SOM and biomass are distributed 80% in the top 25 cm and 20% in the horizon below. Lavahun et al. (1996) found only minor differences for the distribution of C<sub>mic</sub> in profiles of grassland and arable soils. However, grassland contains a higher proportion of Corg in the top 25 cm. This could be attributed to grass sod incorporated initially, and the microbial biomass associated with it. Joergensen (2000) found that considerable C<sub>mic</sub> in rhizosphere material and on grass roots was ignored in standard CFE. Finally, the protective power of these silt-loam soils is underestimated, and the model reaches the equilibrium content,  $N_{mic}(A)$ , too quickly. Net mineralization is predicted as a quasi-linear process during the first 30 years of simulation; however, with 150 kg N ha<sup>-1</sup> year<sup>-1</sup>, it is twice as high as estimated for continuous arable (65 kg N  $ha^{-1}$  year<sup>-1</sup>).

#### **Conclusions**

The static model predicts different equilibria and half-lives for the total and active N fraction. Dynamic simulation proved to be a valuable tool in detecting missing links: fitting a single parameter, we could quantify microbial activity. The concept of 'microbial biomass' in the soil needs to account for  $N_{\rm mic}$  and  $C_{\rm mic}$  associated with litter and crop debris, possibly by regarding two different biomass fractions (bacteria and fungi) as explaining flush and protection in ploughed-up grassland. Further improvements to SUNDIAL should allow the model to be used explicitly to simulate leyarable rotations.

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### Modelling Decomposition of Sugarcane Surface Residues and the Impact on Simulated Yields

2.4

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#### Introduction

The Agricultural Productions Systems Simulator (APSIM; McCown et al., 1996) describes the dynamics of crop growth (for various crops), soil water, soil nitrogen (N) and carbon (C), and plant residues as a function of climate, cropping history (e.g. crop type, sowing date) and soil management (e.g. tillage, fertilizer application). APSIM's linking of crop growth with soil water and N/C dynamics makes it particularly suited for extrapolating the results of agronomic experiments to different soil, management and/or climatic conditions (Probert et al., 1995, 1998b; McCown et al., 1996; Keating et al., 1997). Despite its wide application for this purpose, to date some APSIM modules have been developed and tested in grain and legumes systems in semiarid, rain-fed areas (Probert et al., 1995, 1998a,b; Carberry et al., 1996) and so could benefit from wider testing. This is particularly so of the APSIM-Residue module that describes the decomposition of surface residues.

Recently, increasing attention has been paid to the impact of crop residue retention on soil water and N dynamics of sugarcane systems in the environmentally sensitive humid coastal areas of Australia. APSIM provides a means of modelling these systems (Keating *et al.*, 1997; Thorburn *et al.*, 1999). While the overall agreement between measured and modelled data has been good (Thorburn *et al.*, 1999), the climate, soils and agronomic characteristics (e.g. residue mass, N fertilizer application rates,

frequency of fallows) are markedly different from those where the APSIM-Residue module was developed and most widely used. So it is appropriate that the performance of that module is examined in more detail in this environment.

This study contained three elements: the APSIM-Residue module was parameterized using experimental data (Robertson and Thorburn, Chapter 3.1) from two sites. The applicability of the parameter values to other sugarcane systems was then assessed against independent measurements of decomposition at another site. Finally, the impact of different residue decomposition rates, resulting from the different parameterizations, on simulated yield was assessed from longer term systems simulations for two of the sugarcane residue management experiments.

#### Methods

#### Field experiments

Sugarcane residue decomposition was measured over 1 year within three longer-term residue management field experiments, referred to as Mackay Early (harvested in July each year), Mackay Late (harvested in November) and Harwood Early (harvested in July) by Robertson and Thorburn (Chapter 3.1), who fully describe the methods employed. Harwood is located approximately 1500 km south of Mackay, and has a temperate climate compared with the tropical climate at Mackay (Robertson and Thorburn, Chapter 3.1). After harvest of a sugarcane crop, crop residues were left on the soil surface (as is common practice in the Australian sugar industry) and 8–10 quadrats, 1.5 m × 0.75 m in area, were established in each replicate plot of each experiment. A known mass of residue was enclosed within each quadrat. Residue in the quadrats was in direct contact with the soil, but enclosed over the top with fine nylon mesh ~20 mm in diameter. The mass of residue in individual quadrats was measured destructively over 12 months. Daily weather was measured at each site.

In addition to these measurements, soil moisture was logged hourly with Campbell CR10X dataloggers and CS615 water content reflectometers in one replicate plot of the experiments. Measurements were made over the 0–50 mm soil depth interval, and at 75 and 175 mm depth. Daily average values were calculated from the data.

#### The APSIM Model

The APSIM model configuration used in this study consisted of modules for soil N and C (APSIM-SoilN), soil water (APSIM-SoilWat) and surface

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residue (APSIM-Residue) dynamics (Probert et al., 1998b), and sugarcane growth (APSIM-Sugarcane; Keating et al., 1999). These modules have been fully described in the aforementioned papers, but a short description of them follows. The modules are one-dimensional and driven by daily climatic data. The dynamics of water, N, C and roots are simulated in soil layers, with water (and associated nitrate) moving between layers where gradients exist. The soil organic matter is divided into three 'pools', with fom representing the fresh organic matter (i.e. roots and incorporated plant residues), biom representing the active biomass in the soil, and hum representing the humified material. Part of the hum pool is considered inert. The soil water module is a 'cascading bucket' water balance model, with water between the drained upper limit (dul) and saturation draining to the layer below. The drainage rate is controlled by the parameter swcon. The lower limit of plant available water is defined by the parameter 1115. Evaporation from the soil follows Ritchie's (1972) two-stage evaporation model. The presence of plant residues on the soil surface affects runoff (and hence infiltration) and evaporation. The sugarcane module uses intercepted radiation to produce assimilates, which are partitioned into leaf, structural stalk, roots and sugar. The processes represented in the module are responsive to radiation and temperature, as well as water and N supply. Farming operations (such as fertilization, planting, incorporation of crop residues through cultivation or burning of crop residues) can be specified through the APSIM-Manager module to represent actual or hypothetical conditions.

#### **APSIM-Residue and its Parameterization**

In APSIM, plant residue on the soil surface is considered separately from soil organic matter. C in the residues is transferred into the soil organic matter pools upon tillage, which results in incorporation of residue into the *fom* pool, or decomposition, which transfers C into the *biom* and *hum* pools. The rate of residue decomposition is controlled by first order kinetics;

$$dR/dt = -kR \tag{1}$$

where R is the mass of residue per unit area, t is time and k is the rate coefficient given by

$$k = r_{\text{max}} \cdot F_{\text{C: N}} \cdot F_{\text{temp}} \cdot F_{\text{moist}} \cdot F_{\text{contact}}$$
 (2)

where  $r_{\text{max}}$  is the maximum (or potential) decomposition rate and  $F_{\text{C:N}}$ ,  $F_{\text{temp}}$ ,  $F_{\text{moist}}$  and  $F_{\text{contact}}$  are factors, scaled from 0 to 1, accounting for the limitations to decomposition of residue C:N, temperature, moisture and residue—soil contact, respectively. The functions defining the factors are,

$$F_{C:N} = \exp [j (1 - C: N/C: N_{opt})], C: N > C: N_{opt}$$
 (3a)

$$F_{\text{temp}} = (T/T_{\text{opt}})^2, T \le T_{\text{opt}} (3b)$$

$$F_{\text{moist}} = 1 - (\sum E_{\text{os}})/E_{\text{os, max}} \qquad (\sum E_{\text{os}}) < E_{\text{os, max}} \qquad (3c)$$

$$F_{\text{contact}} = 1.54 - 0.36 \, R,$$
  $1.5 < R \le 3$  (3d)

where j is the coefficient determining the slope of Equation 3a, T is the average daily air temperature,  $E_{\rm os}$  is the potential daily soil evaporation and the subscripts 'opt' and 'max' refer to the optimum and maximum parameter values, respectively. Outside the given limits, the factor values are set equal to 1 or 0, except for  $F_{\rm contact}$  which has a minimum ( $F_{\rm contact}$ , min) of 0.46 in the default values (Table 2.4.1).

For parameterization, APSIM-Residue was programmed into a spreadsheet and the inputs for calculating the factors from Equation 3, i.e. residue C: N, daily air temperature and soil evaporation, were taken from measured data. Soil evaporation data were derived from daily water balance calculations performed on the soil moisture data from the experiments. Using the input data, daily values of k were calculated from Equation 2, and daily residue mass from Equation 1. Optimal values of  $r_{\text{max}}$ ,  $T_{\text{opt}}$ ,  $E_{\text{os, max}}$ ,  $F_{\text{contact, min}}$  were determined by the generalized reduced gradient optimization method of Fylstra *et al.* (1998) to minimize an objective function, D (Table 2.4.1).  $F_{\text{C:N}}$  was not considered explicitly (i.e. was not included in Equation 2) in the optimization since it is constant once calculated from initial residue C: N, which did not vary greatly (75–95) between the three sites. Instead, the optimal value of  $r_{\text{max}}$  included the C: N limitations to decomposition, which were assumed to be uniform across the experiments.

To derive more general parameter values from the optimization, values of  $r_{\rm max}$ ,  $T_{\rm opt}$ ,  $E_{\rm os,\ max}$  and  $F_{\rm contact,\ min}$  were optimized for both the Mackay Early and Mackay Late experiments, i.e. to minimize the sum of the objective function from the two experiments. To provide some test of their generality, the optimal parameter values were used to predict residue decomposition at the third site, Harwood Early.

**Table 2.4.1.** Parameter values and resultant values of the objective function (D) in each experiment for two parameterization schemes, the default parameters (Probert et~al., 1998b; which included those for  $F_{C:N_1}$  not shown here) and the result of the optimization.

Parameter value				e	Values of D <sup>a</sup>		
Scheme	$r_{\rm max}$	E <sub>os, max</sub>	$T_{ m opt}$	F <sub>contact, min</sub>	Mackay Early	Mackay Late	Harwood Early
	0.100 0.007	20 9	20 23	0.46 1	100.3 10.2	384.9 21.4	129.0 18.8

 $<sup>^{</sup>a}D = \Sigma (|R_{p} - R_{o}| + 1)^{2}$ , where the subscripts p and o refer to predicted and observed values, respectively.

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#### Parameterization of APSIM for Systems Simulations

The systems simulations were performed on the residue retention treatments of the Mackay Late and Mackay Early experiments. The experiments commenced with planting in July 1992 for the Early experiment and November 1992 for the Late experiment. The sites were managed similarly, differing only in the harvest date. Crops were grown for 12 months then harvested and allowed to ratoon (i.e. regrow), four and five times for the Early and Late experiments, respectively. Information on fertilization and tillage practices was used to specify crop management in the model. Model parameters were based, wherever possible, on measured data. The soil profile was sampled to 1.5 m depth in 1997, following the residue decomposition measurements, and soil total C and N, mineral N and bulk density measured to initialize N and C in APSIM-SoilN. Measurements of microbial biomass (Robertson and Thorburn, Chapter 3.1) were used to set the biom pool size. All other pools were set equal to values used in previous APSIM simulations of soil N dynamics in sugarcane residue retention experiments (Thorburn et al., 1999). The soil water parameters (saturation, dul and ll15) were estimated from the daily soil moisture data. Default parameter values (Keating et al., 1999) for the sugarcane variety grown in the experiments (O124) were used.

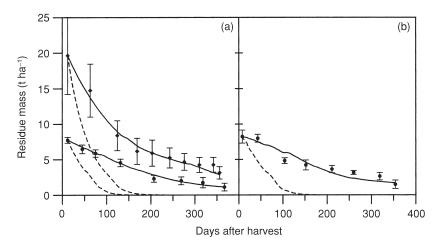
#### **Results and Discussion**

#### Optimization of decomposition parameters

The optimal APSIM-Residue parameter values differed from the default values (Table 2.4.1) and gave predictions of residue mass much closer to measured values (Fig. 2.4.1a) with consequently lower D values (Table 2.4.1). When the optimal values were used to predict residue mass at the third experiment, Harwood Early, predictions of residue mass were also much closer to measured values than those resultant from the default parameters (Fig. 2.4.1b).

The optimal value of  $E_{\rm os,\ max}$  was lower than the default (Table 2.4.1), indicating a greater moisture limitation to decomposition, whereas the value of  $F_{\rm contact,\ min}$  indicated no residue—soil contact limitation. This latter difference was unexpected as initial residue masses were large (10–20 t ha<sup>-1</sup>) in the field experiments, a condition under which residue—soil contact should be a limit to decomposition. It may be that a more appropriate function is required to describe the contact limitation properly.

Another substantial change in the parameter values was the value of  $r_{\text{max}}$ , which was reduced from the default value of 0.1 to 0.007 in the optimization (Table 2.4.1). Since the limitation to decomposition imposed



**Fig. 2.4.1.** Comparison of measured (symbols; bars represent ± 1 sD about the measured values) and predicted (lines) decomposition of sugarcane residue following harvest of a crop. The solid lines are the predictions using the parameter values optimized on the data from the two experiments shown in (a) (◆ − Mackay Late, ● − Mackay Early), while independent predictions for the third experiment (Harwood Early) are shown in (b). The dashed lines are the predictions using the default parameter values (Probert *et al.*, 1998b).

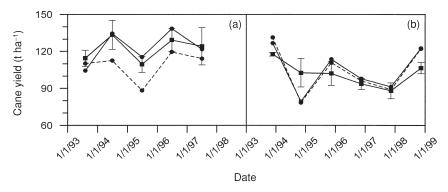
by the high C: N of the sugarcane residue was implicit in the optimal value of  $r_{\rm max}$ , the optimal value is not directly comparable with the default. However, the value of  $F_{\rm C:N}$  (from Equation 3a) was ~0.5 at both the Mackay experiments, implying an  $r_{\rm max}$  value of ~0.014 without the C: N limitation, a value that is still substantially lower than the default. Snow *et al.* (1998) also had to reduce the default value of  $r_{\rm max}$  (to 0.025) to simulate the decomposition of litter in a *Eucalyptus* plantation accurately. These experiences suggest that the value of  $r_{\rm max}$  and other parameters in Equation 2 are not yet generic across the wide range of environments (e.g. semiarid temperate areas to the humid tropics) and agricultural systems (cereals, legumes, sugarcane and forests; both rain-fed and irrigated) where APSIM is being applied.

## Impact of different decomposition rates on cropping system simulations

In simulation of the Mackay Early experiment, the different residue decomposition rates produced by the different parameterization schemes substantially affected simulated sugarcane yield (Fig. 2.4.2a). In all but the first crop, the simulation with the optimized parameter scheme gave higher yields, closer to those measured in the experiment. The effect of

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decomposition on yield was likely to have been caused by impacts of decomposition on the water balance in the simulation (Table 2.4.2). The simulations with the optimal decomposition parameters averaged lower runoff and soil evaporation, and higher deep drainage as would be expected with the greater residue mass throughout the season (e.g. Fig. 2.4.1). These differences were not present in the first crop (data not shown) that was planted into a bare field. Average simulated nitrate leaching from the soil was higher with the optimal parameter scheme (Table 2.4.2) due to increased deep drainage and changes in the patterns of N mineralization (i.e. reduced immobilization of N with the slower decomposition) in the simulation. However, there was little difference in denitrification. Overall, the differences in simulated loss of N were not great enough to affect the crop.



**Fig. 2.4.2.** Comparison of measured (■; bars represent ± 1 sp about the measured values) and simulated (●) sugarcane yields at two experiments; (a) Mackay Early and (b) Mackay Late. Simulations were performed with two parameterization schemes, the default parameters (Probert *et al.*, 1998b; dashed lines) and the result of the optimization (solid lines).

**Table 2.4.2.** Effect of different residue decomposition parameter schemes on simulated components of the water balance and nitrogen losses, averaged across all seasons, at the two Mackay field experiments.

Experiment/ scheme	Water balance (% of rainfall) Nit				trogen losses (kg ha <sup>-1</sup> )	
	Deep drainage <sup>a</sup>	Runoff	Soil evaporation	Nitrate leached	Denitrification	
Early/default	11	13	22	1	26	
Early/optimized	14	11	12	5	24	
Late/default	18	12	21	8	61	
Late/optimized	19	11	20	14	60	

<sup>&</sup>lt;sup>a</sup>Below the active rooting depth.

Contrary to the situation in the Mackay Early experiment, simulated vields (Fig. 2.4.2b) and water balance (Table 2.4.2) in the Mackay Late experiment were little affected by the different decomposition of residues. The difference in the impact of the parameter schemes between the two experiments can be attributed to the timing of residue decomposition in relation to climate at the sites and, primarily, its impact on soil evaporation. Differences in residue mass simulated with the two parameter schemes were greatest from ~100 days after harvest (Fig. 2.4.1). In the Mackay Late experiment (harvested in November each year), this time occurred in the cooler, drier months of late autumn and winter, when potential soil evaporation was low in the Mackay Late experiment, and so the presence (or absence) of residue would have the least impact on soil evaporation. However, the situation was reversed in the Mackay Early experiment (harvested in July each year). There the greatest differences in simulated residue mass occurred in the hotter, wetter months of late spring and summer when potential soil evaporation was high. It is under these conditions that the presence of residue has the greatest impact on soil water balances, and hence vields.

The difference in the simulation results between the two experiments highlights the interactions between climate, crops and soil processes and their impact on cropping system characteristics. Successful use of cropping system models to extrapolate the results of agronomic experiments to different soil, management and/or climatic conditions requires that all these interactions are captured and described adequately.

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### Questionable Assumptions in Current Soil Organic Matter Transformation Models

2.5

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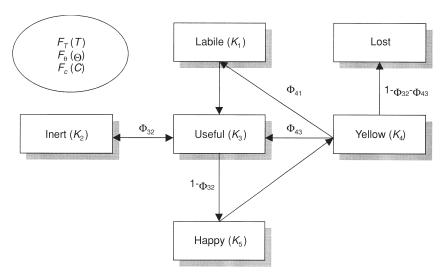
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#### Introduction

Cycling of soil organic matter (SOM) lies at the heart of all terrestrial (agro)ecosystems, controlling the direction and nature of soil–atmosphere trace gas exchange, modulating nutrient availability, and thereby influencing productivity. Current SOM transformation models, especially those designed for incorporation within larger ecosystem-scale models, almost universally assume (i) a soil which is areally homogeneous with respect to all relevant processes and (ii) first-order decomposition kinetics. In this chapter, we analyse these assumptions and examine the conditions which must be satisfied for them to hold. We go on to suggest that in order to test these assumptions it is necessary to depart from a third universal supposition, that (iii) of the existence within the soil of well-defined and functionally discrete SOM pools. Measurements of the transformations of measurable (extractable) SOM fractions are essential in order to secure the foundations on which existing transformation models are built.

It will be clear from the notation that Fig. 2.5.1 represents no published model. It is a caricature, intended to highlight the arbitrary and questionable aspects of the type of model it portrays. Allowing for a few pools more or less, and a little more sophisticated treatment of flux partitioning, nearly all existing SOM transformation models can be represented by a diagram similar to Fig. 2.5.1. We have no space here to mention all the major current contenders; the reader is referred to Powlson *et al.* (1996) for a review. The major apparent exception is the cohort model of Ågren and Bosatta (1987), in which individual additions of organic matter are tracked

Typical SOM model



**Fig. 2.5.1.** A typical SOM transformation model. Pool decomposition rates are determined by intrinsic rate constants  $K_i$  and extrinsic rate modifiers F; partition functions  $\Phi_{ij}$  are shown where required by the reaction network.

through time as their reactivity declines according to a pre-defined pattern. This model, however, becomes mathematically equivalent to the others wherever a sufficient range of (usually geometrically distributed) pool reactivities ( $K_i$ ) is considered. It is conceptually distinct but, given enough pools, functionally equivalent to the majority. It too assumes areal homogeneity and effective first-order kinetics.

Transformation processes within these models are typically represented by some form of Equation 1:

$$V_{i} = K_{i}F_{i}(T, \Theta, C)Y_{i}$$

$$\tag{1}$$

with partitioning between products being handled by some form of equation [2]:

$$\mathcal{J}_{ij} = \Phi_{ij}(T, \Theta, C); \sum_{i \neq 1} \Phi_{ij}(T, \Theta, C) = 1$$
 (2)

In these equations,  $V_i$  is the decomposition rate of substrate  $Y_i$ ,  $Y_i$  is its concentration and  $K_i$  its effective first-order decomposition rate constant;  $F_i$   $(T, \Theta, C)$  is a modifier function representing the effects on  $Y_i$  decomposition of temperature (T), volumetric moisture content  $(\Theta)$  and clay fraction (C);  $\mathcal{J}_{ij}$  is the flux from source pool i to target pool j, and  $\Phi_{ij}$   $(T, \Theta, C)$  is the corresponding partition function. Upper case symbols  $(T, \Theta, C, Y_i, F_i, V_i, V_i)$ 

 $\Phi_{ij}$ ,  $\mathcal{J}_{ij}$ ) denote properties and functions of depth z and time t integrated over the unitary area of the model (usually at least 1 m², commonly as much as  $10^4$  m²). In most models,  $K_i$  is an invariant property of SOM pool i; in the cohort model, it is a function of the age of the cohort. Equations 1 and 2 are actually over-generous in the degree of flexibility they accord to most existing SOM transformation models. Rate modifier functions  $F_i$   $(T, \Theta, C)$  are almost universally assumed to be simple products of individual modifier functions  $F_T$  (T),  $F_{\Theta}$   $(\Theta)$  and  $F_C$  (C) equally applicable to all reactions, while partition functions  $\Phi_{ij}$   $(T, \Theta, C)$  are generally taken to depend only on the clay content C.

#### **Underlying assumptions**

Equations 1 and 2 invoke soil properties, concentrations and reaction rates averaged over areas of 1 m<sup>2</sup> or more. Microbial biomass does not feature in the formulae. Yet (i) SOM decomposition is known to be microbially mediated and (ii) microbial populations and metabolic reaction rates are known to be markedly heterogeneous over distances considerably smaller than 1 m. How are these apparent contradictions to be resolved?

#### Spatial variability and reaction kinetics

Consider a microsite small enough to be treated as homogeneous. Let T,  $\theta$ , c and  $y_i$  represent microsite temperature, moisture content, clay fraction and substrate concentration (and T,  $\Theta$ , C and  $Y_i$ , as before, their macroscale equivalents). Let  $\mu_i$  represent the microbial biomass responsible for bringing about  $Y_i$ -decomposition, with local (microsite) rate  $v_i$ . In general:

$$v_i = v_i(T, \theta, c, y_i, \mu_i)$$
(3)

(i.e.  $v_i$  is a function of T,  $\theta$ , c,  $y_i$  and  $\mu_i$ ) and the corresponding macroscopic rate  $V_i$  is:

$$V_{i} = \int v_{i}(T, \theta, c, y_{i}, \mu_{i}) p(T, \theta, c, y_{i}, \mu_{i}) d(T, \theta, c, y_{i}, \mu_{i})$$

$$\tag{4}$$

where  $p(T, \theta, c, y_i, \mu_i)$  is the probability of an individual microsite having the particular set of controlling properties T,  $\theta$ , c,  $y_i$  and  $\mu_i$ . Assuming – and it is quite a big assumption – that the microsite-scale influences of temperature, moisture content and clay fraction can be disentangled from those of biomass and substrate supply, Equation 3 may be written:

$$v_i = f_i(T, \theta, c)g_i(y_i, \mu_i)$$
(5)

and Equation 4 becomes:

$$V_{i} = \int f_{i}(T, \theta, c)p(T, \theta, c)d(T, \theta, c) \int g_{i}(y_{i}, \mu_{i})p(y_{i}, \mu_{i})d(y_{i}, \mu_{i})$$
(6)

or, equivalently:

$$V_{i} = \overline{f_{i}(T, \theta, c)g_{i}(y_{i}, \mu_{i})}$$

$$(7)$$

where the macron (over-line) denotes integration over the appropriate probability distribution (area averaging). Equating the two macroscale Equations 1 and 6–7 reveals the following assumptions implicit in Equation 1:

$$F_{i}(T, \Theta, C) \equiv F_{i}(\overline{T}, \overline{\theta}, \overline{c}) = \overline{f_{i}(T, \theta, c)} \equiv \int f_{i}(T, \theta, c) p(T, \theta, c) d(T, \theta, c)$$
 (A1) and:

$$K_{i}Y_{i} \equiv K_{i}\overline{y_{i}} = \overline{g_{i}(y_{i}, \mu_{i})} \equiv \int g_{i}(y_{i}, \mu_{i})p(y_{i}, \mu_{i})d(y_{i}, \mu_{i})$$
(A2)

Assumption A1 can be true only where microsite temperature, moisture content and clay fraction are homogeneous, in which case  $F_i$  is by definition equal to  $f_i$ . Under any other circumstances (e.g. in particular, where physical aggregation leads to higher clay fractions and moisture contents in some microsites than in others), there is no simple relationship between  $F_i$  and  $f_i$ . Instead, the macroscale function depends on the probability distributions of the microsite properties T,  $\theta$  and c. Macroscale functions  $F_i$  appropriate for one particular distribution of microsite properties may therefore be inappropriate for another. Macroscale partition functions  $\Phi_{ij}$  are similarly vulnerable.

The assumption that there is one set of rate modifier functions  $F_i$  (and partition functions  $\Phi_{ij}$ ) appropriate for all soils under all climatic and cultivation regimes amounts to an assumption that the probability distributions of the microsite properties T,  $\theta$  and c are always and everywhere roughly the same.

Similar considerations apply with regard to the microscale distribution of substrate concentration  $y_i$  in assumption A2. An additional difficulty concerns the elimination of microbial biomass  $\mu_i$  from the macroscale formula. Two possible scenarios (pictures of microbial life in soil) can be invoked to justify this first-order approximation. The first – probably the most general where the assumption is questioned at all – is of a universal and diffuse war of all against all; all possible microbial life strategies are everywhere present, the most efficient inevitably prevails, successful microbial populations expand until their substrate becomes limiting. The longer the model time step and the larger the simulated area, the more credible such a picture becomes.

What if we want to model short-term SOM dynamics on a time scale of days or weeks (relevant to the crop, but also within the range of microbial population fluctuations)? Can we then make the assumption that potential microbial strategies are universally distributed, and that – basically – whatever makes thermodynamic sense will occur? The evidence seems to be against it (J.M. Tiedje, Los Baños, Philippines, 1999, personal

communication). Spot measurements of microbial DNA point to widely different populations (with, presumably, widely different life strategies) occupying adjacent microsites and exhibiting little short-term mobility. This scenario may lead to first-order kinetics provided that microbial microsites (i.e. population nodes) are sufficiently small and sufficiently diverse that an adequate range of life strategies is represented even within the larger physicochemical microsites defined by approximately uniform  $y_i$ . Most of the microsite volume will then effectively be inert, activity being confined to, and substrate-limited within, those population nodes fortunate enough to be equipped with the relevant enzymes. A corollary of the second scenario is that macroscopic reaction rates must be very much smaller than incubation optima.

Whichever is the case, it should be noted that the first-order assumption is only that: an assumption the validity of which depends on the time and space scales of the model within which it is employed.

#### **Functionally Defined SOM Pools**

Figure 2.5.1 is a caricature. It nevertheless captures one essential feature of the type of model discussed so far: model SOM pools are defined arbitrarily in terms of their effective (macroscale) first-order reaction constants  $K_i$ . These parameters are not amenable to measurement or optimization; they are simply defined as part of the definition of the model. Equally, model pool names – while generally indicative of model pool function – provide no clue for quantification (try to imagine an extraction procedure which reliably isolates all and only that fraction of the soil organic matter with an effective turnover time of 1 day). We have seen that the macroscopic rate modifiers  $F_i$  and partition functions  $\Phi_{ij}$  could not in general be deduced from microscale measurements even were the microscale properties T,  $\theta$ , c,  $y_i$  and  $\mu_i$  measurable, and that  $y_i$  cannot be measured even in principle.

How then is such a model to be evaluated? Its primary predicands (the various SOM pool concentrations  $Y_i$ ) cannot even be measured.

The answer is that SOM transformation models are typically designed to be incorporated into larger models with components representing energy, water, plants, even animals, and driven by various climatic and anthropogenic inputs. They are evaluated not by reference to their primary predicands but in terms of the overall behaviour of the system-level model within which they are embedded. It should be obvious that this provides plenty of scope for fortuitous fits. In view of the load they are expected to bear – forecasts of C sequestration and release over hundreds of years, predictions of indigenous N supply and sustainability under different management regimes – it is clearly desirable that the assumptions which

underlie most SOM transformation models should be tested and shored up wherever possible. How is this to be done?

#### **Prognosis**

Isotope tracing offers the potential for tracking C and N fluxes between measurable (i.e. real) SOM fractions. Where <sup>15</sup>N and <sup>13</sup>C are employed in tandem, it should be possible in theory to measure the fluxes between up to five fractions including the gaseous and solution phases (Arah, 2000a). This in turn should allow the assumptions (approximations) of effective spatial homogeneity and first-order reaction kinetics to be examined in detail. Such work seems to us to be essential.

It calls for the following: (i) development of a fractionation procedure which partitions soil organic matter into five routinely measurable fractions, the first gaseous, the second soluble and the other three defined solely by their method of extraction, such that fractions 1–5 add up to the total organic content of the sample, elemental and isotopic contents of each fraction can be determined, and, ideally, fractions 1–4 are reasonably dynamic over a typical cropping cycle; (ii) incubation of <sup>15</sup>N-and <sup>13</sup>C-labelled soil samples with SOM fractionation and fraction analysis at intervals; and (iii) interpretation of the results via some kind of a model in which <sup>(13)</sup>C and <sup>(15)</sup>N fluxes are linked, to derive the actual C and N fluxes between the measurable (indeed measured) SOM fractions of the incubation. Sohi *et al.* (2000) describe such a fractionation procedure; Arah (2000b) describes such an interpretative model.

Comparison of the results of such an exercise against the constant first-order-reactivity and effective spatial homogeneity assumptions implicit in all current SOM transformation models might do one of three things. It might reveal these assumptions to be unacceptable, necessitating the development of a new generation of models in which more attention is paid to soil structural and microbial kinetic parameters. It might throw up no major contradictions, allowing a relatively seamless shift from the dependence of existing models on functionally defined, unmeasurable SOM pools to a dependence on similarly uniform but procedurally defined, measurable SOM fractions and thereby permitting the development of predictive soil tests for SOM function. Or, most likely, it might fall somewhere between these extremes, suggesting circumstances in which either response is appropriate.

## Acknowledgements

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# A Procedure for Isolating Soil 2.6 Organic Matter Fractions Suitable for Modelling

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## Introduction

Jenkinson and Rayner (1977), and many others since, have modelled the turnover of soil organic matter (SOM) in terms of discrete conceptual pools, each with different characteristic properties and a measure of reactivity (e.g. the first-order reaction constant k). Although the decomposition of plant material in soil is a complex process, such simple models have successfully described the long-term dynamics of SOM.

If a SOM turnover model is to predict nitrogen (N) transformations as well as carbon (C) fluxes, it is necessary to include a functional characteristic of organic matter that determines N mineralized or immobilized during turnover of C. A typical example of such a functional characteristic is the C: N ratio of a conceptual pool. The application of models based on conceptual pools with a first-order reaction constant and a fixed definition of function make for easy modelling but parameters (e.g. initial pool sizes) which are unmeasurable. The overall performance can be optimized against system outputs such as soil C content.

There are drawbacks to such models. Parameterization against systemlevel outputs alone cannot guarantee process-level accuracy. Perhaps the most important drawback is that such models are not directly testable: it is impossible to devise a procedure that will reliably extract all and only that part of the SOM with a first-order reaction constant k. For the same reason, it is not possible to make a measurement in an unknown field and predict future SOM decomposition. Recognition of these limitations has fuelled a debate as to whether we should aim to 'model the measurable' rather than 'measure the modellable' (Elliot et al., 1996; Magid et al., 1997).

If a model is based on measurable fractions, SOM pools are defined as fractions isolated by a specified experimental procedure rather than their reactivity. This has the potential advantage that the size of a SOM pool can be measured in any soil at any time. It has the corresponding drawback that the reactivity of an SOM fraction cannot simply be assumed to be constant.

## **Requirements of the Extraction Protocol**

If the C and N fluxes predicted by the model are to be verified, the number of pools that can be included in the model, and the associated extraction protocol, is determined by the number of tracers employed. Consider a fractionation procedure that divides SOM (including its mineral component) into n fractions. Over any given time period in a closed system, there are n (n-1) carbon fluxes between these fractions, and the same number of nitrogen fluxes. These fluxes represent 2 n (n-1) unknowns for each incubation period. If C is traced by  $^{13}$ C and N is traced by  $^{15}$ N, we have 4 n knowns at each stage of the incubation. Assume, finally, that C and N fluxes are interdependent. If this is the case, then at each stage of the incubation we have n (n-1) unknown fluxes and 4 n known differences.

Therefore, if <sup>13</sup>C and <sup>15</sup>N tracers are used, it should be possible to infer the transformations, and hence derive the effective reactivity, of up to five SOM pools. Therefore, a key requirement of our extraction protocol was that a maximum of five pools should be isolated and that these should account for the total C and N in the system. It is also desirable that the fractionation process should be simple and quick and that the protocol/modelling strategy should identify fractions which differ significantly in measured chemical properties. This difference in chemical properties is important because, as a first approximation, differing chemical properties infer likely differences in reactivity.

## The Extraction Method

We developed an extraction protocol (Sohi et al., 1998) and are currently developing and testing an associated model. In brief, the pools extracted are as follows; fraction 1 is the gaseous phase; fraction 2 is the soluble fraction; fraction 3 is isolated by suspending soil in dense liquid, with no prior energy input (other than swirling) referred to as free material herein; fraction 4, denoted intra-aggregate, comprises material that is released by ultrasonic disruption of aggregates and recovered by a second density separation and;

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fraction 5 is the residual organomineral component. This can be divided further on the basis of particle size.

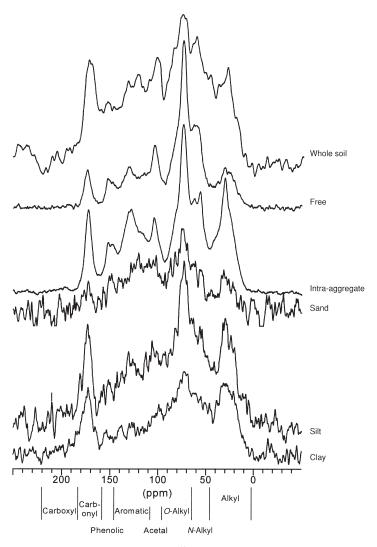
## Chemical composition of solid fractions

Chemical characterization of the solid fractions (fractions 3, 4 and 5 above) was done by  $^{13}$ C cross-polarization magic angle spinning (CPMAS) NMR. Material in the organomineral component was divided further on the basis of size into sand, silt and clay fractions. Full experimental details are published elsewhere (Gaunt et al., 1999). Figure 2.6.1 shows the chemical composition of the solid organic matter fractions separated by our fractionation procedure and that of whole soil, using the <sup>13</sup>C CPMAS NMR technique (Sohi et al., 1998). Carbon in free organic matter was located predominantly in O-alkyl structures. Intra-aggregate organic matter contained a lesser proportion of C in O-alkyl groups, and more in aromatic and alkyl groups. Lowering of O-alkyl to alkyl ratios is characteristic of the early stages of the degradative process (Preston, 1996). This is attributable to the transformation of readily metabolizable carbohydrates, and production and persistence of biomass- and plant-derived alkyl C. Our analysis suggests, therefore, that the free fraction represents less altered organic matter (i.e. closer to the composition of incoming plant material) as compared with the intra-aggregate.

The distribution of NMR-visible carbon for the clay organomineral fractions appears more similar to that of intra-aggregate rather than free organic matter (with slightly greater proportions of carbonyl C, less aromatic and phenolic C). The observed differences between the free and the intra-aggregate and organomineral fractions are potentially important, since the chemical properties of a particular fraction will affect its reactivity. We have found that these differences are consistent across soil type and climate in soils taken from seven long-term experiments, where cereals are grown under mineral fertilization (Gaunt *et al.*, 1999).

## **Isotope Tracing Through Fractions**

If the proposed fractionation protocol and associated model is to be used to predict C and N fluxes over a single crop-growing season, we need to measure, and predict, fluxes of C and N over a period < 1 year. To establish the sensitivity of our fractionation protocol, we set up incubation studies using  $^{13}$ C and  $^{15}$ N tracers. To trace C, we used a  $^{13}$ C natural abundance technique. The stable isotope composition of plants differs depending on the type of photosynthetic pathway ( $C_3$  and  $C_4$ ) of the plant. This results in a difference in the  $^{13}$ C content of 12-14%, with  $C_4$  plants being more



**Fig. 2.6.1.** Chemical composition as seen by <sup>13</sup>C NMR of whole soil and organic matter fractions for a clay texture arable soil from the UK.

enriched. Thus we added  $C_4$  plant material (maize) at approximately -12% <sup>13</sup>C labelled with <sup>15</sup>N to a  $C_3$  soil at approximately -26% <sup>13</sup>C.

Figure 2.6.2 shows an example of the change in the  $\delta^{13}$ C signal of the solid fractions defined by our protocol over a period of ~1 year after incorporation of maize material in pots (unplanted). The disappearance of maize-derived C from the free fraction due to decomposition was accompanied by a small, but detectable, increase of maize-derived material

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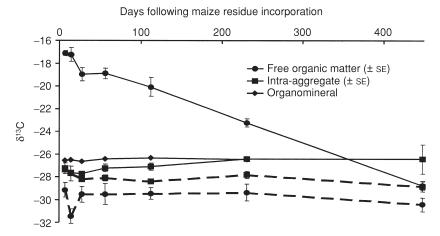


Fig. 2.6.2. The  $\delta^{13}C$  signal of soil organic matter recovered from a  $C_3$  soil after addition of  $C_4$  plant residues. Dashed lines represent the unamended control.

in the intra-aggregate. The appearance of maize-derived material in the organomineral fraction cannot be seen; this may be either because this fraction is less important to describe short-term turnover of C or because the large amount of C in this fraction masks the incorporation of maize-derived material. Results for <sup>15</sup>N show similar patterns and thus are not shown here.

The difference between the  $\delta^{13}C$  enrichment at natural abundance of the free and intra-aggregate fraction in the unamended control soil may reflect isotope discrimination during the decomposition of organic matter. The products of microbial transformation are generally enriched in the heavier isotope form when compared with the substrate. Thus the fact that the intra-aggregate material is less depleted in  $^{13}C$  than the free fraction, and therefore heavier, appears to confirm the results above that suggest that the free fraction represents less altered organic matter when compared with the intra-aggregate.

## Conclusion

An extraction protocol that meets the basic requirements for a model based on measurable pools is presented. The solid organic fractions obtained by our physical fractionation procedure (i.e. free and intra-aggregate fractions) show strong contrast in chemical composition. These differences have been found to be consistent across environment and soil type.

Preliminary isotope data suggest that we can measure fluxes of added C through the pools defined in the model. Thus we can derive the reactivity,

in situ, of the organic matter in these pools. We plan to use the model to test the assumption of first-order reactivity used by most models. Further, the combined modelling and fractionation approach presented provides a unique opportunity to relate the chemical characteristics of soil organic matter to its in situ reactivity. Work to establish these relationships is planned.

## **Acknowledgements**

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# Soil Organic Matter Management: The Roles of Residue Quality in C Sequestration and N Supply

3

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## **Soil Organic Matter Management for What Purpose?**

The transformations of residues into soil organic matter are regulated by three interacting factors, the physicochemical environment and resource quality acting through decomposer communities (Swift *et al.*, 1979). Hence the quantity, the quality and the functional attributes of organic residues are key elements for managing decomposition of soil organic matter (SOM) and nutrient release. Here we will focus on the roles of SOM for different management purposes paying particular attention to the time scale within which potential interactions occur (Table 3.1). The choice of organic resource quality is governed primarily by a particular management objective and resource availability. Hence the classification of resource quality into 'high' and 'low' is somewhat arbitrary depending on the desired effect. For example, residues of low C: N ratio, low lignin and polyphenol content can be termed 'high' quality when fast nitrogen release is required, but the same residues are 'low' quality if the desired outcome is C sequestration.

## The Role of Residue Quality in C Sequestration

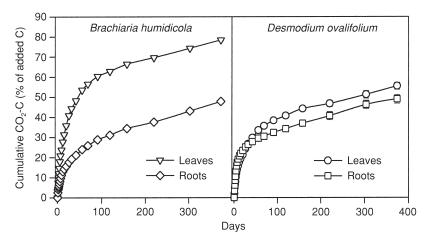
Atmospheric C budgets that ignore the possibility of terrestrial ecosystems to sequester C do not balance, and indicate that there is a 'missing sink' of ~0.4–4 Gt C year<sup>-1</sup> (Gifford, 1994). It has been suggested that the terrestrial biosphere (vegetation and soils) can act as a source as well as a sink of organic C. Land clearing, mostly by tropical deforestation, has been

**Table 3.1.** Choice of plant quality attributes for different management purposes.

Objective	Time scale	Optimal residue quality
C sequestration	Long-term	High lignin and condensed tannins
Erosion control	Immediate	High N, lignin and condensed tannins, structurally protected
Microclimate	Immediate	High N, intermediate lignin and condensed tannins
Weed suppression	Immediate	Low N, intermediate lignin, alkaloids, hydrolizable polyphenols, allelopathic substances
CEC	Immediate	Large specific surface area
Soil pH (Al toxicity)	Intermediate	Cation to anion ratio
Reduce pollution	Immediate	Low soluble C and N, high in active polyphenols (PBC)
Nitrogen supply	Immediate	low C: N ratio, low soluble C, low lignin: N, low in active polyphenols (PBC)
	Medium-term Synchrony	low C : N ratio, intermediate lignin and condensed tannins high C : N with high soluble C, mixed residues

PBC = protein-binding capacity.

estimated to release between 1.5 and 3 Gt C year<sup>-1</sup>. Fisher et al. (1995) suggested that improved pastures which replace native savannas throughout South America could account for an additional sequestration of 100-500 Mt C year<sup>-1</sup> in these tropical soils. Boddey et al. (2000) also found increases in soil C under 9-year-old improved pastures after rainforest clearing in Brazil but the magnitude was lower, and soil C under degraded pastures declined. Substantial soil C inputs may be attributed to the deep-rootedness of grasses in improved tropical pastures (Cadisch et al., 1994). Indeed, 75% of the claimed increased C sequestration was found below 20 cm soil depth and is thus likely to be due to root inputs. Fisher et al. (1997) found that the large increase in SOM under improved tropical pastures (up to 70 t ha<sup>-1</sup>) was associated with a substantial increase in the C: N ratio, giving ratios in the SOM of 33: 1 compared with usual SOM values of ~12:1. It is thus likely that only partial decomposition of roots occurred leading to the increased SOM content. Extrapolation from a fitted double exponential decay model to laboratory incubation data of tropical pasture materials suggested that between 43 and 47% of legume root C and 54-62% of grass roots was theoretically 'non-decomposable' (Urquiaga et al., 1998). Similarly, our recent data showed that after incubation of root materials for 1 year, < 50% of C had been evolved through respiration (Fig. 3.1) whereas 80% of C of leaves of the tropical grass Brachiaria humidicola had been lost. The factors determining residue C degradation in soils are governed by residue quality. Brachiaria and legume roots had a higher C: N ratio and higher lignin content than leaves (Schweizer et al., 1999). Lignin is known to be recalcitrant and to protect cellulose from microbial



**Fig. 3.1.**  $CO_2$  evolution of incubated leaves and roots of the tropical grass *B. humidicola* and tropical legume *D. ovalifolium*. Verticals bars are  $\pm$  1 SEM.

attack due to its entrapment within the cell walls (Chesson, 1997). Thus the lignin: N ratio is widely known to govern residue decomposition of many plant materials and is also used to allocate residue fractions to the slow decomposing structural pool in many models such as CENTURY (Parton *et al.*, 1987).

## **Condensed Tannins-Protein Complexes: A Direct Route to Soil Organic Matter?**

Tropical legumes are well known as a source of a wide variety of polyphenols and of other secondary metabolites (Waterman and Mole, 1994). The importance of polyphenols in decomposition and nitrogen release from the leaves and litters of tropical legumes has been demonstrated in both herbaceous forage species (e.g. Desmodium intortum, Vallis and Jones, 1973) and legume trees (e.g. Acacia, Calliandra, Inga and Peltophorum, Palm and Sanchez, 1991; Constantinides and Fownes, 1994; Handayanto et al., 1994) and summarized by Giller et al. (1998). The combined (lignin + polyphenol): N ratio is now widely recognized as a useful predictor of decomposition and N release for many plant materials (Constantinides and Fownes, 1994; Handayanto et al., 1994). However, the total amount of extractable polyphenols does not necessarily reflect the activity and recalcitrance of these organic residues. The ability of polyphenols to bind protein has been established as a better indicator of short-term N release of tropical legume pruning materials (Handayanto et al., 1995). Longer term implications of polyphenol-protein interactions have been investigated by

Cadisch *et al.* (1998). The residues with high protein-binding capacity released the smallest amounts of N over the second and third crop cycles, giving no indication that smaller N release over the first crop was compensated for in the longer term (Fig. 3.2). Repeated additions of the same prunings at planting of each successive crop led to an increase in N recovery with the prunings of low protein-binding capacity (e.g. *Gliricidia sepium*). However, N recovery from *Peltophorum dasyrachis* prunings, rich in active polyphenols, was reduced even further, again indicating a long-term effect of such species on stabilization of N in the soil organic matter.

The formation and stability of polyphenol–protein complexes is dependent on a number of factors relating to the structure and mass of both the protein and the polyphenol involved. Not only the number of phenol moieties is important, but their stereochemical arrangement and the spatial arrangement of the component molecules within the polymer (Hagerman, 1992). In general, the affinity of polyphenols for proteins increases with molecular mass (Ohara, 1994). Both reversible and irreversible interactions

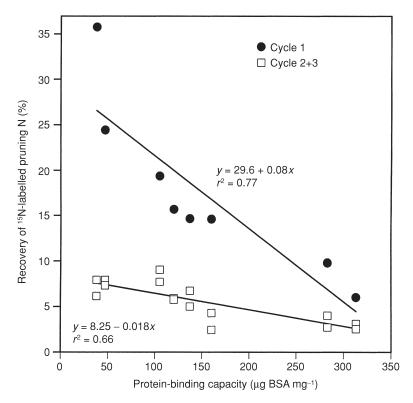
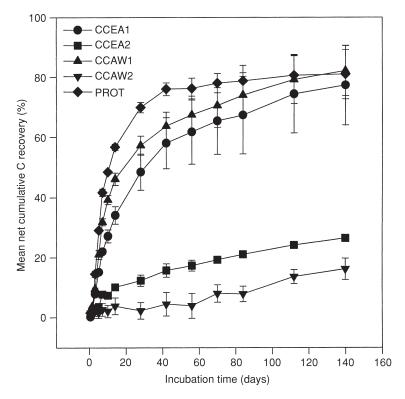


Fig. 3.2. Indirect evidence of role of active polyphenols on long-term N release (Cadisch *et al.*, 1998).

between polyphenols and proteins occur. Irreversible interactions are based on the covalent binding of polyphenols to proteins resulting in complexes which are significantly more stable than those formed by the reversible interactions of hydrogen bonding and hydrophobic association. The interactions occur via the transformation of phenols to quinones, highly reactive intermediates which subsequently react with nucleophilic groups on proteins or other macromolecules to form covalently bound complexes (Beart et al., 1985). Since the conditions required for this irreversible interaction are relatively mild, it is entirely reasonable to predict the occurrence of covalently bound polyphenol-protein complexes in situations such as the microbial decomposition of plant tissues in the soil environment (Haslam, 1989). This is essentially the well-known 'polyphenol theory' of humus formation. The chemical structure of the recalcitrant forms of SOM (the 'humic substances') reveals the presence of linked aromatic rings, and phenolic acids are released when strong oxidation methods are employed (Haynes, 1986). This, and other evidence, led to the 'polyphenol theory' of soil organic matter formation which postulated that phenolic compounds formed by degradation of lignins, together with microbially derived phenols, are oxidized to quinones which polymerize with amino acids and other products of degradation to form 'humus'. Although this theory fails to account for the wide variety of structures found in the humic substances (in particular the large amount of aliphatic C chains), the role of phenolic compounds in reacting with other organic compounds to form stable complexes is generally accepted (Wild, 1988).

Whetton (1999) investigated the difference in decomposition of polyphenol-protein complexes where polyphenols were extracted from leaves of Calliandra calothyrsus and Leucaena leucocephala. The extracted polyphenols were purified by a series of steps including partitioning between ethyl acetate (EA) and water (AW), column chromatography using Sephadex LH20 and preparative high-performance liquid chromatography (HPLC). Although all of the polyphenol fractions had the ability to precipitate protein and had some degree of protein binding, analysis of the crude polyphenol fractions showed that those fractions with very high condensed tannin content and those with the highest molecular weight were the most active in protein binding. In direct comparison, fractions which contained condensed tannins of large molecular weight were shown to be some 20-fold more biologically active than those containing simple flavonoids. Complexes formed from the polyphenol fractions of Calliandra and Leucaena leaves and crude leave proteins (CLP) of G. sepium were added to an acid upland soil from Sumatra. The complexes formed from small molecular weight polyphenols, such as flavonoids, procyanidins and anthocyanins (fractions EA1 and AW1, Fig. 3.3), and CLP were found to degrade rapidly in the soil. Microbial respiration was only slightly reduced when compared with that of CLP alone, with 80% of added C recovered



**Fig. 3.3.** C decomposition from polyphenol–protein complexes from *Calliandra calothyrsus* (see text for explanation of fractions) (adapted from Whetton, 1999).

within the first 6-8 weeks. Complexes formed by polyphenol fractions which were more biologically active in binding proteins (fractions EA2 and AW2) were considerably more recalcitrant. Complexes formed with high molecular weight condensed tannin extracted from C. calothyrsus (AW2) resulted in only 6% net recovery of C via microbial respiration, even after a period of 16 weeks. Corresponding complexes using polyphenols from either Calliandra or Leucaena showed similar decomposition patterns (not shown). However, a specific L. leucocephala fraction (EA2) containing condensed tannins, with molecular weights up to 1500-1600 Da, showed an intermediate degree of recalcitrance, with a linear pattern of degradation over the 16-week period, and a final net C recovery of 50-60%. Thus resistance to microbial degradation of polyphenol-protein complexes in the soil correlated well with their protein-binding or precipitation activity. While some of these complexes had relatively short half-lives, others appeared to be very recalcitrant to microbial attack and may provide a 'fast route' to increase SOM contents.

## Consequences of Increased C Sequestration for N Cycling

Fisher et al. (1997) clearly demonstrated that the large increases in SOM under improved tropical pastures (up to 70 t ha<sup>-1</sup>) were only possible with a substantial increase in the C: N ratio of the SOM. The amounts of N that otherwise would have to be immobilized at the commonly found soil C: N ratio of ~10 or 12:1 would have exceeded by far the soil N supply and would have caused severe immobilization. To increase the C sequestration potential or to improve synchrony of nutrient release from residues with crop N demand, incorporation of 'lower' quality residues (or mixtures) is often promoted. These residues decompose slowly, thereby conserving nutrients, but nutrient release is lower during a given crop cycle. There is little nutrient release over subsequent cycles regardless of initial residue quality, indicating that there is little compensation for initial lower N release from slow decomposing residues (Cadisch et al., 1998). Repeated applications of residues of 'low' quality (high N, lignin and/or polyphenols) should, however, result in build up of soil organic matter and associated increased nutrient release over time. We tested this hypothesis using experimental and modelling approaches to evaluate the time frame within which such a potential compensation effect occurs. Decomposition data were taken from Handayanto et al. (1995) who derived N mineralization rate constants for G. sepium and C. calothyrsus from single exponential decay functions (Table 3.2). Using a single exponential decay model, the change in remaining residue N at equilibrium is (Jenkinson, 1981):

$$dN/dt = 0$$

or:

$$N_{Input} - N_{Equilibrium} \times k = 0$$

**Table 3.2.** Single exponential decay model output values at equilibrium level of repeated applications of legume pruning at a rate of 75 kg N ha<sup>-1</sup> 3 months<sup>-1</sup>.

Species	N	Time	Litter	Accumulated	Mineralization
	Mineralization	to	N	mineral N	rate at
	rate constant	equilibrium	equilibrium	(385 weeks)	equilibrium
	(week <sup>-1</sup> ) <sup>a</sup>	(weeks)	(kg N ha <sup>-1</sup> )	(kg N ha <sup>-1</sup> )	(kg N week <sup>-1</sup> )
Gliricidia sepium	0.074	68	78	1982	$5.8 \pm 3.0$
Calliandra calothyrsus	0.040	125	144	1922	$5.8 \pm 1.3$
Calliandra calothyrsus	0.015	333	385	1698	$5.8 \pm 0.6$

<sup>&</sup>lt;sup>a</sup>From Handayanto et al. (1995).

where k is the mineralization rate constant. Thus the remaining residue N at equilibrium is:

$$N_{\text{Equilibrium}} = N_{\text{Input}}/k$$

Given the amount of residue N remaining at equilibrium, the average mineralization rate (not including fluctuations due to very recent inputs) can be calculated as:

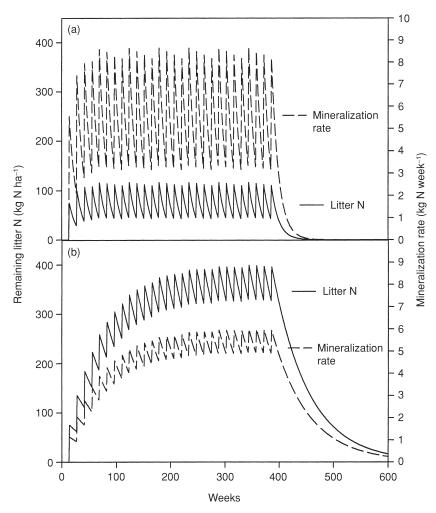
$$N_{\text{Mineralization rate}} = N_{\text{Equilibrium}} \times k$$

An interesting feature of this equation is that it indicates that the mineralization rate at equilibrium is the same for all residues if they are applied at the same rate of N (Table 3.2). If we define the time to equilibrium ( $t_E$ ) as the time required for the mineralization rate to reach ~99% of the maximum mineralization at equilibrium then  $t_E$  can be expressed as (Olson, 1963):

$$t_{\rm E} = 5/k$$

For the investigated legume pruning materials, the time to equilibrium ranged between 68 and 333 weeks for the fast and slow decomposing prunings, respectively (Table 3.2). Thus a farmer would have to wait ~6 years until the average N supply rate of the slowly decomposing prunings is equivalent to that of the fast decomposing prunings, or ~4 years if we consider a 95% achievement of the maximum rate.

The simulated instantaneous mineralization rates, however, reveal that there are agronomically relevant significant differences between the prunings despite the average rate being the same. The instantaneous mineralization rate of the fast decomposing Gliricidia residue fluctuated widely even at equilibrium (Fig. 3.4a). Hence, synchronizing N release with crop N demand remains important. In contrast, the instantaneous mineralization rate of the slowly decomposing Calliandra prunings varied little (Fig. 3.4b). This more constant N release at equilibrium poses less risk for farmers as a more predictable N release is achieved. A further important feature for crop production is that the N supply power from the slowly mineralizing materials remains high between applications. These advantages are associated with an increased organic matter in the soils as suggested by the much larger undecomposed residue N remaining (Fig. 3.4b). The use of slow decomposing materials apparently provides a further advantage in that it is more 'buffered' against withdrawl of continuous residue application. This reduced risk for farmers is associated with the larger stock of remaining residue N in the soil. Long-term alley cropping experiments in Indonesia revealed that the fertility benefit of the slowly decomposing Calliandra and Peltophorum residues was superior to that of the fast decomposing Gliricidia in the seventh year (van Noordwijk et al., 1997).



**Fig. 3.4.** Modelled temporal variation derived from a single exponential decay model using repeated applications of legume pruning at a rate of 75 kg N ha<sup>-1</sup> 3 months<sup>-1</sup>. (a) *Gliricidia sepium* with N mineralization rate constant k = 0.074 week<sup>-1</sup> and (b) *Calliandra calothyrsus* with N mineralization rate constant k = 0.015 week<sup>-1</sup>.

We recognize that this simplified model approach has limitations and needs verification. However, it revealed potentially important long-term implications of systems with different residue qualities in nutrient supply patterns.

## Consequences of Residue Quality Attributes for Quantification of C Sequestration

Differences in the  $^{13}$ C isotopic signatures between  $C_3$  and  $C_4$  species have been used to quantify the contribution of a newly introduced species to soil organic matter, e.g. tropical pastures following rainforest clearing, using the relationship (Cadisch *et al.*, 1996):

$$f_{\rm g(G)} = \frac{\delta^{13} C_{\rm (G)} - \delta^{13} C_{\rm (RF)}}{\delta^{13} C_{\rm (g)} - \delta^{13} C_{\rm (RF)}}$$

where  $f_{g(G)}$  is the proportion of SOM derived from the C<sub>4</sub> species,  $\delta_{(G)}$  the  $\delta^{13}$ C value of the soil under the pure grass pasture (G),  $\delta_{(RF)}$  the  $\delta^{13}$ C value of the soil under  $C_3$  rainforest (RF) and  $\delta_g$  the  $\delta^{13}C$  value of SOM derived from the C<sub>4</sub> species plant material (g). The <sup>13</sup>C signature of SOM entirely derived from the C<sub>4</sub> species under investigation is, however, often not known and the isotopic signature of bulk plant material is used instead. As the <sup>13</sup>C signature of SOM under a particular vegetation is often close to the value of the  $\delta^{13}$ C of its vegetation, this assumption appears to hold (Tieszen and Boutton, 1989). However, plant materials are not uniformly labelled in <sup>13</sup>C (Benner et al., 1987). Recalcitrant plant fractions such as lignin are more depleted in <sup>13</sup>C compared with the bulk plant material (Schweizer et al., 1999; Table 3.3). These materials, due to their slow degradation, may accumulate during decomposition, and hence the <sup>13</sup>C signature of the soil organic matter formed may have a different <sup>13</sup>C signature from that of the original bulk plant material. For the correct use of the <sup>13</sup>C isotopic method for decomposition and SOM studies, it is therefore necessary to know the extent of possible isotopic fractionation which occurs during

**Table 3.3.**  $\delta^{13}$ C signatures of bulk plant and lignin material (adapted from Schweizer *et al.*, 1999) and their potential implications for soil organic matter origin estimations.

	Total plant $\delta^{13}$ C (‰)	Lignin C δ <sup>13</sup> C (‰)	Potential error in grass-derived C (%)
Brachiaria humidicola			
Leaves	-11.4	-16.1	+20a
Roots	-11.9	-16.6	+22a
Desmodium ovalifolium			
Leaves	-27.3	-29.8	-7 <sup>b</sup>
Roots	-25.8	-27.8	-6 <sup>b</sup>
SEM	0.1	0.1	

<sup>&</sup>lt;sup>a</sup>On rainforest soil ( $\delta^{13}$ C = 28‰) (Cadisch *et al.*, 1996).

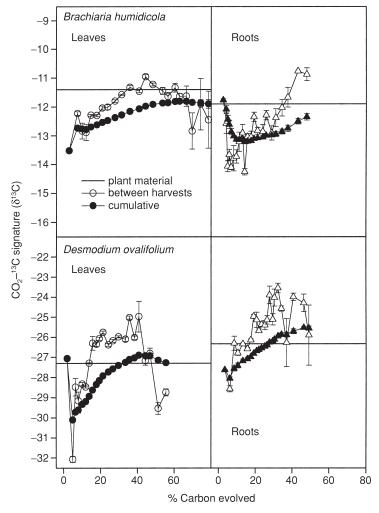
<sup>&</sup>lt;sup>b</sup>On a C<sub>4</sub>-derived soil ( $\delta^{13}$ C = 12‰).

decomposition. We investigated these effects for  $C_3$  and  $C_4$  pasture species introduced in the Latin American savannas. As expected, the grass materials (*B. humidicola*) were less depleted in  $^{13}$ C ( $\delta^{13}$ C -11.4 to -11.9%) than those of the legume (-27.3 to -25.8%) (*D. ovalifolium*) (Table 3.3; Schweizer *et al.*, 1999). Plant lignin C was strongly depleted in  $^{13}$ C compared with the bulk material by up to 2.5‰ in the legume and up to 4.7‰ in the grass. If lignin is a major source of material remaining as 'humus', this would lead to potential errors in the estimation of grass- or legume-derived SOM of 6–22%.

These plant materials subsequently were incubated for 1 year. Significant depletion in <sup>13</sup>C of the evolved CO<sub>2</sub> was observed during the initial stages of decomposition (Fig. 3.5) probably as a result of microbial fractionation as it was not associated with the <sup>13</sup>C signatures of the measured more-decomposable fractions (non-acid detergent fibre and cellulose). The cumulative CO<sub>2</sub>-13C signatures of both grass and legume materials tended to converge towards the <sup>13</sup>C signature of the original bulk material. Analysis of the residual material after 1 year of incubation showed no significant change in the <sup>13</sup>C signature from that of the original bulk material (Table 3.4). Hence, during the incubation time of 1 year, the recalcitrant, <sup>13</sup>C-depleted lignin did not exert a major influence on the isotopic signature of the remaining material, providing evidence against a direct lignin pathway to SOM formation. It is thus likely that lignin is at least partly degraded and that the formation of new SOM is largely the result of microbial transformations. On the other hand, isotopic effects due to decomposer growth rate, efficiency and stabilization of the degradation products increase <sup>13</sup>C concentration in the remaining material (Agren et al., 1996). Although full evaluation of the potential errors associated with isotopic fractionation may necessitate several years of incubation due to the slow decomposition of the more recalcitrant fractions (Fig. 3.1), the importance of <sup>13</sup>C-depleted lignin as a source of errors in calculating the origin of soil organic matter may have been overestimated in the past.

## Conclusions

Definition of residue quality attributes ('high' or 'low') should vary according to the desired management objective (e.g. C sequestration or crop N supply). Soil C sequestration is favoured by lignin and other high molecular weight secondary plant metabolites. Plant roots seem to have a particularly important role in soil C sequestration in productive tropical pastures. Plant quality attributes also affect isotopic carbon signatures of chemical fractions, but <sup>13</sup>C-depleted lignin in residues resulted only in minor errors in identification of sources of SOM within 1 year. Protein–condensed tannin complexes appear to provide a direct route to C sequestration and



**Fig. 3.5.** Cumulative and instantaneous <sup>13</sup>CO<sub>2</sub> signatures of decomposing grass (*B. humidicola*) and legumes (*D. ovalifolium*) leaf and root materials.

SOM build up, but the slow initial N release from lignin- and tannin-rich residues is not compensated by an increased N supply later.

## Acknowledgement

We wish to thank Jon Fear for stable isotope analysis.

	Decomposition time		
	0 day	119 days	374 days
		$\delta^{13}$ C	
Desmodium ovalifolium			
Leaves	-27.3	-27.2	-27.1
Roots	-25.8	-25.8	-25.5
Brachiaria humidicola			
Leaves	-11.4	-11.7	-11.8
Roots	-11.9	-12.1	-12.2
Average SEM	0.1	0.2	0.2

**Table 3.4.** Changes in  $\delta^{13}$ C signatures of plant materials during decomposition.

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## Crop Residue Effects on Soil C and N Cycling under Sugarcane

3.1

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## Introduction

In the Australian sugar industry, there is a progressive move away from burning sugarcane (*Sacharum* spp.) trash (green and senesced leaves) before and/or after harvest (burnt system), to the green cane trash blanketing (TB) system, where trash is retained on the soil surface and cultivation is greatly reduced. Adoption of the TB system is seen as an important move towards a more sustainable production system. When sugarcane trash is burnt, > 80% of the organic matter and nutrients in the trash are lost (P. Larsen and R. Mitchell, unpublished data). Conversely, with retention of trash, nutrients and organic matter may be accumulating in the soil, which has led to speculation that fertilizer applications should be reduced. The effects of these management changes on the fertility of the canelands, however, are largely unknown. This research was undertaken to investigate the effects of sugarcane trash management (burnt or TB) on indicators of short- and long-term soil C and N cycling. The implications of this cycling for fertilizer management are discussed.

## Methods

Burnt and TB systems were compared in five field experiments at three locations with widely differing climates and varying crop ages (Table 3.1.1). The experimental treatments, established at the time of planting, were (i) burnt: trash burnt, soil cultivated to 150 mm depth after harvest and

	Harwood	Mackay	Tully
Climate	Temperate	Tropical	Tropical
Latitude, longitude	29.50 S, 153.20 E	21.10 S, 149.07 E	17.56 S, 145.56 E
Annual rainfall (mm) (30–50 year mean)	1021	1668	4067
Crop age	1st ratoon (late harvest) 2nd ratoon (early harvest)	3rd ratoon (late harvest) 4th ratoon (early harvest)	6th ratoon (late harvest)
Soil texture	Clay/loam	Loam	Clay
Previous cropping	Vegetables, sugarcane	Sugarcane	Sugarcane

**Table 3.1.1.** Summary of the field experiments.

(ii) TB: sugarcane harvested without burning and trash retained on the soil surface, soil not cultivated. The Mackay and Harwood sites each had one experiment harvested early in the harvesting season (July) and one experiment harvested late (December). The experimental designs were randomized complete blocks with three or four replicates. In all cases, the variety was Q124, row spacing was 1.5 m and annual N fertilizer application was 180 kg ha<sup>-1</sup> as urea (applied 1–6 weeks after harvest). At the time of this study, the experiments had been running for between 1 and 6 years, having been ratooned (above-ground portions harvested, remainder left to regrow) annually.

To measure trash decomposition in the TB treatments, a standard mass of trash was placed (on the soil surface) within sufficient quadrats  $(0.75 \times 1.50 \text{ m})$  to allow destructive sampling of one quadrat per replicate on 8–10 occasions over the following (12-month) growing season. The quadrats were held in place by tent pegs, and covered with 20 mm mesh netting.

At each sampling, trash that had not become incorporated with the soil (termed 'free trash', current year's trash only) was sampled first. Soil was then sampled within the quadrat by taking 12 cores (30 mm diameter) in two regular transects across the row. The cores were cut into the depth layers (0–20, 20–50, 50–100 and 100–250 mm) and samples pooled from each depth. Trash that had become incorporated with the surface 10 mm of soil ('incorporated trash', current year's trash and older trash) was then sampled by removing a strip (1750 mm long  $\times$  175 mm wide  $\times$  approximately 10 mm deep) of soil between the holes left by the soil sampler.

Soil microbial biomass C was determined by chloroform fumigation extraction (Vance *et al.*, 1987). This method gave some negative results when C from the unfumigated control was subtracted from C from the fumigated soil. The quantity of C from fumigated soil was highly correlated with (C from fumigated soil – C from unfumigated soil) (r = 0.88-0.95 for all data from individual experiments, negative results excluded), therefore

the fumigated values are presented as an index of microbial biomass C. Soil N and C mineralization potentials were measured as the change in inorganic N and the release of carbon dioxide, respectively, during a 7-day laboratory aerobic incubation at 25°C at standardized soil water content. Soil inorganic N (ammonium + nitrate) was determined by potassium chloride extraction (Bundy and Meisinger, 1994). Total N in soil was determined by Kjeldahl digestion (Bremner and Mulvaney, 1982) and automated colorimetric analysis. Organic C in soil was determined by dichromate oxidation (Heans, 1984). Total N and C in trash were determined using a Leco induction furnace. All soil analyses were done without removing incorporated trash. Incorporated trash was separated from the soil by washing the sample in water and removing floating and suspended organic matter on a 2 mm sieve.

### Results

## Trash decomposition

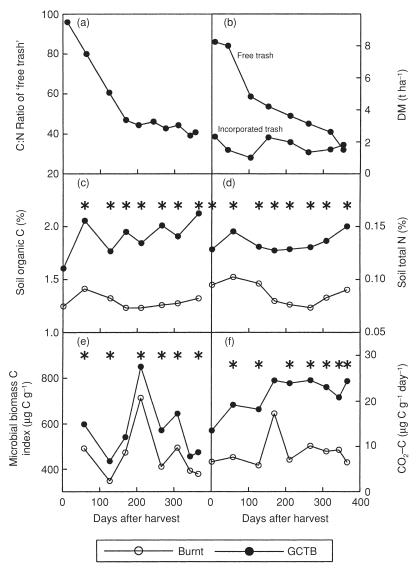
After harvest, 7–12 t ha<sup>-1</sup> of new trash dry matter (DM) was present on the soil surface in TB plots. After 1 year, 98% of the free trash had decomposed at Tully and 82–91% at Mackay and Harwood. The free trash returned after harvest contained 3–5 t C ha<sup>-1</sup>, 28–55 kg N ha<sup>-1</sup> and had a C: N ratio of 70–117. During the season, the C: N ratio declined to ~40 in late-harvested crops and 23–29 in early-harvested crops (e.g. Fig. 3.1.1a). After 1 year, 2–16% of trash C and 5–37% of trash N remained in free trash (i.e. 50–800 kg C ha<sup>-1</sup> and 1.3–20 kg N ha<sup>-1</sup>). Incorporated trash contained 1–2.6 t DM ha<sup>-1</sup>, 370–860 kg C ha<sup>-1</sup> and 12–27 kg N ha<sup>-1</sup> (e.g. Fig. 3.1.1b). There were no apparent trends in the mass of incorporated trash with time. By the end of the year, as much as or more C and N was in incorporated trash than in free trash at all sites.

## Soil properties

Soil organic C, total N and microbial biomass C (e.g. Fig. 3.1.1c–e) were greater under TB than under burning in the older experiments (Mackay and Tully), but only in the 0–20 and 20–50 mm depth layers. At Harwood, neither organic C, total N nor microbial biomass were significantly affected by trash management.

At most sampling dates, C mineralization potential (e.g. Fig. 3.1.1e) was increased by TB in the 0–50 mm depth, but not at greater depths. Unlike C mineralization, soil N mineralization potential generally did not differ significantly between trash management treatments (data not

shown), as variability among replicates was large. However, it tended to be lower in TB soils than burnt soils from 0–20 mm at Mackay and Tully (mean  $0.14 \mu g N g^{-1} day^{-1}$  in TB, and  $0.32 \mu g N g^{-1} day^{-1}$  in burnt soils).



**Fig. 3.1.1.** Representative data from the field experiments: (a) C: N ratio of free trash from Harwood-Late; (b) dry matter of free and incorporated trash from Harwood-Early; (c) soil organic C (0–50 mm) from Tully; (d) total soil N (0–50 mm) from Tully; (e) microbial biomass index (0–50 mm) from Tully; and (f) C mineralization potential (0–50 mm) from Tully. Significant treatment differences marked \* (P < 0.05).

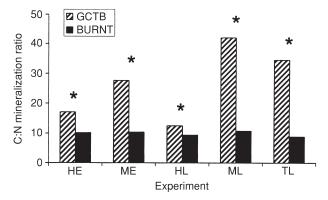
The ratio (C mineralization : N mineralization) in the 0–50 mm depth was greater under TB than under burnt management (e.g. Fig. 3.1.2).

Soil inorganic N content was extremely variable, and did not show consistent differences between trash management treatments. Inorganic N concentration increased dramatically after fertilizer application, and declined to a low level within ~2 months.

## **Discussion**

Although large quantities of C were returned to the soil under TB, most of it was lost through respiration during the following year, so that the proportion remaining in the soil (< 22%) was not much greater than the proportion that remains after trash is burnt (2–20%, as ash and incompletely burnt leaves, P. Larsen and R. Mitchell, unpublished data). However, trash retention did gradually increase total soil C and stimulated microbial growth and activity as crop age increased. In contrast to the biologically labile nature of the C returned under TB, the C remaining after a fire probably contained a significant quantity of charcoal-like material that was biologically inert (Skjemstad *et al.*, 1999). Thus, the quality of C was probably higher under TB.

A greater proportion of trash N was retained in the soil than trash C, so that TB increased total soil N and organic C in similar proportions (C by 7–21% and N by 8–24% in the top 100 mm). Net mineralization of N, however, was not increased by TB, presumably due to stimulated N immobilization in the presence of the increased labile C supply. This hypothesis is supported by the higher ratio (C mineralization: N



**Fig. 3.1.2.** The effect of burnt or TB management on the ratio of [C mineralization: (N mineralization + 1)] from 7-day aerobic incubations of soils (0–50 mm depth). Data are means from all sampling dates. Experiments indicated by ML, Mackay-Late; TL, Tully-Late; ME, Mackay-Early; HL, Harwood-Late; and HE, Harwood-Early. Significant treatment differences marked \*(P < 0.05).

mineralization) in TB than in burnt soils. This difference between the treatments is likely to have been even greater under field conditions, where surface trash (which was excluded from the laboratory incubations) would have contributed additional C to the soil, thereby increasing the ratio in TB soils. Our results are consistent with observations of reduced soil N availability in the first few years of residue conservation (Thompson, 1992).

The magnitude of the trash effect on soil properties in the different experiments depended on the cumulative amount of trash returned to the soil. Cumulative returns of trash DM were estimated as 11% of the fresh sugarcane yield from past years, this percentage being derived from the regression of trash DM against sugarcane yield in our five experiments. Cumulative returns of C and N in trash were estimated by assuming the same trash C and N concentrations as measured in our experiments. Regression of the increase in soil C or N due to TB against the cumulative C or N returns gave coefficients of determination ( $r^2$ ) of 0.94 for soil organic C, 0.80 for microbial biomass C, 0.97 for C mineralization potential and 0.94 for total soil N (n = 5, overall mean from each experiment). Retention of trash C and N in the soil varied among experiments, however, with 10–20% (mean 13%) of cumulative C returns measured as soil organic C and 40–100% (mean 75%) of cumulative N returns measured as total soil N.

The rates of accumulation and mineralization of C and N under TB measured in these experiments can only be considered indicative of the first crop cycle (5–6 years) after conversion from a burnt to a TB system. At the end of a crop cycle, sugarcane soils are normally cultivated several times to 150–200 mm depth, the effect of which may be to reduce differences in soil C and N between burnt and TB treatments (e.g. Thorburn *et al.*, 1999). Furthermore, rates of C and N accumulation in TB systems must be expected to decrease with time and reach an equilibrium level, as measured and simulated in sugarcane systems (Thorburn *et al.*, 1999), and is known to happen in other crop and pasture systems (e.g. Jenkinson, 1991). Accumulation rates decrease because mineralization and loss of C and N from the soil (through respiration, leaching, denitrification and plant uptake) increase.

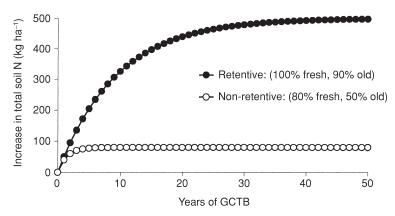
In order to explore a range of possible responses of soil C and N to trash blanketing, we combined measured rates of decomposition and accumulation with assumptions about the mineralization of C and N from trash left from previous years, and calculated equilibrium C and N balances for the top 250 mm of soil at each site. We chose two decomposition scenarios: (i) a 'retentive' system, where for each crop, 100% of trash N and 20% of trash C is retained in the soil in the year following harvest, and 90% of the remaining trash N and 85% of the remaining trash C is retained in subsequent years; and (ii) a 'non-retentive' system, where 80% of trash N and 10% of trash C from each crop is retained in the soil in the year

following harvest, and 50% of this trash N and 60% of this trash C is retained in subsequent years. The effects on soil C and N were calculated (no allowance made for fallow effects), until the systems had reached equilibrium. At the start, the Mackay and Tully soils contained 40,000 kg C ha $^{-1}$  and 2400 kg N ha $^{-1}$ , and the Harwood soils contained 76,000 kg C ha $^{-1}$  and 6500 kg N ha $^{-1}$  (to 250 mm depth).

At equilibrium under the 'retentive' scheme, soil C would have increased by 7–14% after 25 years and soil N (Fig. 3.1.3) would have increased by 8–21% after 35 years. Under the 'non-retentive' scheme at equilibrium, soil C would have increased by 1.4–2.6% after 6 years and soil N would have increased by 1.3–3% after 6 years. Thus, our expectations are for very small to modest increases in soil C and N under long-term TB at these sites.

When equilibrium is attained, maximum mineralization of trash-derived soil N has been reached, and equals annual returns (50 kg ha<sup>-1</sup> year<sup>-1</sup>). This is the maximum amount by which N fertilizer rates could be reduced. In these sugarcane production systems, this point would be achieved after 6 years in the 'non-retentive' system and after 25–35 years in the 'retentive' system. It is possible that fertilizer applications may not ever be able to be reduced to this extent if N loss is greater in TB than in burnt systems. It has been suggested that leaching and denitrification may be promoted under TB due to the increased C and water contents.

The suggestion from this study that trash retention will result in a slight or modest increase in total soil C and N, and a somewhat greater increase in mineralizable soil N in the long term, accords with the findings of Powlson *et al.* (1987), who measured an increase of only 5% in organic C and 10%



**Fig. 3.1.3.** Calculated cumulative increase in total soil N during 50 years of TB management, in a retentive and non-retentive system. See text (Discussion) for more explanation.

in total N, but > 40% in N mineralization and microbial biomass N after 18 years of barley straw incorporation in Denmark. Whilst our approach to prediction of the effects of trash management on soil C and N has provided useful insights, the trash management effects are likely to be more fully understood with the aid of system simulation models such as APSIM (Thorburn *et al.*, Chapter 2.4).

## **Acknowledgements**

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## Effect of Residue Quality on N<sub>2</sub>O Emissions from Tropical Soils

3.2

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## Introduction

One of the potential benefits of N<sub>2</sub>-fixing trees in agroforestry systems is the recycling of nutrients through litter fall or prunings, which become available to crops after decomposition. The quality of the prunings or litter is important in determining nutrient supply and soil organic matter formation. However, crop N recovery from prunings is often < 20% (Giller and Cadisch, 1995), with a lack of synchrony between N release and crop demand, and the potential for substantial N losses. Previous work in temperate systems has shown that incorporation of plant material can significantly increase N<sub>2</sub>O emissions from soils. The magnitude of emissions varies depending on the quality and biomass of added plant material, and also with soil temperature, moisture content, aeration, soil type and cultivation. Greater N<sub>2</sub>O emissions are usually measured after incorporation of material with a low C: N ratio, than material with a high C: N ratio (Baggs et al., 2000). However, for tropical agroforestry residues, lignin and polyphenol contents have a strong influence on determining the availability of N for release (Handayanto et al., 1994) and hence nitrification and denitrification. Emissions from agroforestry systems are of concern, not only due to the loss of valuable N resource from the system, but also due to the role of N<sub>2</sub>O in the greenhouse effect and the destruction of stratospheric ozone.

We report here the results of controlled environment experiments in which N<sub>2</sub>O emissions and N release were measured following amendment of tropical soils with prunings and residues from alley cropping and

improved fallow systems. The effects of C:N ratio, lignin and polyphenol composition on  $N_2O$  emissions were examined during the main period of flux activity.

## **Materials and Methods**

## **Experimental design**

Samples of 50 g of soil and 50 g of acid-washed silica sand were mixed thoroughly and placed in leaching tubes (25 cm length; 4 cm diameter), and 360 mg of dry, chopped prunings (< 7 mm) were incorporated in to the top 5 cm of the soil–sand mixture. Tubes were maintained at 70% of the water holding capacity and incubated at 28°C in the dark.

In the first experiment, leaves of Gliricidia sepium, Calliandra calothyrsus and Peltophorum dasyrrachis were incorporated into a sandy clay loam soil (2.39% C, 0.16% N) from Chitedze, Malawi. Unamended soil–sand mixtures provided controls. Available soil N and N<sub>2</sub>O emissions were measured after incorporation. In the second experiment N<sub>2</sub>O emissions were measured following incorporation of Sesbania sesban leaves and Macroptilium atropurpureum residues into a clay Oxisol (1.50% C, 0.16% N) from Kenya. The effect of residue particle size was examined by comparing emissions from ground and chopped Sesbania leaves.

## **Analyses**

Soil for experiment 1 was sampled from replicate tubes that were not used for gas analysis. Fresh soil was extracted with 1 M KCl in a 1 : 5 ratio of soil to extractant. The concentrations of NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N in the filtered extractant was determined by continuous flow analysis on a Chemlab Instruments autoanalyser. The chemical composition of material (Table 3.2.1) was analysed according to the methods described by Handayanto *et al.* (1994).

## N<sub>2</sub>O fluxes

Tubes used for gas analysis were closed for 1 h with gas-tight lids with a rubber septum. After the hour closure samples were taken using air-tight glass syringes and analysed for  $N_2O$  in a Pye Unicam gas chromatograph fitted with an electron capture detector. Column and detector temperatures were 50 and 250°C, respectively.

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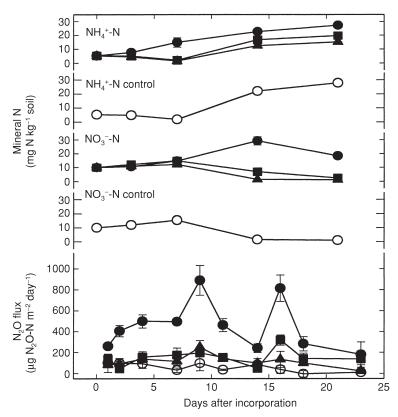
Prunings	N (%)	C : N ratio	Lignin (%)	TEP (%)	PBC (μg BSA mg <sup>-1</sup> )
Experiment 1					
Gliricidia sepium	4.0	12	20	1.3	22
Calliandra calothyrsus	3.6	13	22	3.5	317
Peltophorum dasyrrachis	2.5	20	32	3.9	245
Experiment 2					
Sesbania sesban	3.7	12	5.9	2.4	23
Macroptilium atropurpureum	2.5	16	9.0	2.3	24

**Table 3.2.1.** Chemical compositions of plant material used in this study.

TEP = total extractable polyphenols; PBC = protein-binding capacity of plant extract; BSA = bovine serum albumin.

## **Results and Discussion**

Incorporation of plant material resulted in higher N<sub>2</sub>O emissions (P < 0.05) than from the unamended soil (Fig. 3.2.1; Table 3.2.2). This was probably a result of rapid stimulation of microbial decomposition, possible creation of anaerobic microsites resulting from microbial respiration and the increased C supply and substrate for nitrification and denitrification. The greatest total losses were measured after incorporation of Gliricidia leaves, with 9.7 mg  $N_2O-N$  m<sup>-2</sup> (P < 0.005) emitted over 23 days (Table 3.2.2). The rapid release of N (Fig. 3.2.1) from the Gliricidia, providing the substrate for nitrification and denitrification, was related to the high N and low polyphenol contents of this material. N<sub>2</sub>O emissions were positively correlated with available soil N for the first 11 days after incorporation of Gliricidia leaves (r = 0.91 and 0.86 for NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>, respectively; P < 0.005), but thereafter were negatively correlated. Incorporation of Calliandra and Peltophorum prunings resulted in temporary immobilization of N and low N<sub>2</sub>O emissions. Emissions after addition of Calliandra leaves were significantly lower than from the Gliricidia treatment, despite similar C: N ratios of 13 and 12, respectively. Hence lower N<sub>2</sub>O emissions and mineralization from the former prunings was attributed to their higher polyphenol contents and protein-binding capacities. These findings confirm that the C:N ratio alone is insufficient to predict N release and subsequent  $N_2O$  emissions. The polyphenols of Calliandra and Peltophorum had a high capacity to bind the plant N, thereby reducing the availability of N for nitrification and denitrification. Materials with a polyphenol + lignin: N ratio of < 10 are generally found to result in fast N release (Mafongoya et al., 1998). The Peltophorum leaves had a ratio of 14.4, and exhibited low N release and the lowest N2O emissions of the



**Fig. 3.2.1.** Nitrous oxide emissions and available soil N following incorporation of *Gliricidia* (filled circles), *Calliandra* (filled squares), *Peltophorum* (filled triangles) leaves and control (empty circles). Error bars represent  $\pm$  one standard error.

**Table 3.2.2.** Total cumulative emissions of  $N_2O$  (mg  $N_2O-N$  m<sup>-2</sup>).

	Treatment	Total emission (mg N <sub>2</sub> O-N m <sup>-2</sup> )
Experiment 1 (23 days)	Gliricidia sepium	9.7 (1.7)
	Calliandra calothyrsus	3.4 (0.4)
	Peltophorum dasyrrachis	2.6 (0.6)
	Control	1.1 (0.7)
Experiment 2 (29 days)	Sesbania sesban	8.1 (2.4)
	Macroptilium atropurpureum	8.9 (2.3)
	Ground Sesbania	10.0 (3.7)
	Control	5.1 (3.4)

Values in parentheses represent  $\pm$  standard deviation of the mean.

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amended treatments. Emissions from the Kenyan clay soil in the second experiment were higher (P < 0.1) than from the Chitedze sandy clay loam in the first experiment. This may have been due to the development of anaerobic microsites in the Kenyan soil, as a result of its higher clay content (48% as opposed to 28%).

Incorporation of *Sesbania* resulted in rapid N release and very high, but short-lived (2 days), fluxes of N<sub>2</sub>O (data not shown). Such rapid N release from *Sesbania* is undesirable in improved fallow systems as the N availability is often asynchronous with crop demand (Handayanto *et al.*, 1997) and therefore can result in large, immediate gaseous losses. Over 29 days, there was no significant difference in total N<sub>2</sub>O emissions from the *Sesbania* and *Macroptilium* treatments. Grinding (< 2 mm) as opposed to chopping (< 7 mm) of high-quality *Sesbania* leaves had no significant effect on N<sub>2</sub>O emissions. Jensen (1994) found that decomposition of high-quality residues of different sizes varied only slightly. Greater difference between grinding and chopping could be expected following incorporation of low-quality material, by increasing the accessibility of moderately available C to microbial attack.

This work has shown N<sub>2</sub>O production to be influenced by residue polyphenol content and its ability to bind proteins. There is the potential for these losses to be controlled and N to be retained within the system by regulating N release and improving synchrony of N supply with crop demand by manipulating the quality of organic inputs using varying combinations of plant material of differing qualities, prunings of different ages from the same plant and varying combinations of inorganic and organic N sources. This will be examined in future experiments, and N<sub>2</sub>O losses will also be equated with losses by NH<sub>3</sub> volatilization, NO<sub>3</sub><sup>-</sup> leaching and total denitrification. The relative contributions of nitrification and denitrification to the N<sub>2</sub>O fluxes are unquantifiable without the use of selective nitrification inhibitors, or stable isotope techniques. Further process-based studies will be undertaken to verify this.

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3.3

# Evaluation of the Soil Organic Matter Dynamics Model MOTOR, for Predicting N Immobilization/Mineralization Following Field Incorporation of Paper Mill Sludge in a Horticultural Soil

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### Introduction

The UK output of paper mill sludge (PMS) is expected to be 1 million tonnes in the year 2000. About 0.2 million tonnes are spread on farmland as an exempt waste, providing benefit as a soil conditioner and liming agent (Aitken et al., 1988). PMS, typically with a high C: N ratio, can also be used to reduce nitrate pollution of groundwaters through soil N immobilization. However, accurate predictions of N dynamics related to land application of PMS are necessary to maximize N leaching prevention, and to adjust fertilizer N requirement for crops. Such an approach was attempted in an experiment carried out at Dipper Field, Balmalcom Farm (Fife, Scotland), where PMS was applied to a sandy loam horticultural soil. During 1994/95, the addition of PMS to vegetable crop residues in autumn and winter decreased nitrate leaching without impairing the yield of the next crop (Vinten et al., 1998). The present Chapter is focused on N immobilization/mineralization following PMS application in June 1996, and the immediate effects on the following crop N uptake and the residual

soil N in autumn. The results were compared with simulation of C and N dynamics using the MOTOR model (Hassink and Whitmore, 1997; Whitmore *et al.*, 1997). Laboratory studies were run in parallel to provide additional information on N immobilization/mineralization kinetics related to the physical state of PMS (finely mixed materials are expected to be more accessible to decomposers than large pieces) and the soil biological condition. Hypothetically, if initial bacteria growth following PMS addition occurs without grazing by protozoa or nematodes, N mineralization will occur less rapidly (Bouwman *et al.*, 1994), and the net N immobilization phase will last longer.

### **Materials and Methods**

### Field experiment

There were three replicate blocks of the following treatments: W<sub>95</sub>, soil receiving receiving 40 t DM ha<sup>-1</sup> PMS with C: N = 25 on 6 December 1994 (Vinten *et al.*, 1998); W<sub>96</sub>, soil receiving 40 t DM ha<sup>-1</sup> PMS with C: N = 18 on 14 June 1996; and W<sub>0</sub>, control soil with no PMS application. In 1996, the PMS was ploughed in after spreading, and broccoli was transplanted on 4 July. There were sub-plots fertilized with 125 kg N ha<sup>-1</sup> after planting (N1) and unfertilized sub-plots (N0). Marketable yield and total N and C content were determined. After harvest, the crop residues were left on the surface. Soil samples (0–20 cm) were taken 14, 35, 53, 68, 91, 124, 152, 185 and 213 days after the PMS application. Inorganic N content and microbial biomass C and N (Voroney and Paul, 1984) were determined. Soil was sampled to 2 m on 14 November 1996 and water content, NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N content were determined. Emissions of N<sub>2</sub>O were measured on the N1 treatments using automated cover boxes (Scott *et al.*, 1999) from 2 July to 20 August 1996.

### Laboratory studies

The role of the physical state of the PMS on decomposition processes was investigated in the laboratory, focusing on respiration kinetics. Soils samples were collected from Balmalcolm (Hexpath series, loamy sand), and from a nearby organic horticultural farm (Fourmerk series, sandy loam). Duplicate treatments were established in 11 kilner jars with a vial containing 10 ml of 0.5 M NaOH. Treatments consisted of 35 g of fresh soils thoroughly mixed with 5 mg C g<sup>-1</sup> soil added in the form of: (i)  $0.5 \times 0.5$  cm filter paper squares; (ii) 1 cm diameter spheres of filter paper; (iii) 1 cm diameter spheres of filter paper inoculated with dilute soil

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suspension; and (iv) cellulose powder. A control treatment consisted of fresh soil with no C addition. The jars containing the treated soil were incubated in the dark at 20°C. Alkali traps were changed at 1, 3, 9 and 23 days, and analysed for C content by titration. Nematode counts were made on initial soils, and on soils from a parallel incubation after extraction by flotation (Griffiths *et al.*, 1990).

### **Model description**

MOTOR is an extension of the model concepts of Jenkinson and Rayner (1977) and of the SUNDIAL model (Bradbury *et al.*, 1993). Crop residue C is split into decomposable plant material (DPM) and resistant plant material (RPM). The decomposition rate of the RPM decreases according to the formulation of Parton *et al.* (1987), to allow for the lignin content  $(L_s)$ :

$$K_{\text{RPM}} = K_{\text{DPM}}^{\text{e}(-3L_{\text{g}})} \tag{1}$$

We assumed that in the absence of lignin, the RPM pool (cellulose + chemical addititives) decomposes with the same kinetics as DPM since, following the waste treatment process, enzymes should be available in the PMS for immediate decomposition of cellulose in soil. Crop residue organic C decomposes to microbial biomass (BIO), non-protected organic matter (NOM) and  $CO_2$ . The assimilation efficiency was taken as 60%. Inert organic matter (IOM) is determined in part by the charcoal content of the soil. NOM decomposes to  $CO_2$  and microbial biomass. Microbial biomass turns over to form NOM which in turn may become protected in soil as POM (protected organic matter) with a rate  $K_a$ . POM must first be desorbed (rate constant  $K_d$ ) into the NOM pool before it can decompose (Table 3.3.1). The maximum size of the POM pool (X) is defined by soil texture (Hassink and Whitmore, 1997).

Microbial biomass decays by second-order, not first-order kinetics:

$$d(BIO)/dt = k_2\alpha(1 - \alpha)BIO^2$$
 (2)

where BIO = biomass C,  $\alpha$  = proportion of biomass that consists of primary and secondary consumers (e.g. protozoa and nematodes) and  $1 - \alpha$  = proportion of biomass that is made up of detritivores (bacteria, fungi, etc.). Where  $\alpha$  is reasonably constant, the expression  $k_2\alpha(1-\alpha)$  is also nearly constant, and equation 3 can be simplified to:

$$d(BIO)/dt = k_2' BIO^2$$
 (3)

Whitmore (1996) derived a value for  $k_2$ ' based on fitting Equation 3 to measured soil biomass N and C. PMS-derived biomass is highly labile,

Pool	Decay constant day <sup>-1</sup>	Initial pool size mg C kg <sup>-1</sup> soil	C : N	N mg kg <sup>-1</sup> soil
PMS addition				
RPM	0.035 <sup>a</sup>	3,820 <sup>b</sup>	_	28
DPM	0.035°	652 <sup>d</sup>	4.3	150
Mineral N				88
Initial soil conditions	1			
BIO	0.007	117	4	_
NOM	0.0012	1,000	10	_
POM	0.003 (K <sub>a</sub> )			
	0.00004 (K <sub>d</sub> )	10,292	15	_
Χ		22,975	15	_
IOM	0	597	17	_
Mineral N				10

**Table 3.3.1.** Decay constants, C and N contents for PMS addition (RPM and DPM), initial (day 165) mineral N and soil organic matter conditions (BIO, NOM, POM and IOM) for MOTOR.

unlike stabilized soil biomass, and is assumed to have the same turnover time as plant DPM (Table 3.3.1).

A study on PMS composition (Gilboa *et al.*, 2000) showed that the total N content of the PMS from the mill used in this study ( $N_{tot}$ ) was made up of urea N (~30% of  $N_{tot}$ ), organic additive N (2%),  $NH_4^+$ -N (8%), and biomass N and metabolites (60%). We used this information to estimate the size of the pools of N and C applied as PMS.

### **Results and Discussion**

On N0 plots,  $W_{96}$  caused a complete loss of marketable yield, although total N uptake was not affected. However, on N1 plots, there was a highly significant (P < 0.01) positive effect of  $W_{96}$  on marketable yield and a significant effect (P < 0.05) on N uptake (Table 3.3.2). There was a trend (P < 0.1) to higher N uptake and marketable yield with  $W_{95}$  application than with the control. Residual soil N was significantly higher in the  $W_{96}$  treatment than in the control. There was a small residual effect of  $W_{95}$  and a large effect of  $W_{96}$  on the  $N_2O$  emissions. Although the C:N ratio of the PMS was smaller than desirable to promote immobilization and prevent N leaching, these results suggest that initial immobilization of mineral N may have hampered initial crop development in the N0 plots.

<sup>&</sup>lt;sup>a</sup>Value reduced by fibre content according to Equation 2.

bCellulose and chemical additives.

<sup>&</sup>lt;sup>c</sup>Value reduced by fibre content according to Equation 2.

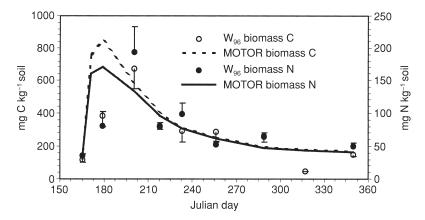
dMicrobial cells and metabolites and chemical additives to paper.

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Table 3.3.2.	Total N uptake and marketable yield of calabrese crop at harvesting time, residual
nitrate N in so	il profile to a depth of 250 mm of pore water storage on 14 November 1996 ( $\pm$ SD,
n = 3).	

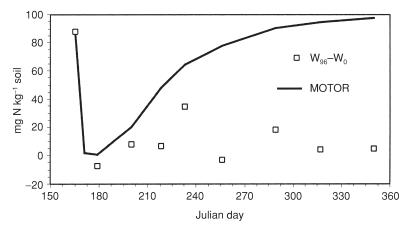
	N uptake (kg N ha <sup>-1</sup> )		Marketable yield (DM ha <sup>-1</sup> )		Residual N (kg N ha <sup>-1</sup> )		N <sub>2</sub> O-N emissions (kg N ha <sup>-1</sup> )	
Treatment	N0	N1	N0	N1	N0	N1	N1	
$W_0$	$79 \pm 8$	$175\pm29$	$5.4\pm2.5$	$9.5\pm1.6$	$14 \pm 2$	$23\pm8$	0.54	
$W_{95}$	$101\pm17$	$201\pm39$	$8.7 \pm 0.7$	$11.0 \pm 4.0$	_	$41 \pm 34$	1.28	
$W_{96}$	$118\pm15$	$234 \pm 41$	$0.0\pm0$	$8.0 \pm 0.9$	_	$37 \pm 4$	15.55	

Nitrous oxide emissions were measured using automated cover boxes sampling eight or four times daily during the period 2 July-20 August 1996 (n = 1).



**Fig. 3.3.1.** MOTOR simulations and measurements of microbial biomass C and N at Dipper field, Balmalcolm Farm after PMS addition of 40 t DM  $ha^{-1}$  (C : N = 18) on day 165; 125 kg N  $ha^{-1}$  applied on day 185.

Soil biomass C and N measurements for the W<sub>96</sub> N1 treatment are compared with MOTOR simulations in Fig. 3.3.1. Peak values of biomass C and N are well simulated. However, the peaks occur up to 3 weeks later than simulated (although more data are needed to confirm this). Biomass C and N in the W<sub>95</sub> treatment were not significantly different from the control treatment (not shown). We calculated the net effect of PMS on soil mineral N by subtracting the mineral N in W<sub>0</sub> plots from W<sub>96</sub> values (Fig. 3.3.2). Values on the day of PMS application were calculated from estimation of the amount of mineral N added with the PMS (Gilboa *et al.*, 2000). The measured remineralization was slower than simulated, but MOTOR simulations do not include crop uptake so simulations cannot be compared directly with field data later in the season (after about day 230). Nonetheless, simulated soil mineral N on 14 November 1996 (212 kg N ha<sup>-1</sup>) was much more than the sum of net (W<sub>96</sub>–W<sub>0</sub>) crop N



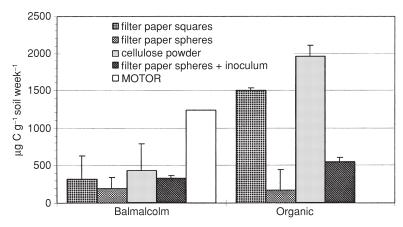
**Fig. 3.3.2.** Net effect ( $W_{96}$ – $W_0$ ) of PMS addition (day 165) on mineral N dynamics simulated by MOTOR compared with field measurements; 125 kg N ha<sup>-1</sup> applied on day 185. MOTOR simulation excludes plant uptake.

uptake, residual soil mineral N and  $N_2O$ -N emissions (88 kg N ha<sup>-1</sup>; see Table 3.3.2). Overall, these comparisons suggest that while the size of the immobilized pool of N was well simulated, the kinetics of both N immobilization and remineralization during PMS decomposition were slower than MOTOR simulates. This may be partly due to shortage of mineral N delaying N immobilization (although MOTOR does allow for this). The soil mineral N content in the  $W_{96}$  treatment on day 179 (prior to fertilizer N application) was very low – only  $2.6 \pm 1.3$  mg N kg<sup>-1</sup> soil compared with  $9.9 \pm 1.3$  mg N kg<sup>-1</sup> soil in the control. The scaling in Fig. 3.3.1 is set so that C and N measurements overlay if biomass C: N = 4. Results show a wider biomass C: N before N fertilizer application (day 185) than immediately afterwards, which also suggests N limitation during initial decomposition.

It is also likely that some physical protection of the cellulose occurs – the PMS was applied as large pieces, which would be colonized only slowly by soil organisms. The laboratory incubation showed that, for both soils, C mineralization was slower for 1 cm diameter spheres than for cellulose powder or filter paper squares (Fig. 3.3.3).

The function used to retard decomposition rates of organic additions in MOTOR needs to account for the decomposition of cellulose in the absence of lignin. In the MOTOR simulations, we assumed that cellulose was 100% DPM on the basis that (i) no lignin was present and (ii) that microorganisms present in the PMS from the waste treatment process would ensure a ready supply of the cellulase enzyme. In the laboratory incubation (Fig. 3.3.3), all forms of cellulose initially decomposed more slowly in the Balmalcolm soil than the MOTOR-simulated soils. However,

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**Fig. 3.3.3.** C emissions during days 3–9 of incubation of experimental soils at 20°C with 5 mg C g<sup>-1</sup> soil as cellulose in various physical forms.

if we assume the cellulose is 100% RPM, immobilization and mineralization are too slow. Dalenbergh and Jager (1989) postulated that cellulose decomposition may show a lag phase as cellulolytic microorganisms need to be activated by supply of soluble C. However, this was not expected to be the case in this work as cellulose decomposers would have the opportunity to develop in the PMS during storage prior to spreading.

Finally, the initial and potential biological activity of the soil will influence the kinetics of decomposition. In the laboratory, incubation decomposition rates for the organic soil were close to or above those predicted by MOTOR, on the cellulose powder and filter paper square treatments (Fig. 3.3.3). Counts in field soil sampled in May 1999 showed that bacterivorous nematode numbers in the Balmalcolm soil (~3 g-1 of soil) were only 20% of those in the organic soil (Sattar, 1999). The ratio of bacterial grazer C to bacterial C at the start of a parallel incubation experiment was 1% for the Balmalcolm soil and 11% for the organic soil (Sattar, 1999). Thus initial grazing of bacteria on the Balmalcolm soil would be slow, extending the period of immobilization. Incubation with cellulose for 4 weeks did not lead to a significant increase in nematode numbers with Balmalcolm soil, but did in the organic soil (Sattar, 1999). The initial value of  $\alpha$  (the proportion of total biomass that consists of primary and secondary consumers) on the Balmalcolm soil was only ~2%, compared with a value of 7% used in the parameter estimation work of Whitmore (1996). This could retard mineralization significantly and make invalid the assumption that  $k_2\alpha(1-\alpha)$  is constant, implicit in the rate constant used for biomass C decomposition. We have made calculations of the expected steady-state mineralization rate of Balmalcolm soil using the food web model of de Ruiter et al. (1993). These show that mineralization during decomposition

of an N-rich residue (C: N = 10) is 50% faster with 15 bacterivorous nematodes per g of soil than with three nematodes per g of soil.

### Conclusion

MOTOR simulated the peak amount of C and N immobilized in biomass following PMS application in June 1996 well, but the prediction of the timing of the peak N immobilization was up to 3 weeks early. We suggest that shortage of soil mineral N, physical accessibility of substrate and level of initial soil biological activity affected the kinetics of C and N immobilization/mineralization.

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### Organic Matter Management in Practice – The Potential to Reduce Pollution

3.4

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### Introduction

Soil nitrogen (N) supply to arable crops can be separated into a background mineral N supply from soil organic matter (SOM) mineralization and a 'pulse' from mineralization of fresh residues (Bjarnason, 1989). Arable crop residues can be an important source of N for the following crop (MAFF, 1994). Residue size depends on the balance between N uptake and N removed from the field in produce, and so varies between crops (Table 3.4.1). The quality of the residue will also influence both the amount of N mineralized and the rate of mineralization. It is clear from the quantity of N that is potentially available (Table 3.4.1) that we need to utilize it as efficiently as possible in the soil—crop system to minimize environmental effects (e.g. leaching of nitrate to water courses). Here we describe experiments that have measured (i) the influence of time and method of residue incorporation on subsequent mineral N release and (ii) the contribution of above- and below-ground residues to soil mineral N, and we discuss the implications for efficient N management in the soil—crop system.

### **Description of Experiments**

Standard measurement methods were used in all experiments. Nitrate leaching was measured using porous ceramic cups (Lord and Shepherd, 1993) and estimated drainage volume (Bailey and Spackman, 1996) below 1 m. Soil mineral nitrogen (SMN) was measured (to 60 or 90 cm) by 2 M

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**Table 3.4.1.** Calculation of N returns in arable crop residues in England and Wales, based on Sylvester-Bradley (1993). Calculations exclude 210,000 ha of 'miscellaneous' crops and 126,000 ha of vegetable crops.

Crop	Area <sup>a</sup> 000 ha	Residue	N index <sup>b</sup>	Yield of produce <sup>a</sup> (t ha <sup>-1</sup> )	N content of produce (kg t <sup>-1</sup> )	Maximum N uptake (kg ha <sup>-1</sup> )	N content of residue (kg ha <sup>-1</sup> )	Total residue N (kt)
Wheat	2036	Straw	0	7.43	17	193	67	136.4
Barley	1358	Straw	0	5.78	17	150	52	70.6
Oats	100	Straw	0	5.41	17	141	49	4.9
Rye	12	Straw	0	5.41	18	141	43	0.5
Triticale	8	Straw	0	5.41	18	141	43	0.3
Linseed	73	Straw	0	1.49	38	75	18	1.3
Sugarbeet	196	Leaf/crown	0	45	1.7	200	124	24.3
Peas	78	Haulm/straw	0.5	3.79	130	265	135	10.5
Beans	99	Haulm/straw	0.5	3.8	155	285	130	12.9
Oilseed rape	446	Haulm/straw	1	3.23	30	240	143	63.8
Potatoesc	16	Haulm	1	23.8	3.5	167	83	1.3
Potatoes <sup>d</sup>	150	Haulm	1	45.3	2.5	227	113	17.0
Total	4572						Total	343.8

<sup>&</sup>lt;sup>a</sup>Based on the statistics for 1997 (Anon, 1998); <sup>b</sup>from MAFF (1994); <sup>c</sup>early crop; <sup>d</sup>maincrop.

KCl extraction from soil cores taken in 30-cm increments (MAFF, 1986). Crop yields were measured following harvesting by hand or determined using a plot combine harvester. Sub-samples were taken to determine moisture and N content, with the latter measured by near infrared spectrophotometry.

### The effect of time and method of residue incorporation

This was investigated in an experiment on a free-draining loamy sand soil (Bridgenorth series, Badger Farm, Salop). A crop residue (cereal foliage cut and air-dried for storage: average composition 87% dry matter and 2.0% N) was spread on to the bare soil surface at a rate of 10 t ha<sup>-1</sup> fresh weight to separate, replicated plots on 7 June, 2 August, 2 September, 5 October, 7 November 1994 and 11 January 1995. Each time, the residue was incorporated immediately by ploughing to a maximum depth of 25–27 cm, and throughout the experiment period (until March 1996) the entire site was kept free of plants. The effect of incorporation method (plough, disc or left on the surface) was also investigated for a limited range of dates (August, September and October) in the same experiment.

### The effect of residue type

The effect of residue type was investigated on the same soil type (Bridgenorth series) at a different location (Weston, Staffs). The above-ground residues of a range of arable and horticultural crops (see Fig. 3.4.2) were incorporated either with their own below-ground residues, or with the below-ground residues left by a winter wheat crop (fertilized at 80 kg ha<sup>-1</sup> N). In addition, plots were also included where only the below-ground residues were incorporated for each of the harvested crops. Overall, the trial was set out as three fully replicated randomized blocks, with crop type and residue incorporation arranged on main and split plots, respectively. Although crops were harvested at different times (as appropriate), residue incorporation by ploughing for all treatments was carried out in a single operation on 12 September 1995. Subsequently, a winter barley crop was established and managed according to normal agronomic practice.

### **Results and Discussion**

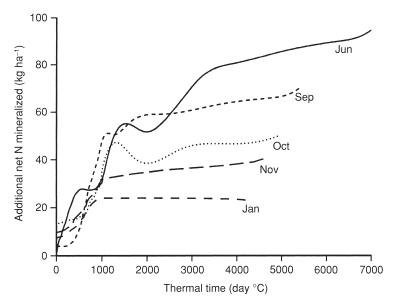
### Effect of incorporation time

Time of residue incorporation is important for nitrate pollution. Delaying cultivation decreased leaching only when a nitrogen-rich residue was incorporated; there were no differences in mineralization/leaching between different cultivation dates without residue incorporation (Table 3.4.2). The implication of this is that time of residue incorporation might also affect the fertilizer N requirement of the following crop. However, it was only extreme differences in timing (June to January) and large inputs of residue N (~180 kg ha<sup>-1</sup>) that showed measurable effects. In this experiment, rainfall was such that NO<sub>3</sub>-N was completely eluted from the soil profile; with less rainfall, a proportion of the mineralized N would have been available to the following crop, stressing the need to take account of winter rainfall in recommendation systems.

**Table 3.4.2.**  $NO_3$ -N (kg ha<sup>-1</sup>) leached during the winter following incorporation of a green residue. Standard error (n = 3) in parentheses.

	Residue incorporation date						
	June	August	September	October	November	January	
With residue	204 (6.1)	164 (25.7)	181 (24.1)	133 (7.0)	139 (3.3)	129 (2.3)	
No residue	135 (13.8)	147 (5.8)	133 (8.7)	114 (2.9)	139 (10.7)	125 (9.4)	

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**Fig. 3.4.1.** Calculated additional net N mineralized from residues incorporated on different dates. These data were smoothed with a least squares cubic technique (Steinier *et al.*, 1972) applied with a 15-point window and three passes using Fig-P software (Biosoft, Cambridge). 'Thermal time' is the accumulated day degrees (above 0°C).

The effect of residue incorporation on SMN was observed for up to 18 months. The residue incorporation date affected the total N release (Fig. 3.4.1), calculated as the change in the total of mineral forms of N (essentially SMN plus leached NO<sub>3</sub>-N) over the period of the experiment. Significant N loss by denitrification seems unlikely from the loamy sand soil; alternatively, mineralized N may have been re-immobilized rapidly. However, this result has important implications for N cycling simulation models; and also implications for fertilizer N recommendations which currently assume a fixed N supply irrespective of incorporation date.

### Method of incorporation

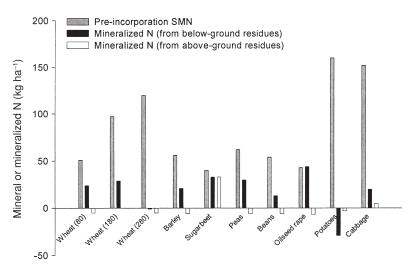
The method of incorporation had no differential effect on leaching (154 and 161 kg N ha<sup>-1</sup> for shallow cultivation and plough, respectively, as an average of three cultivation dates). The results from other workers indicate that the decomposition of added plant material is more rapid in sandy than in more clay-rich soils under undisturbed conditions, whereas the reverse is true under conventional cultivation (Silgram and Shepherd, 1999). However, since mineralization is greater following cultivation, this

would tend to cancel out any difference between cultivated and undisturbed treatments in a sandy soil (whilst exaggerating those in clay-rich soils). These results lend support to this conclusion. In practical terms, it is unlikely that the cultivation method has importance in influencing fertilizer N advice for sandy soils, since it is common practice to plough these soils.

### The effect of residue type

The SMN immediately before residue incorporation (0–90 cm) in autumn 1995, and the calculated contributions of above-ground and below-ground residues to mineralized N at harvest of the winter barley test crop (autumn 1996) are shown in Fig. 3.4.2. The main N contribution to the winter barley test crop (autumn 1995 to autumn 1996) from the crop residues came from a below-ground component. Much of this was already in SMN form when the barley was sown. It therefore derived from fertilizer and/or rapidly mineralized residues arising from root turnover or leaf drop during the growing season. The impact of fertilizer rate on SMN was demonstrated by the three wheat crops which received different N rates (Fig. 3.4.2), agreeing with other results (e.g. Chaney, 1990). With the exception of sugarbeet, the majority of mineralized N following incorporation was derived from below-ground residues. In general, above-ground residues made a negative contribution to the mineral N pool.

Although the general trend of increasing N supply with N index (MAFF, 1994) was apparent, there were significant differences between the



**Fig. 3.4.2.** Pre-incorporation soil mineral nitrogen and subsequent mineralization of above-ground and below-ground residues. Numbers in parentheses indicate fertilizer N applied (kg ha<sup>-1</sup>).

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source of this N. Nitrogen reserves for cereals (an N index 0 residue) were derived mainly from pre-incorporation SMN, whereas for sugarbeet (also an N index 0 residue) the contributions from pre-incorporation SMN, mineralized N from below-ground residues, and mineralized N from above-ground residues were approximately equal. Similarly, for N index 1 residues such as potatoes and cabbages, N reserves were derived mainly from pre-incorporation SMN, whereas those for oilseed rape were derived from pre-incorporation SMN and mineralized N from below-ground residues. The crops considered to be intermediate between N index 0 and N index 1 (peas and beans) were similar to oilseed rape, except that pre-incorporation SMN tended to be slightly higher and mineralized N from below-ground residues was lower.

The significance of these results is twofold. First (in general), the largest proportion of N from arable crop residues is vulnerable to leaching, as it is already in mineral form in the autumn. Secondly, (for most crops) further N is derived from mineralization of below-ground residues, implicating root turnover as the main mechanism for recycling plant N through the SOM; the effect of above-ground residues is small and generally immobilizes N. The exception is sugarbeet, for which above-ground residues are also important as a source of mineral N.

### **Conclusions**

Crop residue returns are an important N source to the following crop. The amount of N made available to the next crop depends on the amount of N returned and its 'mineralizability'. This is recognized in current advice. However, our experiments have shown where there is scope for improving recommendations and/or N management.

- For many crops, much of the returned N is already in mineral form in the autumn. Management options to reduce losses of nitrate are therefore limited: an early sown crop (or cover crop) to trap some of the N, or delaying cultivation.
- Fertilizer advice will be affected by residue incorporation date and winter rainfall because of effects on mineralization and leaching: current advice does not do this.
- Residue incorporation date appeared to affect the *total* N release. This
  may have been because for earlier incorporation dates much of the mineralized N was leached from the soil system, whereas for later incorporation, SMN retained in the soil was available for re-immobilization.
  The result is in contradiction to accepted wisdom and needs further
  investigation.
- Sugarbeet tops act as a 'nitrogen store' during winter. Consequently, this organic N is protected from leaching and will be made available to

the following crop if the tops are mineralized in the growing season. Other arable residues tend to mineralize in the late summer/autumn leaving a large pool of SMN. Thus, N supply from beet residues on sandy soils will be greater than from other residues where SMN can be leached. Fertilizer recommendations on sandy soils need to reflect this.

### **Acknowledgements**

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## Carbon and Nitrogen Losses after Ploughing out Grass and Grass-Clover Swards

3.5

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### Introduction

Growing clover can be an environmentally friendly way of increasing the nitrogen supply in soils because bacteria in the clover root system fix atmospheric nitrogen. Potentially, this biologically fixed nitrogen could reduce reliance on energy-intensive fertilizers. However, we need to be sure that this nitrogen is available to subsequent crops and does not contribute to pollution of air or water. The process of mineralization releases ammonium from nitrogen-rich crop residues and soil organic matter. In well-aerated soils, ammonium undergoes nitrification to nitrate, which contributes to the pollution of surface and ground water. Denitrification, which occurs under anaerobic soil conditions, converts nitrogen compounds to nitrous oxide and nitrogen gas. Nitrous oxide emitted from soil contributes to the greenhouse effect.

The nitrogen cycle in soil is inseparable from the carbon cycle. Storage of carbon within the soil system is beneficial to agriculture and the environment. Soil organic matter (SOM) provides the largest terrestrial sink of carbon and affects the atmospheric content of carbon dioxide, carbon monoxide, methane and other greenhouse gases. It is a source of crop nutrients and is one of the dominant factors influencing soil structure. Structure in turn affects many processes that occur in soil. Examples are the rate at which heat and material move through the soil, its suitability as a medium for root growth and its resistance to degradation. Therefore, any changes to SOM status will affect crop productivity and its sustainability, environmental pollution and soil quality.

From both an agricultural and environmental perspective, we need to know what farming practices are best for retaining nitrogen and carbon in the soil system. Ley-arable systems increase the organic nitrogen content of the soil through fixation and thus improve soil quality. However, ploughing out the swards releases CO<sub>2</sub> from the extra organic matter. This CO<sub>2</sub> makes a large extra contribution to the atmosphere. Furthermore, crops may not use the released nitrogen efficiently, compared with continuous arable systems (Lloyd, 1992; Djuurhuus and Olsen, 1997). There is some evidence to suggest that winter cereals may be less efficient for nitrogen retention than spring cereals, because of early autumn cultivation and higher fertilizer N use.

Deep tillage mixes organic matter over a greater depth of soil than conventional tillage. This leads to immobilization and fixation of nitrogen in mineral sub-soils but also to higher CO<sub>2</sub> emission rates. Shallower tillage reduces disturbance and fossil fuel use, and so may reduce CO<sub>2</sub> loss. Using published data from long-term experiments in the USA, the average soil carbon level was 285 g m<sup>-2</sup> higher under no-tillage than under conventional tillage (Paustian *et al.*, 1997). A reduction in tillage thus may allow significant sequestration of carbon in the soil.

We conducted a field experiment over 3 years to measure the agronomic and environmental effects of ploughing out grass and grass-clover swards. Our overall objective was to measure the effects of timing of cultivations, tillage methods and fertilizer use on crop growth and the storage of carbon and nitrogen in soil.

### Methods

We sited the experiment on grass and grass-clover swards that were  $\sim$ 14 years old and had been part of a sheep grazing experiment. It should be emphasized that both types of sward were managed to obtain similar gross margins, and productivity in terms of liveweight gain was  $\sim$ 20% lower on the grass-clover swards (Vipond *et al.*, 1997).

Average annual rainfall at the site is 870 mm. Mean monthly temperature varies between 2.5°C in January and 14.8°C in July.

There were four paddocks, two in grass and two in grass-clover. Table 3.5.1 summarizes the treatments. Within each paddock, there were four main plots, one for each of the rotations. These main plots were split into three sub-plots for the tillage methods, and the tillage sub-plots were split further for nitrogen fertilizer levels. The tillage methods were conventional ploughing to 200 mm depth and deep ploughing to 300 mm depth. We also included a no-tillage treatment to provide the widest range for comparison and to allow us to separate the effects of ploughing from sward destruction.

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<b>Table 3.5.1.</b> The t	reatments ir	n the field	experiment.
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Two swards:	Grass	Grass-clover	A1	
Three tillage methods:	Conventional ploughing to 200 mm	Deep ploughing to 300 mm	No-tillage	
	Р	D	Z	
Two crops	Spring barley	Winter barley		
Four nitrogen rates (kg ha <sup>-1</sup> )	Spring barley	Winter barley		
N0	0	0		
N1	40	60		
N2	80	120		
N3	120	180		
Four crop rotations	1996	1997	1998	
SSS	Spring barley	Spring barley	Spring barley	
SWW	Spring barley	Winter barley	Winter barley	
GWW	Grass/grass-clover	Winter barley	Winter barley	
GWS	Grass/grass-clover	Winter barley	Spring barley	

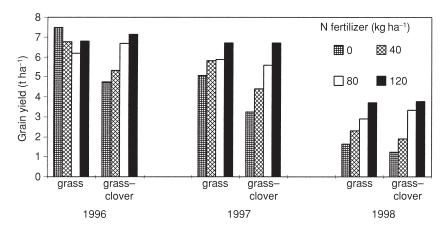
We measured gaseous losses of carbon and nitrogen and nitrate leaching, as well as crop uptake.

### Results

### Crop growth

In 1996, which was the first year after ploughing, spring barley grown after grass–clover responded strongly to nitrogen fertilizer, as shown in Fig. 3.5.1. However, there was no response to nitrogen after ploughing out from grass. These results indicate that the potentially mineralizable nitrogen after grass–clover is much lower than after grass, contrary to expectations. This surprising outcome may be a result of the lower fertilizer inputs to the grass–clover swards, so they may have had higher C: N ratios. We are awaiting results to clarify this. In an earlier (1992) study on this site, the macroorganic matter (mainly root material) in the 0–4 cm depth range prior to ploughing out had an N content of 1.27  $\pm$  0.04% and 1.44  $\pm$  0.10% for grass–clover and grass plots, respectively. When these swards were ploughed out, the N input from this material was 152.1  $\pm$  8.7 and 204.6  $\pm$  11.8 kg N ha $^{-1}$  for grass–clover and grass plots, respectively (Davies, 1996).

In the final year, all treatment combinations showed a response to nitrogen fertilizer, indicating that the residual effects of the original grass



**Fig. 3.5.1.** Spring barley yields in response to four fertilizer nitrogen rates in the first 3 years after ploughing out long-term grass and grass—clover swards. The results are averages of the three tillage methods.

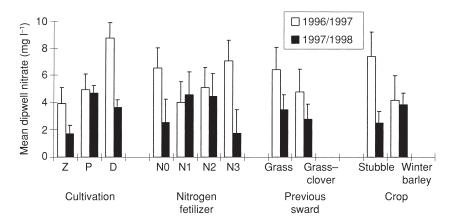
and grass—clover swards had disappeared. Overall, there was little difference between the two ploughing treatments, but the no-tillage treatment yielded less, especially at zero nitrogen. The effects of tillage and previous sward on yield were less for winter barley than for spring barley because of better and more uniform crop establishment and lower weed populations. Yields were low in the last year because a wet spring delayed sowing, and seedbed preparation compacted the soil.

### Nitrate leaching

The most reliable method of estimating nitrate leaching is to sample the tile drainage (Vinten *et al.*, 1994). However, the drainage works required made this approach impractical. Therefore, we estimated nitrate leaching on selected plots by sampling from dipwells. Such samples represent water that flows laterally to the artificial drainage system in the upper, most permeable, part of the soil profile. We collected samples weekly during winter months of 1996/97 and 1997/98. Rainfall from October to March inclusive was 596 and 514 mm in 1996/97 and 1997/98, respectively.

Figure 3.5.2 summarizes the main treatment effects for the two winters. There were significant effects of cultivation in both years, with no-tillage showing the smallest leaching losses. Nitrate concentration in dipwells was greatest from the deep cultivated plots in 1996/97. We also found significant fertilizer effects in both years. The largest leaching losses were from the N0 and N3 plots in 1996/97 but from the N1 and N2 plots in 1997/98. More nitrogen leached from the grass plots than from the

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**Fig. 3.5.2.** The effects of the main treatment on nitrate leaching, assessed from the nitrate concentration in dipwells.

grass—clover plots in 1996/97, the first winter after ploughing. Winter crop cover also affected nitrate leaching significantly in both years. More nitrate leached from the plots left as undisturbed spring barley stubble than from those planted with winter barley in 1996/97, but there was little difference between the crops in 1997/98.

We estimated the total amount of nitrate leached from each treatment in each year by multiplying mean nitrate concentration by the pipe drainage measured on a nearby experiment. Annual leaching loads were relatively low (6.4–19.6 kg N ha<sup>-1</sup>), compared with swards that were ploughed out in the late summer of 1993 (Davies, 1996).

### Gaseous losses of nitrogen and carbon

In the first year, nitrous oxide emissions shortly after drilling spring barley were high, reaching a peak of 510 g N ha<sup>-1</sup> day<sup>-1</sup>. Surface conditions were important for nitrous oxide emission. Emissions from the areas around drill slits in the no-tillage treatment were more than double those in the area between drill slits. This indicates the importance of macropore volume and continuity near the surface in regulating gas emissions.

In the following year, nitrous oxide emissions from no-tillage were even higher, reaching a peak of 2000 g N ha<sup>-1</sup> day<sup>-1</sup> after heavy rainfall. Soil measurements showed that much of this nitrous oxide was produced or accumulated near the soil surface. Nitrous oxide emissions on the other treatments were typically lower, giving peaks of ~200 g N ha<sup>-1</sup> day<sup>-1</sup> under conventional ploughing and ~120 g N ha<sup>-1</sup> day<sup>-1</sup> under deep ploughing. In the first two seasons, N<sub>2</sub>O fluxes showed a marked and rapid response to

rainfall. These were particularly high from no-tillage under spring barley in 1997 (Fig. 3.5.3). Existing models of soil nitrogen dynamics failed to simulate these large, rapid responses, indicating that we need more sophisticated models to enable us to predict the behaviour of such systems.

Effects of tillage on  $CO_2$  emissions were less consistent than for  $N_2O$ , and average rates were generally < 20 kg C ha<sup>-1</sup> day<sup>-1</sup>. High fluxes (up to 130 kg C ha<sup>-1</sup> day<sup>-1</sup>) shortly after tillage were related to ploughing depth. Emissions were low from no-tillage after heavy rain due to soil anaerobic conditions in the soil.

In 1997, tillage differences in CO<sub>2</sub> emissions (Fig. 3.5.4) were small, with typical emissions of between 5 and 25 kg C ha<sup>-1</sup> day<sup>-1</sup>. Emissions reduced almost to zero under no-tillage after a period of particularly heavy rainfall. These emissions related well to soil temperature at 200 mm depth. Under winter barley, weekly estimates of nitrous oxide emission were much lower and differed little between treatments, with typical values of 50 g N ha<sup>-1</sup> day<sup>-1</sup>. In the autumn, there was a pulse emission of 75 kg CO<sub>2</sub>-C ha<sup>-1</sup> day<sup>-1</sup> which lasted only 2 h after ploughing and was associated with soil disturbance releasing trapped CO<sub>2</sub>.

In 1998, we measured gas emissions intensively in the period immediately after cultivation. CO<sub>2</sub> emissions showed a strong flush in the

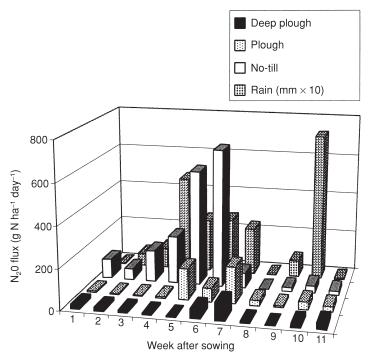


Fig. 3.5.3. N<sub>2</sub>O emissions from soil in the weeks after sowing spring barley in 1997.

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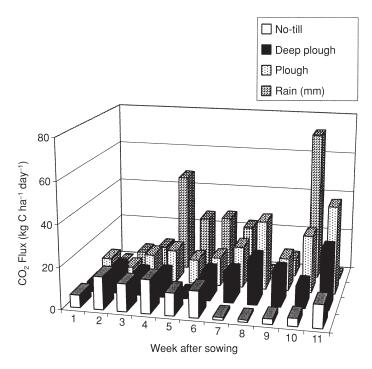


Fig. 3.5.4. CO<sub>2</sub> emission from soil in the weeks after sowing spring barley in 1997.

hour after ploughing for spring barley.  $N_2O$  emissions, monitored on the no-tillage treatment using automatic chambers, ranked with the fertilizer N levels. We also monitored  $N_2O$  emissions using manual chambers distributed throughout the experimental area. These showed marked spatial variability in emissions, which was associated with differences in hydrology resulting from both the treatments and the topography of the experimental site.

### Conclusions

The residual N value from long-term, low-input grass—clover swards to subsequent arable crops was lower than the residual N value from moderately fertilized grass swards. This was probably due to the lower N content of the macroorganic matter (roots, etc.) in the grass—clover swards before cultivation. In the first year after ploughing out of long-term grass swards, less nitrate leached from winter cereals than from spring cereal stubble. In subsequent years, there was no significant difference between spring- and winter-sown crops.

Reduced tillage reduced mineral N losses to drains, but, on this soil type, it also resulted in a larger requirement for fertilizer N after ploughing out of long-term grass swards. No-tillage and deep ploughing retained more carbon than conventional ploughing. However, there were large losses of nitrogen as nitrous oxide from no-tillage and, initially, significant losses of nitrate from deep ploughing. Thus, deeper ploughing than normal is likely to be a better option for C and N conservation than no-tillage.

Experimental responses to treatments were strongly influenced by climatic and topographical variability.

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### Fate and Behaviour of Organic Contaminants During Composting of Municipal Biowaste

3.6

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### Introduction

Degradation and transformation processes in the course of municipal biowaste composting affect both organic matter and organic contaminants present in the biowaste. During composting, a minute amount of contaminants will be degraded partially to metabolites or completely to CO<sub>2</sub> (Förstner and van Raaii, 1996).

A potential hazard to the environment results from contaminants, which form non-extractable residues (NERs) or are sorbed to dissolved organic matter (DOM), acting as a potential sorbent and carrier agent. Release of NERs may occur due to further transformation processes of humic substances in the compost matrix (DFG, 1998).

The aim of the project was to investigate the fate of three <sup>14</sup>C-radiolabelled organic contaminants (bis(2-ethylhexyl)(carboxyl-(<sup>14</sup>C)) phthalate (DEHP); pyrene-(4,5,9,10-<sup>14</sup>C) and, simazine-R-UL-<sup>14</sup>C] during composting over a period of 200 days to elucidate the hazard potential of compost application.

### Composting procedure

The composting process was simulated close to realistic conditions using high-tech simulation reactors. The stainless steel reactors had a volume of 1.8 m<sup>3</sup>, they were aerated from the bottom to the top and had internal as well as external temperature control. The segmented interior enabled

separate composting for each test substance. Municipal biowaste was used as starting material. The application of each of the test substances to the biowaste was carried out individually and took place prior to composting. The thermophilic phase was simulated for 29 days, reaching temperatures of ~70°C. The material was cooled to 30°C and the maturation phase was simulated for another 49 days. The long-term fate of the test substances was monitored for 6 months at temperatures of 30°C and a water content of 40%. Samples were taken after the thermophilic phase, after the maturation phase and after 120 and 200 days during the long-term study.

### **Analytical procedure**

The characterization of the test substances involved both analysing the compost matrix and examination of the reactor exhaust gas.

A sequential fractionation was carried out with the compost matrix, differentiating the <sup>14</sup>C-radioactivity between extractable fractions and non-extractable residues. The extraction was performed by shaking with water and acetonitrile, by Soxhlet with toluene, and by shaking with sodium hydroxide. The <sup>14</sup>C-radioactivity in the extracts was determined by liquid scintillation counting (LSC; Packard). The identity of <sup>14</sup>C-radioactivity in the extracts was analysed via <sup>14</sup>C-HPLC (RP18 hypersil with gradient elution acetonitrile/water). NERs were transformed into <sup>14</sup>CO<sub>2</sub> by incineration using a Packard oxidizer and the radioactivity was determined by LSC.

For quantification of the mineralization rate, the exhaust air was passed through PUF plugs to trap organic volatiles and subsequently passed through NaOH traps where  $CO_2$  was absorbed. The  $^{14}C$ -radioactivity trapped in the sodium hydroxide was determined daily by LSC.

### **Results and Discussion**

### Mineralization

Degradation of [<sup>14</sup>C]DEHP to <sup>14</sup>CO<sub>2</sub> occurred rapidly during the thermophilic phase. After 200 days, 70% of the initial [<sup>14</sup>C]DEHP was mineralized (Fig. 3.6.1). The degradation of [<sup>14</sup>C]simazine to <sup>14</sup>CO<sub>2</sub> was minor, leading to a mineralization rate of only 8% altogether. The degradation of pyrene was intermediate between these values. After a low mineralization rate during the thermophilic phase, the degradation to <sup>14</sup>CO<sub>2</sub> increased rapidly and reached 56% of the initial [<sup>14</sup>C]pyrene after 6 months. This delayed mineralization was also observed by Martens (1982), who found that in fresh composts only minor amounts of four-ring polycyclic aromatic hydrocarbons (PAHs) could be degraded. However,

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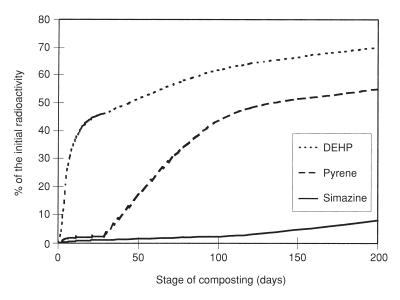


Fig. 3.6.1. Mineralization of DEHP, pyrene and simazine measured as <sup>14</sup>CO<sub>2</sub> in the reactor exhaust air.

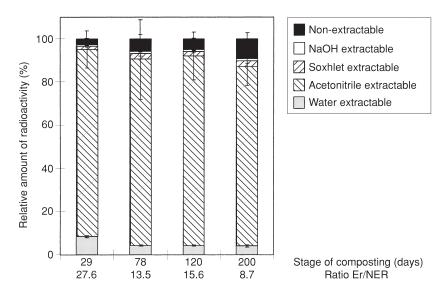
microbial populations of ripe composts possessed considerable capabilities to mineralize PAHs.

Other organic volatiles were observed to < 0.1% of initial  $^{14}$ C-radioactivity only.

### Distribution in the compost matrix

### [14C]DEHP

Figure 3.6.2 shows the distribution of <sup>14</sup>C-radioactivity in the compost matrix at different stages of composting. The main portion of radioactivity was solvent extractable and identified by HPLC analysis as [<sup>14</sup>C]DEHP. The amount of water-extractable radioactivity was too small for the HPLC analysis. Most probably metabolites were present in the aqueous extracts, as was found by Weber (1989). The remaining NERs showed a slight increase with time. Considering the loss by mineralization and applying the data to the initial content of <sup>14</sup>C-radioactivity, the following values were calculated: the water-extractable radioactivity did not exceed 4% of the applied radioactivity at any time during the experiment. The NER fraction amounted to 1.5% of applied radioactivity after the thermophilic phase and increased to 1.7% after 200 days of composting.



**Fig. 3.6.2.** Relative distribution of [¹⁴C]DEHP-radioactivity amongst different extracts of compost material during various stages of composting. Er = extractable radioactivity; NER = non-extractable residues.

The data of the [<sup>14</sup>C]DEHP analysis showed strong estimated deviations demonstrated with error bars. This is typical for [<sup>14</sup>C]DEHP analysis and was mentioned by Förstner and van Raaji (1996) who performed similar composting experiments. The deviations are not attributed to analytical errors, but are probably due to a heterogeneous distribution of DEHP in the sample material.

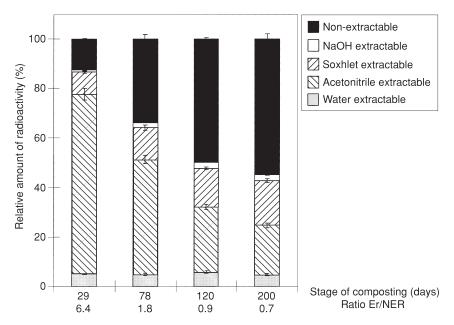
The ratio of extractable radioactivity (Er) and NERs decreased with time, making a release of NERs very unlikely. However, ratios were calculated using mean values. Considering the strong deviations, no clear prediction about the potential release of NERs can be made. In view of the low percentage of NER, only small amounts of applied [14C]DEHP could be released.

### [14C]Pyrene

As shown in Fig. 3.6.3, the water-extractable fraction of applied <sup>14</sup>C-radio-activity was small and an identification by HPLC analysis was not possible. However, the presence of polar metabolites is very likely (Hartlieb and Kördel, 2000). In relation to the initial content of <sup>14</sup>C-radioactivity, the water-extractable radioactivity did not exceed 5% during the experiment.

Solvent-extractable <sup>14</sup>C-radioactivity was identified by means of HPLC analysis as pyrene. During the thermophilic phase, ~12% of the initial

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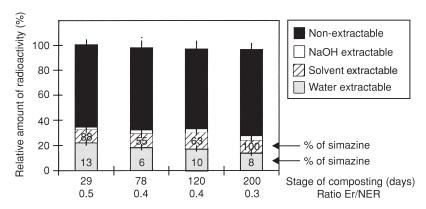


**Fig. 3.6.3.** Relative distribution of [1<sup>4</sup>C]pyrene-radioactivity amongst different extracts of compost material during various stages of composting. Er = extractable radioactivity; NER = non-extractable residues.

<sup>14</sup>C-radioactivity was bound to the compost matrix and could not be extracted with the described methods. During this period, the mineralization rate was negligible. With an increasing mineralization rate of pyrene after the thermophilic phase, the fraction of NERs grew rapidly, finally reaching 25% of the initial <sup>14</sup>C-radioactivity. The results indicate that there was a correlation between mineralization and formation of NERs. This corresponds with the results of Eschenbach (1995), who found no further formation of NERs in sterile soil samples. It can therefore be concluded that the formation of NERs, at least to some extent, can be attributed to microbial-mediated processes.

Moreover, the ratio of acetonitrile-extractable to toluene-extractable pyrene showed a decrease with time (Fig. 3.6.3). This indicates that apart from microbial processes there was a sorption of pyrene due to adsorption and ageing processes. Accordingly, the formation of NERs cannot be explained by one mechanism alone.

Release of NERs is very unlikely, which is demonstrated by the strong decrease in the Er: NER ratio and the small deviations of the data. This corresponds with investigations of Eschenbach (1995), who could not find any remobilization of PAHs from residual waste soil.



**Fig. 3.6.4.** Relative distribution of [14C]simazine-radioactivity amongst different extracts of compost material during various stages of composting. Er = extractable radioactivity; NER = non-extractable residues.

### [14C]Simazine

The HPLC analysis showed that the <sup>14</sup>C-radioactivity in the aqueous extracts and organic extracts consisted of metabolites as well as simazine (Fig. 3.6.4). Already after the thermophilic phase, NERs comprised up to 64% of the initial <sup>14</sup>C-radioactivity, and this increased slightly with time. Since metabolites were present in the extracts, it can be assumed that NERs preferably consisted of metabolites and only to a lower extent of the parent compound, simazine.

Figure 3.6.4 shows a very slight decrease in the ratio value. However, considering the deviations of the data, release of NERs cannot be ruled out. However, if there was a release of <sup>14</sup>C-radioactivity, it is questionable as to whether it would consist of simazine or its degradation products.

### Conclusions

The investigated test substances had a very different fate during composting and the following long-term study. The formation of NERs could not be explained by one mechanism alone. The hazard potential of compost application due to release of NERs under the experimental conditions has to be discussed:

1. DEHP: NERs were formed to a small extent only. Considering the large deviations of the data, no clear prediction can be made about release of NERs. However, a potential release should be expected for small amounts only.

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2. Simazine: during the thermophilic phase, > 60% of the initial <sup>14</sup>C-radioactivity was bound to the compost matrix and could not be extracted with the described methods. The formation of NERs can be due to both physical entrapment in the matrix and covalent binding of metabolites. Release of NERs cannot be ruled out, but is most likely to consist of metabolites rather than simazine, especially with increasing compost maturity.

**3.** Pyrene: the amount of NERs increased rapidly during the maturation phase. The formation of NERs can be attributed both to microbial activity and to adsorption and ageing mechanisms. Release of NERs during the experimental period is very unlikely.

### **Acknowledgements**

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## The Impact of Changing Farming Practices on Soil Organic Matter and Soil Structural Stability of Fen Silt Soils

3.7

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### Introduction

Increases or additions of soil organic matter can improve soil properties in many ways. Better plant nutrition (N, P, S and micronutrients), ease of cultivation, penetration and seedbed preparation, greater aggregate stability, reduced bulk density and improved water-holding capacity have all been observed (Johnston, 1986). This implies that a decrease in organic matter, e.g. by oxidation following cultivation, will adversely affect most, if not all of these properties, even if, in the long term, organic matter reaches an equilibrium with additions of organic matter in crop residues (Arrouays and Pelisser, 1994).

Evidence from the Representative Soil Sampling Scheme (1969–1985) suggests that there has been a reduction in the proportion of arable soils with 'high' (> 8%) and 'low' (< 1.8%) organic matter levels in England and Wales (Skinner and Todd, 1998), whilst average organic matter levels for all crops and grass have remained static during the same period.

In the 1960s, 14 sites were selected on the fertile silt-lands of the Holland district of Lincolnshire to assess changes in soil organic matter levels, because of concerns that declining soil organic matter levels were compromising the sustainability of agricultural production on these soils. Results from 1964 to 1989 were reported by Johnson and Prince (1991). This chapter presents results from the last sampling in 1996.

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### Methods

The sites were located on typical silt-land soils of the Romney and Blacktoft Association, with one Downholland Association site (Hodge *et al.*, 1984). The sites had been ploughed out of pasture land pre-1960 (higher organic matter status) or were long-term arable land (lower organic matter status). Crop rotations following sampling were either arable with vegetables, or ley-arable rotations.

At each site, two points were selected where soil samples consisting of ten cores were taken to 15 cm depth, within a 5 m radius of each of these points. Samples were analysed for soil organic matter content (MAFF, 1986). In the 1960s, measurements had been made on an annual basis, but less frequently thereafter until 1989 (Johnson and Prince, 1991). The soil sampling strategy was repeated in spring 1996 at 13 sites, labelled A–N (access to the Downholland Series site (D) was not granted). The soils were again analysed for organic matter content but, in addition, particle size distribution (MAFF, 1986) and soil structural stability measurements were made. Structural stability was assessed using a dispersion ratio technique adapted from the standard method outlined in MAFF Reference Book 441 (MAFF, 1985), where the stability of 'natural' soil aggregates (size range 5–30 mm) was measured, rather than the < 2 mm ground fraction.

Regression analysis using Genstat 5 (Payne et al., 1987) was carried out on the soil organic matter data over the ~30-year measurement period at each of the 13 fen silt sites, to assess if soil organic matter levels had increased, decreased or stayed the same. The models applied were simple linear and four-parameter logistic functions. It is appreciated that many of the logistic fits approach a simple exponential function, however, use of the exponential function on the data imposed an excessive bias on early sampling points.

### Results

Results of the soil organic matter and structural stability measurements, and estimates of 'stabilized' organic matter levels inferred from the regression analyses are summarized in Table 3.7.1.

At eight sites (C, E, F, I, J, K, M and N), there were no changes over time in soil organic matter levels (P > 0.05). Soil organic matter decline was evident at five sites, with the lower asymptote having minimum values between 2.5 and 3.4%. This minimum value was taken to represent the 'stabilized' soil organic matter content.

'Fitted' regressions of soil organic matter change over time for ex-pasture sites (Fig. 3.7.1) and arable sites (Fig. 3.7.2).

	Cropping	Cropping	Initial	1996	OM	'Stabilized'	Dispersion
Site	1961–1996	1996	OM (%)	OM (%)	change (%)	OM (%)	ratio (SEM)
Α	Arable	Calabrese	6.4ª	3.5	-2.9***	2.5	8.6 (0.24)
В	Arable	Cauliflower	5.3 <sup>a</sup>	3.0	-2.3***	3.3	6.0 (0.44)
С	Arable	Winter wheat	2.2a	2.3	+0.1	2.2	7.8 (0.51)
Е	Ley-arable	Ley	2.4	2.7	+0.3	2.6	8.2 (0.12)
F	Ley-arable	Ley	3.1	3.1	0	3.3	5.8 (0.11)
G	Arable	Cauliflower	3.0	2.5	-0.5***	2.6	8.0 (0.60)
Н	Arable	Cauliflower	5.2	3.6	-1.7**	3.3	7.7 (0.17)
I	Ley-arable	Peas	3.4	3.1	-0.3	3.1	8.0 (0.57)
J	Ley-arable	Peas	3.5	3.1	-0.4	3.2	7.0 (0.20)
K	Arable	Winter wheat	2.6	2.5	-0.1	2.8	8.7 (0.32)
L	Arable	Winter wheat	4.4	3.9	-0.5***	3.4	7.1 (0.33)
M	Ley-arable	Ley	2.6	2.7	+0.1	2.8	6.9 (0.34)
N	Ley-arable	Ley	4.3	4.0	-0.3	3.9	3.1 (0.43)

**Table 3.7.1.** Measured soil organic matter content and dispersion ratios, and estimated 'stabilized' organic matter contents.

Sites A and B were in permanent pasture until the early 1960s (ploughed out in 1963 and 1962, respectively) and showed an exponential decline in soil organic matter content. The initial rate of decline was similar for both sites. However, both sites appear to be at or approaching 'stabilized' soil organic matter levels of ~2.5 and ~3.3%, respectively. The topsoil at site B was more stable than at site A (dispersion ratios of 6.0 and 8.6, respectively), corresponding to the higher organic matter levels at site B.

The two initially higher organic matter sites (H and L) showed a decline in soil organic matter levels over time (P < 0.05). The rapid initial decline at site H suggests that this site may have been ploughed out of pasture prior to sampling, although this cannot be confirmed with the available historical land management data. The 'stabilized' organic matter levels were estimated at 3.3 and 3.4% for sites H and L, respectively, with the sites having similar structural stabilities. Site G showed a slow decrease in soil organic matter content (P < 0.05) and sites C and K no change (P > 0.05). It is interesting to note the 'apparent' decreases in the early 1980s at sites G and L which may be due to deeper ploughing, diluting topsoil organic matter levels.

The ley-arable sites showed no changes (P > 0.05) in soil organic matter content over time. 'Stabilized' organic matter values were estimated

<sup>&</sup>lt;sup>a</sup>Measured in 1964, all other initial figures measured in 1969. SEM = standard error of the mean.

<sup>\*\*</sup>P < 0.01; \*\*\*P < 0.001

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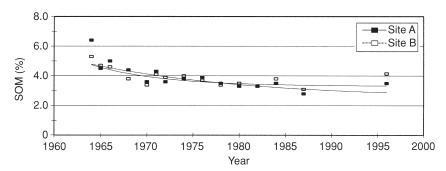
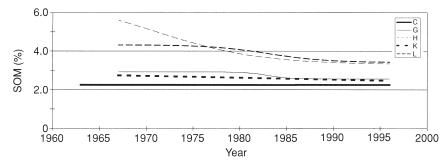


Fig. 3.7.1. Changes in soil organic matter content over time on ex-pasture sites, including fitted regressions.



**Fig. 3.7.2.** Changes in soil organic matter content over time on anable sites (fitted regressions only for clarity).

to range from 2.6 to 3.9% (measured organic matter levels ranged between 2.7 and 4% in 1996).

Regression analysis was used to establish whether there was a relationship between the 'stabilized' and total soil organic matter contents measured in 1996 and soil structural stability. There was a weak positive relationship between total soil organic matter contents and structural stability (P = 0.05, adjusted  $r^2(adj) = 20\%$ ; y = 0.39 + 7.3/(1 + EXP(4.4(x-4.1)))). Surprisingly, there appeared to be a much stronger relationship between 'stabilized' soil organic matter levels and soil structural stability (P = 0.002, adjusted  $r^2(adj) = 74\%$ ; y = -3.1 + 11.4/(1 + EXP(2.9(x-4.0)))). However, this relationship was very dependent on the 'stabilized' organic matter estimates for sites A, B and L.

There was no relationship (P > 0.05) between soil structural stability and the clay, silt (fine and coarse) and sand (fine and medium) content of the soils. Also, the strength of the relationships between soil structural stability and total/'stabilized' organic matter levels was not improved by the inclusion of sand, silt and clay content data.

# **Discussion and Conclusions**

Many authors (Johnson, 1991; Jenkinson *et al.*, 1994) have reported rapid changes in soil organic matter contents after major land use changes (e.g. ploughing out of pasture). The effects of ploughing out grassland on soil organic matter levels were particularly evident at sites A and B, with a rapid decline over the first 5 years. Sites H and L, where the cropping history was uncertain, exhibited a similar decline from relatively high initial organic matter levels and may well have been in pasture before the study commenced.

The estimated 'stabilized' soil organic matter levels were well related (P < 0.01) to the soil structural stability measurements. However, there was only a weak (P = 0.05) relationship between total soil organic matter levels and structural stability. Results from this work are in agreement with the findings of other workers (Wander and Traina, 1996) which suggested that there was a rapidly decomposable organic matter fraction, which does not contribute to soil stability. However, as previous work has suggested, other soil properties can affect structural stability, such as clay content (Sorenson, 1981), soil water regimes and microbial activity (Scott *et al.*, 1996).

At three of the long-term arable sites, soil organic matter levels declined by 0.5–1.7% over the monitoring period. At two of these arable sites, the decreases may have been related to deeper soil cultivations, as the 'apparent' changes occurred over a short period of time with steady-state organic matter levels before and after. At the six ley–arable sites and two of the arable sites, there were no changes in soil organic matter levels over the monitoring period.

The data presented here suggest that all sites will reach stable or very slowly changing organic matter levels under continuous arable cropping regimes. It is concluded from the study that modern farming systems are not causing a decline in the organic matter content of fen silt soils.

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# Crop Management Systems to Conserve Soil Fertility After Long-term Setaside in Southern Italy

3.8

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# Introduction

Setting aside land from agricultural production for a long period seems to reduce the adverse effects of intensive agricultural systems by conserving biodiversity, reducing soil erosion and restoring soil quality (e.g. increasing soil organic matter content and metabolic activities). To an extent, the degree to which this is true is determined largely by agricultural system management, soil type, initial level of soil organic matter content and climate (Fullen, 1998; Masciandaro et al., 1998). When brought back to cultivation, soil organic matter content usually decreases (Huggins et al., 1998; Reeder et al., 1998). In Southern Italy, where climatic conditions favour soil organic matter mineralization, continuous cropping systems based upon deep ploughing have brought about a dramatic drop in average soil organic matter content, whose preservation is crucial in such conditions in order to sustain future agricultural productivity and environmental quality. However, soil quality preservation should be matched with acceptable crop yield and favourable energetic and economic budgets; therefore, there is a need to devise sustainable agricultural management systems which would encompass all these objectives. To evaluate the optimum management options for agricultural land that has lain unproductive for a long period in accordance with EU policy (reg. 1765/92 and subsequent modifications that also provide for leaving land uncultivated for periods of 20 years) in Southern Italy, two crop management systems and three crop

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rotations were combined factorially and studied over a 4-year period on land left uncultivated for 25 years.

# Materials and methods

Research was carried out at the University of Reggio Calabria, in Southern Italy, at a hilly site (6–8% slope, 250 m a.s.l., 485 mm mean annual rainfall, of which 86% occurs between October and May, 16 and 23°C average minimum and maximum temperatures). The main soil physical and chemical characteristics as measured at the beginning of the experiment are shown in Table 3.8.1. A field that had been abandoned for 25 years, where a mixture of native prairie species was the dominant vegetation, was brought back to cultivation according to two different crop management systems: a conventional system (CS), based on the standard agricultural practices in the study area (i.e. ploughing to a 30-35 cm depth, fertilization to reintegrate total crop nutrient uptake, and chemical weed control) and a low-input system (LIS), based on sustainable agricultural practices (i.e. no-tillage, fertilization to reintegrate only nutrients taken up by crop-marketable product, and mechanical weed control, except for glyphosate spraying prior to sowing). Crop residues were removed in CS and left on the soil surface in LIS. Three crop rotations, specifically durum wheat (Triticum durum Desf.) continuous cropping (typical of the study area) (Rotation 1), a horse bean (Vicia faba L. var. equina Persoon)-durum wheat 2-year rotation (Rotation 2) and a

**Table 3.8.1.** Soil characteristics measured at the beginning of the experiment (1993).

		Depth	n (cm)
Characteristic	Analytical method	0–7	7–30
pH (H <sub>2</sub> O)	Mc Lean, 1982	7.2	7.2
Clay (%)	USDA (Gee and Bauder, 1982)	41.0	44.2
Silt (%)	USDA (Gee and Bauder, 1982)	22.6	19.8
Sand (%)	USDA (Gee and Bauder, 1982)	36.4	36.0
Organic matter (%)	Walkley-Black (Nelson and Sommers, 1982)	2.8	2.0
Total nitrogen (%)	Kjeldahl (Bremmer and Mulvaney, 1982)	1.1	0.9
$P_2O_5$ (mg kg <sup>-1</sup> )	Olsen (Olsen and Sommers, 1982)	12.3	11.6
$K_2O$ (mg kg $^{-1}$ )	Knudsen et al., 1982	373	216
Aggregate stability (%)	Malquori (Lotti and Galoppini, 1980)	57.7	56.0
Field capacity (0.3 bar) <sup>a</sup>	Richard chamber	28.5	30.1
Wilting point (15 bar) <sup>a</sup>	Richard chamber	17.8	18.5

<sup>&</sup>lt;sup>a</sup>As a percentage of dry weight.

subterranean clover (*Trifolium subterraneum* L.)—durum wheat 2-year rotation (with the legume interseeded in wheat) (Rotation 3), were compared for each crop management system. Each phase of the rotation was present every year. Berseem clover (*Trifolium alexandrinum* L.) replaced subterranean clover in CS from the second rotation cycle onwards. Both legumes were used in CS as a forage crop, whilst in LIS, subterranean clover was used as a cover crop. Manure was applied in neither CS nor LIS according to mainstream Italian agricultural systems, which are often characterized by complete separation of fodder production and animal husbandry. The experimental design was a split plot with three replicates, with crop management systems in the main plots and crop rotations in the sub-plots. Sub-plot size was  $360 \text{ m}^2$  ( $9 \times 40 \text{ m}$ ). Soil was sampled before tillage in the

Table 3.8.2. ANOVA results.

		Sc	Soil organic matter		Total nitrogen		Assimilable $P_2O_5$			Aggregate stability			
Soil depth (c	cm)	0–7	7–30	0–30	0–7	0-7 7-30 0-30		0–7	7–30	0–30	0-7 7-30 0-30		0–30
1993													
System	mean effect	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Rotation	mean effect	ns	*	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
$\text{Sys.} \times \text{rot.}$	interaction	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
1995													
System	mean effect	*	ns	ns	_	_	_	_	_	_	ns	ns	ns
Rotation	mean effect	ns	ns	ns	_	_	-	_	_	-	*	ns	*
$\text{Sys.} \times \text{rot.}$	interaction	ns	ns	ns	-	-	-	-	-	-	ns	ns	ns
1997													
System	mean effect	**	ns	**	**	*	**	*	ns	ns	ns	ns	ns
Rotation	mean effect	ns	ns	ns	ns	ns	ns	ns	ns	ns	*	*	*
$\text{Sys.} \times \text{rot.}$	interaction	ns	ns	ns	*	ns	ns	ns	ns	ns	ns	ns	ns
Year	mean effect	*	*	ns	*	*	*	*	*	*	ns	ns	ns
Systema	mean effect	*	ns	*	*	ns	ns	ns	ns	ns	ns	ns	ns
Rotationa	mean effect	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	*
$\text{Sys.} \times \text{rot.}^a$	interaction	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Year $\times$ sys.	interaction	*	*	*	*	*	*	*	ns	ns	ns	ns	ns
$Year \times rot.$	interaction	ns	*	ns	ns	ns	ns	ns	ns	ns	*	ns	ns
$\text{Year} \times \text{sys.}$	$\times$ rot.	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

<sup>&</sup>lt;sup>a</sup>Analysis for the 3 years.

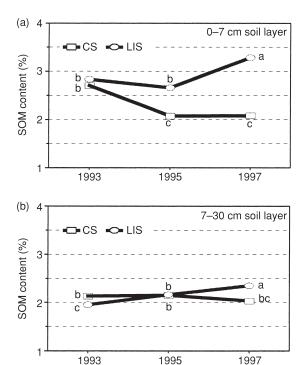
ns, Not significant; \*significant at 5% level; \*\* significant at 1% level.

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summers of 1993 (at the onset of the experiment), 1995 and 1997 (at the end of the first and second rotation cycles, respectively). Thanks to a low soil heterogeneity, four soil sub-samples were taken in each plot down to a 30 cm depth (0–7 and 7–30 cm layers) by means of a 2-cm diameter probe, and then pooled to obtain a bulk sample. Total soil organic matter content (Walkley-Black method) and aggregate stability (Malquori method) were measured at each sampling date, whilst total N (Kjeldahl method) and assimilable  $P_2O_5$  (Olsen method) were measured in 1993 and 1997. To reduce variance heterogeneity, soil organic matter and total N percentage data were transformed to  $\sqrt{(x + 0.5)}$  while  $P_2O_5$  data were transformed to arcsin  $\sqrt{(x)}$  prior to statistical analysis. Data were then subjected to analysis of variance according to a split plot experimental design. The analysis of variance was carried out separately for each year and, after carrying out Bartlett's test, a combined analysis over years was performed using the time factor as a random variable. ANOVA results are shown in Table 3.8.2.

## Results

Soil organic matter was affected significantly and differently by the management systems during the experimental period as shown in Table



**Fig. 3.8.1.** Soil organic matter: 'year × management system' interaction.

3.8.2 (year × system interaction). Soil organic matter content in the upper soil layer (Fig. 3.8.1a) decreased sharply (by 24%) in CS during the first 2 years and then stabilized in the subsequent 2 years, whilst there was no initial decrease in LIS and a subsequent significant increase (23%) in 1997. No significant differences in the 7–30 cm layer were observed under CS over the 4-year period; in contrast, soil organic matter progressively increased in LIS over the trial period (from 1.95 to 2.35%) (Fig. 3.8.1b). Values of soil organic matter content in the combined soil layer (0–30 cm) were mainly a result of those found in the upper layer. Soil organic matter content was observed to be significantly higher in LIS than in CS in all layers after 4 years when averaged over crop rotations (Table 3.8.3).

Probably due to high initial values, aggregate stability was not affected by crop management systems to the same extent as soil organic matter, but was more affected by crop rotation (Table 3.8.2). Rotation 3, which includes clover, improved aggregate stability in 1995 and 1997 (Table 3.8.4). It is worth observing the interaction 'year × rotation' illustrated in Fig. 3.8.2, showing an aggregate stability increase in the 0–7 cm layer over time in the rotation which included clover.

**Table 3.8.3.** Crop management system and rotation effects on soil organic matter.

	Soil organic matter (%)									
		1993				1995			1997	
Soil depth (cm)	0–7	7–30	0–30		0–7	7–30	0–30	0–7	7–30	0–30
Mean effects										
CS	2.71	2.13	2.27		$2.07^{b}$	2.15	2.16	2.08 <sup>b</sup>	2.03 <sup>b</sup>	2.09 <sup>b</sup>
LIS	2.83	1.95	2.18		2.66a	2.16	2.31	3.28a	2.35 <sup>a</sup>	2.58a
Rot. 1	2.70	2.25 <sup>a</sup>	2.38		2.37	2.22	2.31	2.58	2.13	2.27 <sup>b</sup>
Rot. 2	2.80	1.90 <sup>b</sup>	2.09		2.38	2.13	2.21	2.67	2.32	2.43 <sup>a</sup>
Rot. 3	2.82	1.97 <sup>b</sup>	2.20		2.34	2.12	2.18	2.79	2.12	2.29
Interactions										
$CS \times Rot. 1$	2.65	2.41	2.46		2.08	2.18	2.16	2.08	1.99	2.01
$CS \times Rot. 2$	2.86	2.02	2.22		2.13	2.20	2.18	2.11	2.06	2.07
$CS \times Rot. 3$	2.62	1.97	2.12		2.01	2.07	2.06	2.05	2.05	2.05
LIS $\times$ Rot. 1	2.75	2.10	2.25		2.67	2.26	2.35	3.08	2.28	2.47
LIS $\times$ Rot. 2	2.74	1.79	2.01		2.64	2.06	2.20	3.23	2.58	2.73
LIS $\times$ Rot. 3	3.03	1.97	2.21		2.67	2.17	2.28	3.53	2.19	2.50
Year	2.77	2.04	2.23		2.37	2.16	2.24	2.68	2.19	2.33

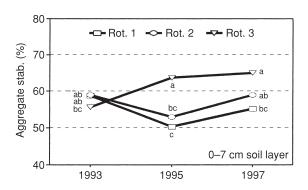
Values followed by the same letter in the same column are not statistically different at the 0.05 probability level.

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Table 3.8.4.	Crop management system and rotation effects on aggregate stability.

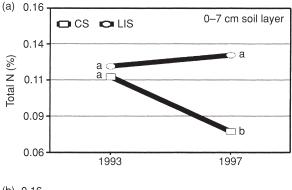
	Aggregate stability (%)								
		1993			1995			1997	
Soil depth (cm)	0–7	7–30	0–30	0–7	7–30	0–30	0–7	7–30	0–30
Mean effects									
CS	58.0	56.5	56.8	54.6	52.9	53.7	60.7	55.9	57.0
LIS	57.4	55.6	56.0	54.7	54.5	54.5	58.6	56.8	57.2
Rot. 1	58.7	52.3	53.8	50.3 <sup>b</sup>	50.2	50.2 <sup>b</sup>	55.1 <sup>b</sup>	53.7 <sup>b</sup>	54.0 <sup>b</sup>
Rot. 2	58.9	57.9	58.1	52.8	54.6	54.1	58.9 <sup>ab</sup>	57.0 <sup>ab</sup>	57.4 <sup>b</sup>
Rot. 3	55.6	57.8	57.3	63.6a	56.4	58.1 <sup>a</sup>	65.0 <sup>a</sup>	58.4a	60.0a
Interactions									
$CS \times Rot. 1$	60.0	53.2	54.8	49.6	47.9	48.3	56.9	54.6	55.1
CS × Rot. 2	58.8	59.0	58.9	55.0	55.8	55.6	60.1	57.5	58.1
$CS \times Rot. 3$	55.2	57.2	56.7	64.6	55.1	57.3	65.3	55.7	57.9
LIS $\times$ Rot. 1	57.4	51.5	52.9	50.9	52.5	52.1	53.4	52.8	52.9
LIS $\times$ Rot. 2	59.0	56.8	57.3	50.7	53.3	52.7	57.6	56.5	56.8
LIS $\times$ Rot. 3	55.9	58.4	57.8	62.7	57.6	58.8	64.7	61.2	62.0
Year	57.7	56.0	56.4	54.6	53.7	54.1	59.7	56.4	57.1

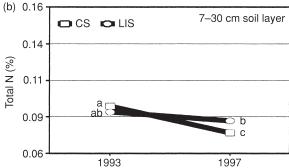
Values followed by the same letter in the same column are not statistically different at the 0.05 probability level.



**Fig. 3.8.2.** Aggregate stability: 'year × rotation' interaction.

Adoption of LIS preserved soil total N content both in the upper (1.19%) in 1993 and 1.27% in 1997) and lower layers (0.96) and 0.93%, respectively) (Fig. 3.8.3a and 3.8.3b) and, consequently, in the combined layer. In contrast, a significant decrease was observed in both layers under CS (from 1.12 to 0.74% in the 0–7 cm layer and from 0.92 to 0.74% in the 7–30 cm layer). The effect of crop management systems on assimilable  $P_2O_5$  was significant only in the upper soil layer in 1997 (as mean effect)





**Fig. 3.8.3.** Total nitrogen: 'year × management system' interaction.

and as an interaction 'year  $\times$  system' (Table 3.8.5). In fact, a sharper increase was observed in LIS (155%) than in CS (86%) after 4 years. No appreciable effect of crop rotation was observed on either total N or assimilable  $P_2O_5$  content.

# Discussion

Overall, the effect of crop management systems on soil fertility parameters prevailed over that of crop rotation and was observed mainly in the upper soil layer (Table 3.8.2). Use of no-tillage in LIS very probably helped in preserving both soil organic matter and major nutrient content (N and P<sub>2</sub>O<sub>5</sub>), as has been observed elsewhere (Dalal *et al.*, 1995; Reeder *et al.*, 1998). Besides reduced soil disruption, variations in these parameters are also related to the amount of crop residues produced (Campbell *et al.*, 1996) and the use of fertilizers (e.g. phosphorus). In this respect, the observed soil organic matter increase (compared with the initial level) under LIS in the combined layer (0–30 cm) probably depended upon the higher quantity of crop residues left on the soil surface, which out-yielded setaside natural productivity in the study area (14.5 and

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**Table 3.8.5.** Crop management system and rotation effects on total nitrogen and assimilable  $P_2O_5$  content in the soil. Values followed by the same letter in the same column are not statistically different at the 0.05 probability level.

	Total nitrogen (%)					Assimilable P <sub>2</sub> O <sub>5</sub> (p.p.m.)						
		1993			1997			1993			1997	
Soil depth (cm)	0–7	7–30	0–30	0–7	7–30	0–30	0–7	7–30	0–30	0–7	7–30	0–30
Mean effects												
CS	1.12	0.92	0.97	0.74 <sup>b</sup>	0.74 <sup>b</sup>	$0.74^{b}$	13.0	11.6	11.8	24.2 <sup>b</sup>	16.1	15.1
LIS	1.19	0.88	0.96	1.27 <sup>a</sup>	0.82a	$0.93^{a}$	11.6	11.5	11.6	29.6a	11.7	15.9
Rot. 1	1.15	0.93	0.98	1.02	0.77	0.83	13.4	12.0	12.6	24.6	15.6	15.3
Rot. 2	1.17	0.89	0.95	0.98	0.81	0.85	11.5	11.7	11.8	27.1	13.6	15.3
Rot. 3	1.16	0.89	0.95	1.02	0.76	0.82	12.0	11.0	10.9	29.2	12.6	15.9
Interactions												
$CS \times Rot. 1$	1.13	0.97	1.01	0.78	0.74	0.74	14.8	12.0	12.6	22.4	19.6	20.3
$CS \times Rot. 2$	1.20	0.89	0.96	0.77	0.74	0.75	11.6	12.2	12.1	25.5	15.6	17.9
$CS \times Rot. 3$	1.04	0.91	0.94	0.69	0.75	0.73	12.7	10.6	11.0	24.8	13.2	15.9
LIS $\times$ Rot. 1	1.17	0.89	0.96	1.27	0.80	0.91	12.1	12.0	12.0	26.7	11.5	15.1
LIS $\times$ Rot. 2	1.13	0.88	0.94	1.20	0.89	0.96	11.3	11.2	11.2	28.7	11.6	15.6
LIS $\times$ Rot. 3	1.29	0.87	0.97	1.35	0.78	0.91	11.4	11.4	11.4	33.5	12.0	17.0
Year	1.16	0.90	0.96	1.01	0.78	0.83	12.3	11.6	11.7	26.9	13.9	15.5

1.8 t DM ha<sup>-1</sup> year<sup>-1</sup>, respectively). Average crop residues produced under CS (4.4 t DM ha<sup>-1</sup> year<sup>-1</sup>) did not allow the preservation of the initial soil organic matter content, presumably because of the higher mineralization rate associated with the use of deep ploughing in such climatic conditions. Observed soil organic matter trends in our study are in accordance with those found by Blevins and Frye (1993) for the effects of tillage systems on soils having a relatively low initial soil organic matter content. Positive effects of legume crops on soil characteristics – not yet pronounced in our study – were also observed by Dalal *et al.* (1995) in a similar environment. However, these authors noted a sharp decrease in soil fertility soon after adoption of conventional tillage, which was not observed in our case. It is worth stressing that a thorough evaluation of the effects of agricultural management systems on soil fertility can only be made in the long term (Janzen, 1995).

The preliminary results of our study suggest that in Southern Italy, where high summer temperatures enhance soil organic matter mineralization rates and low rainfall limits biomass production, the adoption of a low-input system based upon no-tillage seems more appropriate for preserving the beneficial effects of setaside on soil properties. In this

respect, crop rotation and inclusion of legume crops seem less important than the crop management system type, at least in the initial phase. This type of research may provide useful indications for future management, within the European Common Agricultural Policy schemes, of land that has been setaside for 20 years.

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# Modelling Traditional Manuring Practices in the North Atlantic Context: Soil Sustainability of a Shetland

**Island Community?** 

3.9

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# Introduction

There is increasing interest in the application of soil science to questions of past land management, and sustainability of early agricultural communities. Modelling has the potential to create hypotheses linking land management and soil properties over extended historical periods. Soil properties, however, have yet to be modelled from historical and ethnographic data and tested against present-day measurements. If soil models are demonstrated to be accurate predictors of soil properties in historic contexts then this will allow questions of historical sustainability to be addressed. The island of Papa Stour, Shetland, can be examined as a site where long-term use of traditional manuring practices has been abandoned only relatively recently. These traditional practices were replaced by uniform extensive sheep grazing across the whole island. Without the burden of interference from modern agricultural practices, the island is a unique 'laboratory' with detailed ethnographic and historical information on early land management and a legacy of relict plaggen soils known to contain detailed evidence of such (Davidson and Carter, 1998; Simpson et al., 1999). In this chapter, we seek to use the ethnographic and land management information to predict plaggen soil organic carbon levels using the CENTURY soil turnover model (Parton et al., 1987) and test the predictive performance of the model against present-day soil organic carbon levels. The model is then

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used to assess the historic sustainability of manuring practices in arable areas (rig land), and its consequent implications for the wider landscape and social dimension in North Atlantic regions.

# Site

Papa Stour (60°20'N 1°42'W) is a small (2000 ha) island off the west coast of the Shetland mainland. The island is a unique site for the study of deepened soils, since traditional manuring practices continued until 1960, having probably been introduced to Papa Stour during the late Norse (~800 AD) period (Simpson, 1997; Davidson and Carter, 1998). Following the cessation of these practices, there has been little human disturbance of the soil. The manuring practice involved the translocation of soil materials and intensive use of animal manures. The USDA Soil Taxonomy classifies the resulting soils as Plaggenthrepts (Soil Survey Staff, 1998) although a more refined definition may be possible (Adderley et al., submitted). If there were no translocated material, the soils are likely to fall within the Walls association found elsewhere on mainland Shetland (Soil Survey of Scotland, 1982), which would be classified under USDA Soil Taxonomy as Humods (Soil Survey Staff, 1998). The land use of Papa Stour has a distinctive division between the intensively managed 'infield' and the extensive 'outfield' area, separated by a head dyke. Within the 'infield', it is evident that various specific types of land use practice were adopted for selected areas. The most complete ethnographic information relates to Bragasetter farm (HU 174 595) and arable rig land, and these, therefore, provide the focus of this study.

# **Ethnographic and Historical Information**

From semi-structured interviews during 1998 with present-day farmers on Papa Stour and from ethnographic evidence collected during the 1960s, a detailed picture of the now abandoned traditional manuring practices on Papa Stour emerges (Irvine, 1846; Ordnance Survey, 1886, 1901; Fenton, 1978). Other historical sources, used to provide greater time depth for the model, include the Sinclair (1791–1799), Low (1879), Skirving (1874), Donaldson (1954), Brand (1683) and Evershed (1874), confirming the general pattern of early manuring in defined areas of the landscape, which mirrors practices used in continental Europe described previously by, amongst others, Neimeier and Taschenmacher (1939) and Spek (1992). Quantification of these manuring practices was made possible by calculating volumes of kishie (basket carried by back ~90 dm<sup>-3</sup>) used to

carry turves, seaweed and manure. The manure application is expressed on a per area basis of management areas (Table 3.9.1).

# **CENTURY Modelling**

The CENTURY model developed by Parton et al. (1987) has been applied successfully to a variety of long-term experiments in a variety of climatic and cropping situations worldwide (Kelly et al., 1997). Because of this wide applicability, transparency and peer-reviewed acceptance (Smith et al., 1997), CENTURY was selected for this study. Since CENTURY models only the top 20 cm of the soil, a uniform bulk density of 1.35 g cm<sup>-3</sup>, based on the mean of five measurements from fixed depth 15-23 cm samples was used throughout. CENTURY model runs were for periods of 14 years (1946–1960) and 250 years; in this case the climatic data (e.g. Hulme, 1997) were rewound several times to simulate short-term variations. The starting values of soil C and N were derived from mean values of organic carbon measurements of three Walls series soil samples obtained from profiles close to the rigged land at Bragasetter (mean = 2.04%), and an assumed C: N ratio of 15.6. This ratio was obtained from the C: N ratio of the total C and total N measurements made on the same soils. The allocation of initial soil C and N to 'active surface', 'active soil', 'slow' and 'passive' pools is defined arbitrarily (respectively, 5, 5, 40 and 50%) but, by considering data generated from the model only after allowing the pool distributions to equilibrate in the model over a period of 10 years, this effect is minimized. This problem is also found in long-term experiments and is overcome by similar practices (e.g. Kelly et al., 1997). Parameters, where available, relating to the oat crop Avenia strigosa were obtained from field trial data. Addition of organic manures (400–500 gC m<sup>-2</sup> per manuring;

**Table 3.9.1.** Quantification of traditional manuring practice of Papa Stour infield.

Land area	Farming practice	Proportion Bragasetter farm (%)	Manure type	Frequency	Quantity per application (kg m <sup>-2</sup> )	Mass C per application (kg m <sup>-2</sup> )
Rigged	Cropping of oats (Avena strigosa)	9	Compost from midden	Biennial	~4.14	~0.44
Kailyard	Brassica and potato crops	4	Compost from midden	Annual	> 4.14	> 0.44
Planticrue	Plant nursery	< 1	Seaweed (Tang)	Annual	~5.75	~3
Grazing	Tethered grazing	86	Grazing animal	N/A	Random	Random

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Table 3.9.1) was scheduled within the model on a biennial basis, with measurement of the input made on composition of manures and semi-quantitative evidence for manure addition. A range (100–300 gC m<sup>-2</sup> per manuring) of manure addition input values have also been modelled to allow discussion of various cultural scenarios.

# Testing the model

Soil samples were collected during August 1998 on a volumetric basis from 15–23 cm fixed depths at four profiles within the rigged land. Organic carbon content was measured using a Strohlein Instruments Coulomat 702 carbon analyser adapted to analyse  $CO_2$  liberated from  $H_3PO_4$  digestion. Each sample was analysed a total of four times and a mean value calculated. All these values are expressed initially in percentage terms and subsequently have been converted, using the bulk density values obtained from replicate samples, to a mass per unit area basis to be consistent with CENTURY model outputs.

## Results

Soil organic C values output from the CENTURY model for a 250-year period demonstrate rising soil organic matter levels from ~6000 g m<sup>-2</sup> to final values of 14,880 g m<sup>-2</sup> (organic matter addition 500 g m<sup>-2</sup>) and 12,760 g m<sup>-2</sup> (organic matter addition 400 g m<sup>-2</sup>), approaching equilibrium after 150 years (Fig. 3.9.1, 250 years). These predictions relate well to the mean measured soil organic carbon (1998) value of 14,526 g m<sup>-2</sup>, and suggest that the soil predictions based on ethnographic and historical data are valid. Issues that remain to be resolved include the depth resolution limits of the model (top 20 cm), which is of particular significance in deepened soils. Short-term scenarios modelled demonstrate the biennial rise and decline of organic matter as cropping and manuring interact (Fig. 3.9.1, 14 years).

# Conclusion

The validity of the model opens up the possibility of constructing alternative manuring strategies as a way of assessing the sustainability of the study area. One critical question of the study area relates to the use of a finite turf resource on Papa Stour for fuel and animal bedding. If lower levels of turf were applied as manure to the rig land, as a way of conserving the outfield turf resource for future generations, how would this affect

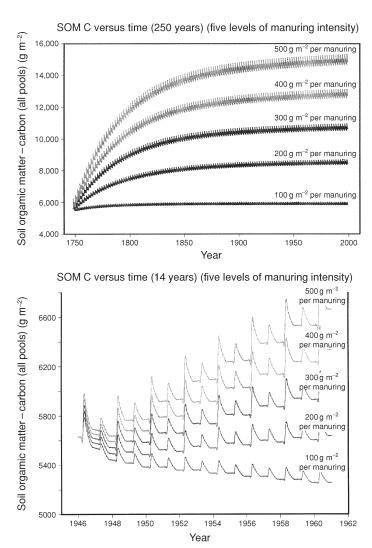


Fig. 3.9.1. Outputs of CENTURY model: 250 years and 14 years.

infield soil productivity and food security? The additional model runs (100–300 g m<sup>-2</sup>) suggest that the manuring intensity was greater (~40%) than required to maintain both short- and long-term food security. A further consequence of this was that the outfield was overexploited and reduced the longevity of the traditional land management systems. Social explanations for the continuation of land management practices limited in their sustainability include the short-term land tenure system, and insufficient cultural knowledge of long-term cultural landscape change.

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# Organic Matter and Anthrosols in Amazonia: Interpreting the Amerindian Legacy

3.10

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# Introduction

Amazonian dark earths (ADE), commonly referred to as terra preta do Índio, occur in a variety of landscape contexts throughout Amazonia in patches ranging in size from less than a hectare to many square kilometres. During the last 30 years, research by soil scientists employing diverse sampling and analytical methods has produced a sizable body of data describing these anomalous soils (e.g. Falesi, 1972; Kern and Kampf, 1989; Pabst, 1991; Glaser, 1999). ADE exhibits a wide range of chemical and physical properties, though the following suite of persistent characteristics distinguishes them from surrounding oxisols and ultisols: dark colour, high nutrient content, high soil organic matter (SOM), high pH and association with cultural debris. Recently, ADE has attracted increasing attention from geographers, anthropologists and others who are interested in what the soils may reveal about prehistoric lifeways and settlement patterns (e.g. Smith, 1980; Mora, et al., 1991; Heckenburger et al., 1999). Nonetheless, important basic questions regarding origin, formation, persistence, variation, distribution and use remain unresolved. Most discussions of ADE assume that it is a single definable soil type created by the accumulation of human waste debris in the context of long-term habitation. This chapter has three objectives. First, we assess the validity of this prevailing 'midden' model of dark earth formation. Secondly, we propose a more detailed classification of ADE that reflects its variability. Thirdly, we discuss the implications of our findings for management of tropical soils.

# Reassessing the 'Midden Model' of Dark Earth Formation

With the goal of obtaining information from the greatest variety of dark earth contexts occurring in the lower Tapajós and Arapiuns River basins of Brazilian Amazonia, we physically inspected a large number of dark earth areas from uplands, river bluffs, beaches and plateau outliers. The sites ranged in size from 0.5 ha to > 120 ha, and included both sand- and clay-dominated soil matrices. We analysed samples collected from 20 of these sites using the following procedures: (i) elemental extraction through HNO<sub>3</sub>/HCl digestion with determination by inductively coupled plasma emission spectrophotometer (ICP); (ii) organic carbon (OC) by loss-onignition; and (iii) electrometric measurement of pH in 1 : 2 soil : water mixture. Figure 3.10.1 is based on aggregate data from 75 samples taken from 12 sites.

The results of the field and laboratory analysis showed that the dark earths in the study area are generally not middens, or aggraded piles of human garbage (Woods and McCann, 1999). While some of the sites did indeed exhibit the classic indicators of human habitation – abundant potsherds and high concentrations of P and Ca – the majority did not. Furthermore, significant human-induced aggradation was not apparent even at the chemically rich sites. On the other hand, natural aggradation clearly played an important role in the formation of many ADE profiles, most notably at riverine sites subject to high rates of deposition of windand water-borne sand. At Vila Franca near the confluence of the Arapiuns and Tapajós, for example, the dark zone extended beyond 2 m below the surface. Deposition of garbage by humans was indeed important to the formation of these soils, not as a bulk contributor of mass, but rather for the indirect chemical changes that stimulated soil biota, enhanced fertility and melanized the soil.

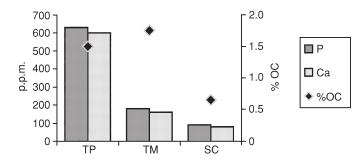


Fig. 3.10.1. Two kinds of dark earth.

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# Classification of Amazonian Dark Earths

Are we dealing with a single type of dark earth, several different entities or is each occurrence of ADE so unique as to defy classification based on characteristics attributable to human activities? Based on our laboratory and field analyses of diverse dark earth locations, we found that ADE is indeed classifiable, but it is not a discrete, single entity. Rather, we found that all of the dark earths fell into two clearly distinct categories. The first group, which we call terra preta (TP), is darker, richer in P, Ca and other elements, and contains abundant cultural artefacts (mostly ceramics, but also some lithic material). The second, and more widespread type of dark earth, is typically dark greyish brown, not black, corresponding to a Munsell colour 10YR 4/2 or darker. It is often found surrounding smaller, darker zones of TP. Ca and P levels are not significantly higher and cultural artefacts are rare. Like TP, however, it has elevated concentrations of organic carbon when compared with the background sands and clays (SC). To distinguish it from terra preta, we call this brown soil terra mulata (TM), following Sombroek (1966). In his reconnaissance of soils in the Tapajós region, Sombroek also observed expanses of slightly lighter soils devoid of ceramics encircling darker TP sites, almost certainly the same phenomenon that we now describe. We will return to the question of how TP and TM were formed and why their unique characteristics persist, but first we will present the results of additional research we designed to test the idea that two distinct types of ADE indeed exist.

# **Near Infrared Reflectance Spectroscopy (NIRS)**

NIRS holds great promise for efficient sample grouping and organic characterization – precisely those qualities needed in the dark earth study where important questions revolve around: (i) the nature of the organics that produce the dark coloration; and (ii) the degree of organic commonality between both types of the dark soils. Because laboratory infrared techniques have not yet been used extensively in the study of soils (White and Roth, 1986), we will provide some background on the technology, and describe our specific methodology.

NIRS has become a commonly used tool for both the qualitative and quantitative determination of structural information concerning organic substances. Dramatic advances in computer technology during the last 20 years have led to great improvements in NIRS instrumentation, preparation techniques and data processing capabilities. Common applications of NIRS include assessing the nutritional quality of forages and feed grains and identifying sewage sludge decomposition products. The technology is also used to analyse pharmaceuticals, textiles, dairy products, petrochemicals

and tobacco. Archaeologists have used NIRS to identify dyes, pigments, oils and resins (Goffer, 1980).

Using NIRS to test our preliminary classification of ADE, we employed relatively simple methods of sample preparation, processing and analysis. We dried the samples in a forced air oven, removed large inclusions by hand picking and then ground the soil matrix and screened it through a 2.00-mm mesh. The screened material was placed in a simple ring sample cup and automatically processed through the scanning monochrometer. The wavelength interval between data points for our analysis was 4 nm. Projected over the measured spectral range of 1108–2492 nm, this interval results in a total of 346 data points for each sample. Sample spectra interpretation was facilitated by calibration and comparison with the results from other chemical and physical determinations. We chose a chemometric statistical package called WINISI (Infrasoft International, LLC) from among the various statistical packages designed for NIRS analysis because its use of modified partial least squares regression was best suited for our investigation (Shenk and Westerhaus, 1991a,b).

# Results

For the initial phase of our test, we scanned 65 samples from the three distinct cultural and non-cultural soil contexts - TP, TM and the background sands and clays (SC) – as identified by field and laboratory methods described above. Distinctive NIR spectra of the three groups were identified and a calibration statistic developed (Table 3.10.1). To test the validity of this calibration, the individual group placement of an additional 143 samples from other known contexts in the Rio Tapajós region was then queried based upon their individual spectral signatures. Projected placement based on field observations, Munsell colour and chemical characteristics was correct for > 90% of these samples. In other words, there is discrimination between the three groups, but with variation within each data set and a small number of samples with a spectral signature that falls outside of the expected category. This is exactly what we would expect from a relatively continuous, real world, data set. Many of our samples were collected along transects initiated outside the dark earth zone and traversing both TP and TM zones, and the transitional areas between them.

In this ongoing study, we presently are testing the distinctiveness of these NIRS groupings and identifying the band ranges that account for the variation between groups. For our data set, the shapes of the TP, TM and SC spectra are very similar throughout most of the spectral range. Both the greatest activity and the greatest differences in the spectral patterns occurred at the high end of the spectrum, between 2149 and 2492 nm. As

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### Table 3.10.1. NIRS process and results.

1. Calibration process: scan a set of 'known' samples - classified on the basis of colour, chemistry and presence/abundance of artefacts – and analyse with the calibration programme to develop a function with the fewest possible terms.

Provided data:

13 Terra Preta (TP) 24 Terra Mulata (TM)

28 Background Sands and Clays (SC)

65 Total known samples

2. Test #1: use the function developed above to discriminate between a set of 'unknown' samples that included the original 65 samples.

Provided data:	74 TP	Results received:	74	TP
	49 TM <sup>a</sup>		48	TM
	85 SC		85	SC
	208 Total unknown		207	Total discriminated samples

3. Re-calibration process: the 207 discriminated samples were reprocessed with the calibration programme to create a new, more accurate function with more terms and tighter tolerances.

Provided data:

74 TP 48 TM

85 SC

207 Total known samples

4. Test #2: the revised function was used to process the same set of data used in test #1.

Results received: 71 TP

44 TM

83 SC

198 Total discriminated samples

The resulting rejections were as follows:

Of 208 samples:	Rejections consisted of:	Providing an accuracy of:
74 TP	2 TM 1 SC	95.9% for TP
49 TM	3 TP 2 SC	89.8% for TM
85 SC	1 SC 1 TM	97.6% for SC

<sup>&</sup>lt;sup>a</sup>One of the TM samples was classified as TP.

for the specific organic compounds that are affecting these spectra, there has not been sufficient work to demonstrate what these may be in our soils. Fortunately, this analysis did not appear to have been affected by the long-standing problems resulting from particle size variation, scatter and multi-colinearity encountered by other investigators (Barnes et al., 1989). We can therefore conclude then that intergroup variation is not a function of soil texture, water content or grinding.

These results support our classification of two types of ADE. The initial groupings based upon field and chemical criteria appear to be sound when their NIR spectra are compared with those of other samples from known contexts. Moreover, specific bandwidths – hence specific organic compounds – contribute markedly toward the group placement. We feel confident that NIRS can be applied fruitfully to a number of pedogenic, agronomic and cultural questions relating to Amazonian black earths. With a robust calibration, NIRS holds great potential for grouping samples on the basis of their spectral characteristics and then processing large numbers with a high degree of reliability as to their group placement. Aside from the difficulties in collecting samples in the field, a major stumbling block to studies of these anthrosols has been the high price of quantitative procedures. Consequently, only a relatively few samples on any project routinely are subjected to detailed analysis. With appropriate controls, NIRS would greatly alleviate this problem.

### **Discussion**

We have identified two distinct types of dark earth, but how were they formed? Also, especially in the case of TM, how do we know that they are anthropogenic? Our analyses have shown that TP samples are high in organic carbon – and thus organic matter content – and also have greatly elevated concentrations of Ca and P, elements strongly associated with human habitation. These characteristics, and the abundance of cultural materials, attest to the importance of household waste deposition (e.g. food processing, bones, blood, faeces, urine) in the formation of TP. It is incorrect, however, to view these soils as middens.

In the case of TM, the samples contain only slightly more Ca and P than the background sands and clays, though organic matter concentrations are equal to or even greater than those in TP. The consistently high organic concentrations in TM samples, regardless of texture, parent material or geomorphic context, strongly suggest an anthropogenic origin. However, in situ habitation waste deposition is clearly not a likely source. The most plausible explanation is that organic content was elevated through long-term soil management practices (especially mulching and burning) under intensive agriculture. Though perhaps temporarily reducing near-surface soil biota, fire more importantly contributes charcoal and ash, which affect the soil in two important ways. First, these products of incomplete combustion provide charged surfaces largely absent from both the highly weathered clays and sands in the local soils, thereby increasing nutrient retention capacity. The charcoal and ash also increase soil pH, suppressing Al activity toxic to soil biota. Increased microbiological activity adds colloidal-sized organic decomposition products to the soil

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matrix, providing still more charged surfaces and greater fertility. It is these organic complexes that coat soil particles and give the dark earths their distinctive coloration (Glaser, 1999). The spatial associations of the two types of dark earths – TP zones embedded within TM – support the hypothesis outlined above and suggest a pre-European landscape of long-term settlements and associated fields.

However, we must ask why are the distinctive qualities of high fertility, dark colour and high SOM so persistent in both types of dark earth, despite the severe tropical weathering effects of high temperature and high rainfall, and even long after cultural manipulations have ceased. They persist, we believe, primarily because of the coupled attributes of high biological activity and high nutrient-retaining capacity. Recent research has demonstrated the long-lasting influence of ash deposition in terms of increased pH (de Moraes, 1996) and higher concentrations of nutrients such as Ca and Mg (Ludwig et al., 1999). Moreover, black carbon is notoriously long lived, and not entirely inert as is often assumed (Glaser, 1999). Most telling, perhaps, is the nature of the humus in dark earth soils. Zech et al. (1990) compared TP with an Oxisol developed from the same parent material and found stable, high molecular weight, fused aromatic ring structures in the dark earths, as opposed to the readily degraded polysaccharides of the Oxisol. They identified the source of the stable organics in the TP as lignin and concluded that the process responsible was the mineralization and humification of large amounts of nutrient-rich organic materials by culturally stimulated microbial activity. Apparently, at some threshold level of biotic activity and soil nutrient retention status, dark earth attains the capacity to perpetuate – even regenerate – itself thus behaving more like a living 'super' organism than an inert mineral. Local farmers in our study area have also noted the dark earths' unique ability to maintain fertility under intensive cultivation, and those who dig it up for sale as potting soil have learned that it will regenerate itself if some of the organic-rich, biologically active dark layer is left intact.

# Implications for Tropical Soil Management

The benefits of low input farming technologies in the tropics are becoming increasingly apparent when compared with high-tech agriculture with its expensive chemical inputs and associated environmental costs. Ash contributes important nutrients, increases the cation exchange capacity and encourages microbes by suppressing the activity of toxic Al. The Kayapó people of Brazil have even developed specially formulated ashes for their specific nutritive value (Hecht and Posey, 1989). Empirically measured benefits of mulching include increased fertility, reduced soil temperature,

increased soil moisture, reduced erosion, increased SOM, conservation of SOM, weed control and pest control (Patrick and Toussoun, 1965; FAO, 1975; Sanchez, 1976, p.181; Kamara, 1986). Likewise, the importance of soil microbiota in healthy and productive agroecosytems is well documented (Lopez-Real and Hodges, 1986; Sylvia et al., 1998). Cochrane and Sanchez (1982) demonstrated that Amazonian soils managed with inputs of ash and mulch can produce high yields continuously for at least 5 years. The Kayapó, whose agricultural technologies have remained relatively intact, cultivate their fields intensively for 5 years, and less intensively for an additional six, employing a sophisticated array of soil management techniques including composting, mulching and burning.

Many local farmers throughout Amazonia also recognize the benefits of applying mulch and ash (by burning slash) but, unlike the Kayapó and the prehistoric farmers responsible for the creation of the TM expanses, most choose to practise short fallow shifting cultivation instead of a more intensive cultivation supported by soil management practices. The advent of steel tools with European contact has certainly made clearing new fields easier (Denevan, 1992), and the rapid rotation of fields explains why TM does not appear to form under today's shifting cultivation (though local farmers in our study area do notice a gradual darkening of continuously cultivated home gardens). Mulch-based cultivation need not be prohibitively labour intensive, however, as the Urarina people of Upper Amazonia recognize (Kramer, 1977). Moreover, as the persistence and regenerative qualities of ADE attest, improvements to notoriously infertile tropical soils through burning and organic inputs (or other direct manipulations of pH, cation exchange capacity and soil biota) can be significant and long lasting. Once infused with the self-perpetuating life force of an active soil biota and an adequate nutrient retention capacity, under proper management additional inputs may not be necessary to maintain a reasonable fertility. As growing numbers of shifting cultivators place ever greater pressure on forest resources, the soil-enhancing agricultural practices of pre-contact Amerindians and their legacy of rich, 'living' soils warrant further study in the search for high-yield, land-intensive, yet sustainable forms of land management in the humid tropics.

# Acknowledgements

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# Persistence of Soil Organic Matter in Archaeological Soils (Terra Preta) of the Brazilian Amazon Region

3.11

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# Introduction

Within the Oxisol and Ultisol landscape of the Brazilian Amazon region, patches of very fertile and stable anthropogenic soils, known as Terra Preta soils, occur. These soils are characterized by a large and stable soil organic matter (SOM) pool and by high stocks of nutrients such as N, P and Ca (Sombroek, 1966; Zech et al., 1990; Sombroek et al., 1993). Terra Preta soils are favoured by the local farmers because they enable high crop yields. The establishment of stable SOM seems to be decisive for the sustainable soil fertility (Duxbury et al., 1989; Zech et al., 1990). Frequent charcoal findings (Sombroek, 1966; Saldarriaga and West, 1986) provided evidence that black carbon may be responsible for the SOM stability in these soils as it is known that this carbon species can survive in the environment over centuries (Goldberg, 1985). For this reason, the main objective of the current work was to verify to what extent black carbon contributes to SOM of Terra Preta soils. Furthermore organomineral stabilization of SOM was investigated by particle size and density fractionation, and scanning electron microsopy (SEM) and energy-dispersive X-ray spectroscopy (EDX) on the separates.

## Materials and Methods

The investigation area is located in the Brazilian Amazon basin. Mean annual temperature is ~27°C and mean annual precipitation is 2050 mm at Santarém and a bit higher at Manaus. At Santarém, 90% of the rain falls in

the rainy season between December and July, whereas the yearly rainfall is more evenly distributed at Manaus (Otzen, 1992). Parent materials of all soils are Tertiary sediments. For this study, we investigated five Terra Preta sites in comparison with nearby Oxisols. Sand-rich soils (sites 1–3) are located near Manaus and clay-rich substrates (sites 4 and 5) were found in the north of Manaus and Santarém, respectively. Location and general characteristics of the investigated soils are described by Glaser (1999).

Black carbon was analysed in the fine earth, in particle size fractions (2000–250  $\mu$ m, coarse sand; 250–20  $\mu$ m, fine sand; 20–2  $\mu$ m, silt; < 2  $\mu$ m, clay), and in density fractions (< 2 g cm<sup>-3</sup>, light fraction; 2.0–2.4 g cm<sup>-3</sup>, medium fraction; > 2.4 g cm<sup>-3</sup>, heavy fraction) of Terra Preta soils and surrounding Oxisols with a novel technique using benzenecarboxylic acids (BCAs) as molecular markers for black carbon (Glaser *et al.*, 1998). The analytical procedure includes acid digestion, oxidation, sample clean up, derivatization and gas chromatography. BCAs were not produced upon oxidation of model humic substances.

# **Results and Discussion**

The SOM of Terra Preta consisted of up to 35% of black carbon, sometimes down to 80 cm soil depth, whereas in the Oxisols only the topsoil contained considerable amounts of black carbon (up to 14% of TOC). In both Terra Preta soils and Oxisols, most of the organic carbon was located in the silt and clay fractions (Fig. 3.11.1). Terra Preta soils contained significantly higher concentrations of black carbon in the fine sand and silt fractions (not shown here). In Terra Preta soils, a major part of black carbon is located in the inert carbon pool of the silt fraction. The contribution of black carbon in the clay fraction increased with increasing soil depth (Fig. 3.11.1), indicating transport by illuviation. SEM of the clay fraction showed aggregated amorphous material representing organomineral complexes besides dark wood-like particles larger than 10 μm (Fig. 3.11.2). The investigation of the elemental composition of the surface of these particles by EDX gave O: C ratios smaller than 0.5, indicating black carbon. Cellulose would have a theoretical O: C ratio of 0.88 (Stoffyn-Egli et al., 1997). Due to the low density of black carbon, particle size fractionation leaves particulate black carbon > 2 µm in the clay fraction (Fig. 3.11.2, spot 5).

Density fractionation, on the other hand, allowed the separation and the subsequent characterization of particulate black carbon in the light fraction ( $< 2.0 \text{ g cm}^{-3}$ ), whereas organomineral complexed black carbon was isolated in the medium fraction ( $2.0-2.4 \text{ g cm}^{-3}$ ) and black carbon coatings on minerals in the heavy fraction ( $d > 2.4 \text{ g cm}^{-3}$ ). Investigation of black carbon in density fractions of soils from 0-10 cm and 30-40 cm depth (Fig. 3.11.3) in combination with SEM and EDX (not shown here)

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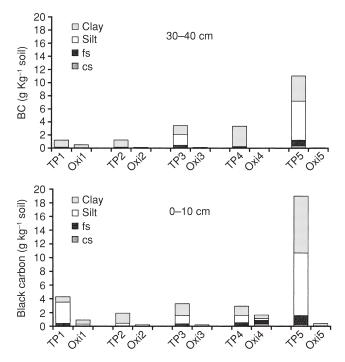
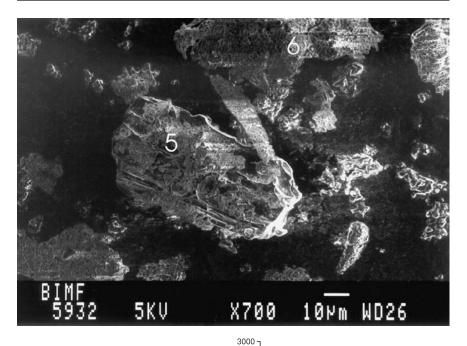


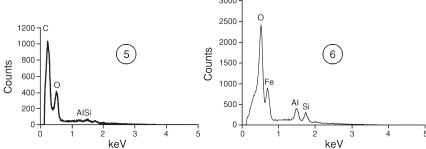
Fig. 3.11.1. Black carbon in particle size fractions.

gave evidence for a complex dynamics of black carbon in Terra Preta soils. From 20 to 70% of the total black carbon content was found in the light fraction even in 30–40 cm soil depth (Fig. 3.11.3), identifying a major part of black carbon as particulate and therefore chemically and biologically inert. The occurrence of particulate black carbon in 30–40 cm soil depth indicates transport of black carbon into deeper soil depth by turbation. In the clay-rich Terra Preta, however, the major part of black carbon was located in the medium density fraction and, therefore, was organomineral complexed. Oxidation on the edges of the polyaromatic core of black carbon produces carboxylic groups, which are involved in organomineral complexation, but also contribute exchange sites for cations and increase the cation exchange capacity (CEC). It was surprising that black carbon was also found to a minor extent in the heavy fraction. SEM and EDX showed that black carbon in this fraction was physically embedded within plaques of iron and aluminium oxides.

# Conclusions

Our investigations showed that black carbon plays a central role in the stability and therefore the sustainability of Terra Preta soils. Due to the





**Fig. 3.11.2.** Scanning electron micrograph and energy dispersive X-ray spectra of the clay fraction of a Terra Preta. 5; Particulate black carbon greater than clay size. 6; Aggregated organomineral complexes.

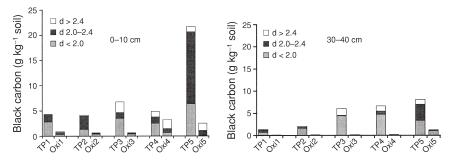


Fig. 3.11.3. Black carbon in density fractions.

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polyaromatic structure of black carbon (Haumaier and Zech, 1995; Glaser et al., 1998), it is chemically and microbially resistant and survives in the environment over thousands of years (Glaser, 1999). Therefore, charring transforms a labile carbon pool into stable SOM. Partial oxidation produces aromatic acids, which increase the CEC and also the reactivity and solubility of black carbon. Therefore, the application of black carbon together with inorganic nutrients as sustainable soil conditioner seems very promising to improve food production on infertile soils of the humid tropics.

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# Organic Matter Dynamics in Soils of the Former Lake Texcoco, Mexico

3.12

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# Introduction

Lake Texcoco (east of Mexico City, Mexico) was drained from the 17th century onwards to reduce the possibility of flooding of the city. An area was created with little or no vegetation as the soils from volcanic origin are saline or saline–sodic with a pH locally > 11. The natural drainage is poor, with a water table at 0.25–1 m depth. Weathering is progressive, and high evaporation (2000 mm) increases the salt concentration of the soil solution. The bare soil is eroded readily by wind and causes severe dust pollution over Mexico City. Pathogens carried in the dust aggravate the nuisance. A programme was therefore started at the beginning of the 1970s to vegetate the former lake, and a drainage system was installed to lower the water table. Irrigation with sewage effluent was used to remove salts and improve aeration in the root zone. A plan was also developed to apply sewage sludge to fertilize the pioneer vegetation.

The application of 1000 mg of glucose C kg<sup>-1</sup> soil did not result in a significant increase in CO<sub>2</sub> production within 70 days of an aerobic incubation at 22°C, nor did it do so in soils drained for 1 and 5 years (Beltrán-Hernández *et al.*, 1999). However, application of glucose to a soil drained for 8 years resulted in a significant increase in CO<sub>2</sub> production. To detemine the reason for this phenomenon, we applied <sup>14</sup>C-labelled glucose and investigated its fate in soil sampled from four locations characterized by different periods of drainage and vegetation density.

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# **Materials and Methods**

The experimental site is located in the former lake Texcoco in the valley of Mexico City at an altitude of 2240 m above sea level with a mean annual temperature of 16°C and mean annual precipitation of 705 mm (Beltrán-Hernández et al., 1999). Soil was sampled on 27 March 1998 at random from the 0–15 cm layer of four different plots: undrained soil and soils drained for 1, 5, and 8 years. The undrained site and the site drained for 1 year had an 80% cover entirely of Distichlis spicata, a grass adapted to high salinity. The site drained for 5 years was completely covered with D. spicata and that drained for 8 years had a diverse flora of several Gramineae, Cacti and Compositae. Characteristics of the different plots can be found in Beltrán-Hernández et al. (1999). The soil was passed through a 5-mm sieve and dried for up to 2 days until a 40% water-holding capacity (WHC) was obtained. The soil was conditioned for 6 days in drums treated with 100 ml of distilled H<sub>2</sub>O to avoid desiccation and containing a beaker with 100 ml of 1 M NaOH to trap any CO<sub>2</sub> evolved.

Fifty-four sub-samples of 50 g of soil from each site were added to 120 ml glass flasks. Half of the sub-samples were treated with a solution containing <sup>14</sup>C-labelled glucose (~40 μCi kg<sup>-1</sup> soil) and (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, and the remainder were treated with an equal amount of distilled H<sub>2</sub>O. The amount of water added resulted in a soil moisture content of ~50% WHC, and the amounts of C and N as glucose and NH<sub>4</sub><sup>+</sup> were ~1000 and 200 mg N kg<sup>-1</sup> soil. Three flasks were chosen at random from each treatment. A 20 g aliquot of soil was extracted for inorganic N with 80 ml of 0.5 M K<sub>2</sub>SO<sub>4</sub> to provide zero time samples. The samples were shaken for 30 min and filtered through Whatman No. 42 paper; the NH<sub>4</sub><sup>+</sup> concentration was determined by distillation with MgO (Bremner and Keeney, 1966), and the NO<sub>3</sub><sup>-</sup> concentration was measured colorimetrically (APHA AWWA WPCF, 1989).

The glass flasks were placed in 945 ml glass jars treated with 10 ml of distilled H<sub>2</sub>O and containing one vessel with 20 ml of a 1 M NaOH solution to trap CO<sub>2</sub> evolved. The jars were sealed to be airtight and stored in the dark for 97 days at 22°C. After 1, 3, 7, 14, 28, 42, 69 and 97 days, three jars were selected at random from each treatment, opened, the vessel containing NaOH removed and the NaOH solution titrated with appropriate concentrations of H<sub>2</sub>SO<sub>4</sub>. The soil was removed from three flasks and 20 g was extracted with 80 ml of 0.5 M K<sub>2</sub>SO<sub>4</sub> solution. The samples were shaken for 30 min and filtered through Whatman No. 42 paper and the inorganic N was measured as described for zero time samples. All remaining flasks were opened, aired for 10 min to avoid anaerobicity, sealed and incubated further.

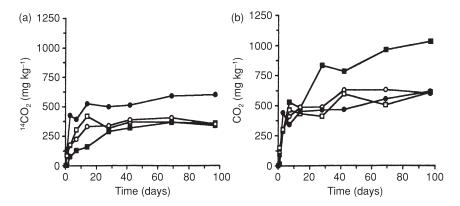
#### Results

#### Production of CO<sub>2</sub> and <sup>14</sup>CO<sub>2</sub>

The production of CO<sub>2</sub> was greatest in the undrained soil and least in the soil drained for 1 year. Approximately 19, 18, 25 and 25% of the soil organic matter mineralized within 97 days in the undrained soil and soils drained for 1, 5 and 8 years, respectively. The production of <sup>14</sup>CO<sub>2</sub> showed the same pattern in each soil; an increase in the first days of incubation, reaching a maximum with no further production thereafter (Fig. 3.12.1a). The rate of increase and the time at which the maximum value was attained, however, changed from soil to soil. Approximately 36, 36 and 38% of the glucose C added was evolved as <sup>14</sup>CO<sub>2</sub> in the undrained soil and soils drained for 1 and 5 years, respectively, but the amount was 60% in the soil drained for 8 years. The difference in CO<sub>2</sub> production between the untreated soil and the soil treated with <sup>14</sup>C-labelled glucose was greater in soil drained for 1 year than in the other soils (Fig. 3.12.1b).

#### Inorganic N

Concentrations of  $\mathrm{NH_4^+}$  in the untreated samples were variable over the first 14 days of incubation without a clear pattern, and became undetectable in each soil thereafter. Concentrations of  $\mathrm{NH_4^+}$  in the glucose-treated soil sharply decreased within the first day and were undetectable or small after 14 days (Fig. 3.12.2a). Concentrations of  $\mathrm{NO}_3^-$  in the untreated



**Fig. 3.12.1.** (a) Cumulative  $^{14}$ CO $_2$  production and (b) difference in CO $_2$  production between the glucose-treated and the untreated soil (mg C kg $^{-1}$  dry soil) in soil from an undrained site (□) and sites drained for 1 (■), 5 (○) and 8 years (•) treated with 1000 mg of  $^{14}$ C-labelled glucose C kg $^{-1}$  dry soil and 200 mg of NH $_4$ +-N kg $^{-1}$  dry soil incubated aerobically at 22°C for 97 days.

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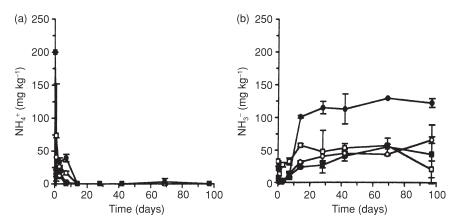


Fig. 3.12.2. (a)  $NH_4^+$  and (b)  $NO_3^-$  concentrations (mg N kg<sup>-1</sup> dry soil) in soil from an undrained site ( $\square$ ) and sites drained for 1 ( $\blacksquare$ ), 5 ( $\bigcirc$ ) and 8 years ( $\bullet$ ) treated with 1000 mg of  $^{14}C$ -labelled glucose C kg<sup>-1</sup> dry soil and 200 mg of  $NH_4^+$ -N kg<sup>-1</sup> dry soil incubated aerobically for 97 days at 22°C. Bars are  $\pm$  1 sp.

sample remained  $< 50 \text{ mg NO}_3^-\text{-N}$ . The  $\text{NO}_3^-$  concentrations in the glucose-treated soil showed a decrease in the first 3 days, but increased thereafter in all soils; the largest increase was found in the soil drained for 8 years (Fig. 3.12.2b).

#### **Discussion**

#### Production of CO<sub>2</sub> and <sup>14</sup>CO<sub>2</sub>

The undrained soil and soil drained for 1 year, with an electrolytic conductivity of 40 and 80 dS m<sup>-1</sup>, respectively, showed the smallest percentage of organic C mineralized, suggesting that the salts inhibited the microbial activity. The decrease in CO<sub>2</sub> production from 2.1 to 0.89 g C kg<sup>-1</sup> after 90 days reported by Pathak and Rao (1998) with an increase in electrolytic conductivity in the soils from 1 to 97 dS m<sup>-1</sup>, was much larger than we found. However, they added salts to soil (NaCl and CaCl<sub>2</sub>) so the microorganisms may not have been adapted to osmostic and/or specific ion stress and this might have inhibited their activity.

The percentage mineralized glucose C in the soil drained for 8 years was similar to values reported by Bremer and Kuikman (1994), but it was less in the other investigated soils. The differences in percentages of mineralized glucose C pointed to a possible effect of large salt concentrations on metabolic processes, but possible effects of other soil characteristics, such as pH, cannot be excluded.

In a previous experiment, we applied 1000 mg of glucose C kg<sup>-1</sup> soil. A significant increase in CO<sub>2</sub> production was found in soil drained for 8 years compared with the untreated soil, but not in undrained soil or soils drained for 1 and 5 years (Beltrán-Hernández et al., 1999). The results found in the experiment reported herein confirm a difference in mineralization of organic material added, but not its sequestration, i.e. its availability as C-substrate for soil microorganisms. The difference between the two experiments presumably was related to a difference in the time of sampling. Soil used in the experiment reported herein was much wetter upon sampling than the soil used by Beltrán-Hernández et al. (1999). Gutiérrez-Castorena (1997) stated that the amorphous materials provide the soil with a very high water retention capacity (2.4-5 kg kg<sup>-1</sup>) which decreases upon drying; presumably also affecting other soil characteristics and thus the capacity to sequester added organic material. Compared with the untreated soil, the application of glucose increased the CO<sub>2</sub> production more than the amount of <sup>14</sup>CO<sub>2</sub> produced. Accelerated decomposition of unlabelled soil organic matter following the addition of organic material has often been referred to as a 'priming effect' (Brookes et al., 1990). These authors described situations where a true priming effect is caused mainly by an increased turnover of microbial cell C (after addition of glucose) or by an increased decomposition of native soil organic matter (after addition of rye grass). It was difficult to pinpoint what happened in our soil, but it appeared to be related to an increased turnover of microbial cell C (no data shown).

#### Inorganic N

Soils from the former lake Texcoco are characterized by limitations in inorganic N, and > 150 mg of N kg<sup>-1</sup> soil could not be accounted for after 3 days of incubation in the glucose-treated soil. Losses through NH<sub>3</sub> volatilization were < 10 mg of N kg<sup>-1</sup> soil (no data shown) so it was immobilized in the soil microbial biomass. Nitrifiers in soil of the former lake Texcoco were adapted to elevated salt concentrations, as indicated by increases in NO<sub>3</sub><sup>-</sup> concentrations in the glucose-treated soil without accumulation of NH<sub>4</sub><sup>+</sup>.

It was concluded that sequestration of added organic C as found in previous experiments was not confirmed, but sod characteristics changed by drainage affected the decomposition of added organic material.

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## Low-input Ecological Rice 3.13 Farming in Bangladesh

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#### Introduction

The population in Bangladesh is 120.8 million people and currently growing at an annual rate of 1.8% (Bangladesh Bureau of Statistics, 1997). With a reduction in the per capita availability of cultivatable land, productivity must be increased. Modern cereal varieties introduced throughout Asia during the 1960s differed from the traditional varieties in their short stature and stiffer straw. These plants do not lodge when N fertilizer is applied, allowing them to produce higher yields. Generally farmers have sought to maximize economic gain through the use of fertilizers and agricultural chemicals to control weeds, diseases and pests. Modern varieties are less sensitive to day length than traditional varieties, and where other climatic factors and water supply are not constraints, continuous rice cropping is possible. Using pumps to extract groundwater for irrigation, modern varieties are now grown in the dry season in addition to the crop traditionally grown in the monsoon season between July and September. The emergence of the double-cropped rice system was central to the green revolution.

However, there is concern that rice yields may be declining under such intensive cultivation in several Asian countries including Bangladesh (Cassman and Pingali, 1995). There is some evidence for this from experimental stations, but the anecdotal evidence from farmers – who are more constrained by productivity factors such as labour, fertilizer and pesticide – supports the view that greater inputs are required to maintain the same level of yield.

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The intrinsic sustainable fertility of traditional rice farming systems, maintained by biochemical processes, will normally provide a low level of yields, of the order of 2 t ha<sup>-1</sup> (Giller and Wilson, 1991). Rice farming systems in Bangladesh still rely substantially on returns of organic matter and biological N fixation from blue-green algae to maintain their inherently high level of fertility. PROSHIKA, a non-governmental organization, and others promote 'ecological farming'. Their goal is to 'promote an ecological agricultural system which is productive, equitable and cost effective'. Farmers are taught that using no-tillage, mulches, compost and green manures improves soil fertility. In principle, no chemical fertilizers or pesticides are to be used. PROSHIKA provides motivation and formal training to farmers, who then are encouraged to adopt ecological agricultural practices and pass on information to others.

Due to the shortage of fuel, farmers have to burn crop residues and up to 80% of their cowdung (Parikh, 1988), whereas the landless will burn all that their cow produces rather than using this manure to improve or maintain soil fertility. Thus, in our experience, farmers rarely convert all their fields to ecological management.

During field visits, farmers working with PROSHIKA suggested that fields which they converted to ecological management practices were more fertile. During discussions, they cited improvement in soil physical properties, described as ease of ploughing and increased biological activity, as indicators of these changes. This chapter seeks to see whether these observations can be confirmed using established quantitative methods to compare conventional farming systems that include chemicals with the ecological farming system.

#### **Materials and Methods**

Four different village sites were located in the following thanas (smallest administrative unit): Dhamrai, Daulatpur, Gabtali and Shibganj; the thana name was used to identify the field sites. The field sites were chosen on the basis of different flooding regimes and were located in two districts of Bangladesh, Dhaka and Bogra. Dhamrai and Daulatpur represent lowland areas (Dhaka district), which are subjected to flooding from the local rivers during the monsoon season; flood depths range up to 2 m and both are south of the Jamuna River. Gabtali and Shibganj represent medium high land flooding conditions with flooding depths up to 1 m, mostly due to rainwater flooding at the same time (June–September).

Ten fields were selected at random from an immediate village area for each of the two farming systems, although the selection process depended upon farmers following the necessary management system and being willing to participate in the experiment. At Dhamrai, fields had been under 7 years of ecological management; all other fields had been under ecological management for between 3 and 5 years. Soils did vary across sites and a composite soil sample was taken from the plough layer (i.e. to ~12–15 cm). Concurrently, samples were collected for soil moisture content and bulk density determination using undisturbed 5-cm cores taken from just below the soil surface.

Soils were dried and ground to pass through a 2-mm sieve before analysis. Soil properties measured were bulk density, total C, anaerobic N mineralization (Anderson and Ingram, 1996) as an index of potential N mineralization, and soil organic matter fractions that are postulated as being sensitive to changes in management. The organic matter fractionation procedure (Sohi *et al.*, 1998) uses sequential density separations to separate free organic matter (with no dispersion other than manual swirling) and intra-aggregate organic matter (after ultrasonic disruption of aggregates). Carbon and N in the fractions were determined by combustion, using a combined C and N analyser and mass spectrometer (Europa Integra CN). Sub-samples were also re-wetted and incubated for 11 days at 30°C until they came to microbial equilibrium. Soil microbial biomass C and basal respiration were then determined (Witt *et al.*, 2000).

#### **Results and Discussion**

It is known that organic matter influences soil physical, chemical and biological properties upon which soil fertility depends (Gaunt *et al.*, 1995). One of the main benefits from using regular organic inputs on the ecological fields was a significant difference in the bulk densities (Table 3.13.1). Ecological management led to lower average bulk densities in three villages, with Shibganj being the exception. Shibganj lies on the Barind Tract and has higher clay content than the other sites, the parent material consisting of Madhupur clay. These results appear to confirm farmers' perception of the ecological practice that the fields were easier to cultivate, whilst the conventional fields were found to be 'hard'.

		Bulk densit	y (g cm <sup>-3</sup> )	
Farming system	Dhamrai	Daulatpur	Gabtali	Shibganj
Conventional	1.07	1.29	1.29	1.28
Ecological	0.94*	1.23*	1.22*	1.31*

**Table 3.13.1.** Bulk density of soils from all four villages.

<sup>\*</sup>Significant difference at the 1% level.

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At two of the sites (Dhamrai and Daulatpur), total C was significantly greater in the ecological fields than in conventional fields, but this was not so in the two other sites. Indeed, at Gabtali it was significantly lower in the ecological fields (Table 3.13.2). Total soil C and N change slowly and are less sensitive than light fractions to management practices (Christensen, 1987). The free organic matter would be expected to be most sensitive because it turns over most rapidly. However, it will, by its nature, be affected by sampling time, whereas intra-aggregate material, whilst still sensitive, is less susceptible to seasonal fluctuations (S.P. Sohi, personal communication, 1999). Significantly greater amounts of free (P = 0.05) and intra-aggregate (P = 0.001) material were recovered in the ecological fields at Dhamrai and Daulutpur, whereas lower amounts of free (P = 0.05) and intra-aggregate (P = 0.001) material were recovered at Gabtali and Shibganj. These findings are in line with the differences in soil total C (Table 3.13.2). Dhamrai has a difference of 2072 kg C ha<sup>-1</sup> for the

**Table 3.13.2.** Parameters of 'soil fertility'.

		, .			
Management	Soil fertility parameter	Dhamrai	Daulatpur	Gabtali	Shibganj
Conventional	Total C (kg ha <sup>-1</sup> )	29,241	30,177	30,373	33,661
Ecological	Total C (kg ha <sup>-1</sup> )	30,737*	42,129*	27,234*	33,294
Conventional	Free organic matter C (kg ha <sup>-1</sup> )	981	1,364	1,760	1,469
Ecological	Free organic matter C (kg ha <sup>-1</sup> )	1,500*	1,715*	1,370*	1,500
Conventional	Intra-aggregate C (kg ha <sup>-1</sup> )	1,161	1,119	1,119	2,216
Ecological	Intra-aggregate C (kg ha <sup>-1</sup> )	3,233**	1,816**	1,056	1,925**
Conventional	Microbial biomass C (kg ha <sup>-1</sup> )	210	226	333	377
Ecological	Microbial biomass C (kg ha <sup>-1</sup> )	253**	323	150**	404
Conventional	Basal respiration (kg C ha <sup>-1</sup> h <sup>-1</sup> )	2.89	1.98	2.41	1.68
Ecological	Basal respiration (kg C ha <sup>-1</sup> h <sup>-1</sup> )	2.42*	2.97	1.25	2.36*
Conventional	Pot. available N (kg N ha <sup>-1</sup> h <sup>-1</sup> )	9.98	3.17**	6.66	2.52**
Ecological	Pot. available N (kg N ha <sup>-1</sup> h <sup>-1</sup> )	9.95	8.56*	7.76	5.20*

<sup>\*</sup>Significant at the 5% level; \*\*significant at the 1% level.

intra-aggregate between the two management practices after 7 years of organic inputs.

The soil microbial biomass is responsible for the organic matter turnover and nutrient cycling in soils. Microbial biomass C ranged from 150 to 404 kg C ha<sup>-1</sup>, equivalent to 0.67-1.2% in the soils investigated (Table 3.13.2), comparable with the typical range of 1-4% (Gaunt *et al.*, 1995). Microbial biomass C was greater in ecological fields at all villages, with the exception of Gabtali. Basal respiration followed a pattern similar to total C (Table 3.13.2), being greater in ecological than conventional fields at Daulatpur and Shibganj, but less at Gabtali and Dhamrai; differences at Shibganj and Dhamrai were significant (P = 0.05). There was no significant difference in potential N mineralization at Dhamrai and Gabtali, whereas both Daulatpur and Shibganj had significantly larger mineralizable N pools under the ecological management (Table 3.13.2).

Farmers who are engaged in ecological farming also continue with 'conventional' practices on other fields, but the two systems were not practised in isolation. Nutrient budgets for the same fields (data not shown) showed, even on conventional fields, that farmers added up to two-thirds of their fertilizer as organic manure.

Thus it is not surprising that there are not large differences in soil properties between the two systems. Fields at Dhamrai that have been under ecological management for a longer time demonstrate a significant increase in total soil C, C in free and intra-aggregate organic matter fractions, microbial biomass C and basal respiration.

#### Conclusion

Given that the organic inputs varied annually for both farming systems, it is not surprising that differences between systems after 3 years of ecological management were not entirely consistent. We will re-analyse these fields after a further 3 years to assess the longer term impact of ecological management. The data in Table 3.13.2 suggest that a suite of measurements should be used as indicators of soil fertility rather than an individual measurement.

If farmers' observations and measurable soil properties can be linked, then we can develop learning exercises that are related to this biophysical understanding. We also wish to link our predictive tools and quantitative indicators. In this way, we hope to use organic matter fractions as measurable pools in a soil organic matter turnover model (Gaunt *et al.*, 1999). Linking models and indicator measurements in this way has the advantage that models can, in principle, be used in a predictive manner without previous site knowledge (Arah and Gaunt, 2000).

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# The Influence of Cultivation on the Composition and Properties of Clay-Organic Matter Associations in Soils

### 3.14

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#### Introduction

One of the main questions about the effect of land use on soil properties is which soil constituents are affected and what is the impact of land use change on soil properties. Cultivation decreases soil organic matter (SOM) stocks, in particular labile fractions, such as particulate organic matter (Cambardella and Elliott, 1992). Organic matter in the  $< 2 \mu m$  fraction of soils is less depleted by cultivation (Christensen, 1992; Balesdent *et al.*, 1998). This is ascribed to the slow turnover rate of  $< 2 \mu m$  SOM, due to its chemical nature and presumably also to its protection from decomposition by the clay minerals (Balesdent *et al.*, 1998).

The basic units of soil aggregates are elementary associations of clay minerals and oxides with organic matter (Emerson *et al.*, 1986). Clay-associated organic matter is identified in most studies with the organic matter present in the  $< 2 \mu m$  fraction of soils. However, in this fraction, organic matter can be associated with clay particles or present as discrete entities (Chenu *et al.*, 1998).

The aims of this chapter are to assess the effect of cultivation on the organic matter present in the < 2  $\mu m$  particle size fraction, and in particular on the forms of organic matter present in this fraction and their degree of association with clay particles, and on the physical properties of this fraction. We focused on clay dispersibility because it seriously affects large soil pores and leads to surface sealing and associated problems (Kay and Dexter, 1990).

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#### Materials and Methods

Soil samples were collected from the French Pyrenean piedmont, from a *Pinus pinaster* forest and from adjacent agricultural sites, converted to intensive maize cropping for 7 and 35 years. Soils are thick humic acid loamy soils, developed from Quaternary silty alluvial deposits, classified as vermic Haplumbrepts in the soil taxonomy (Arrouays, 1992). Samples were collected from the ploughed layer or from an equivalent depth in the forest soil (Balesdent *et al.*, 1998). Soils were air dried and clods were broken apart to < 4 mm.

Particle size fractionation of the soil was based on mechanical dispersion of the soil and is described in detail by Balesdent *et al.* (1998). Briefly, in a first step, soil was dispersed by agitation with glass beads. The > 50  $\mu$ m particle size fraction was discarded by sieving, sonicated to disrupt microaggregates, and particle size fractions were separated by sedimentation. The < 2  $\mu$ m fractions were recovered after flocculation with CaCl<sub>2</sub> and then freeze dried. Aliquots were kept for transmission electron microscopy without flocculation and freeze drying. All separations were performed in triplicate. C and N contents were determined by dry combustion. Results are expressed on an oven dry mass basis. Mineralogy of the < 2  $\mu$ m fractions was determined with an X-ray diffractometer after OM oxidation with H<sub>2</sub>O<sub>2</sub>.

For transmission electron microscopy, the < 2 µm suspensions were first concentrated by centrifugation and equilibrated with 0.32 kPa. Millimetre sub-samples were fixed with glutaraldehyde and osmium, then exchanged with methanol, propylene oxide and embedded in a Spurr resin (Chenu and Jaunet, 1992). Two samples were embedded for each soil situation, each coming from different replicate fractionations. Ultrathin sections (80 nm) were made and deposited on metal grids. The sections were stained with uranyl acetate, lead citrate (Lewis and Knight, 1976) or silver proteinate (Thierry, 1967) to contrast specific functional groups of the organic matter and were observed on a Philips EM 420 TEM coupled with a AN 10000 EDS elemental X-ray analyser. The organic constituents were identified on the basis of their morphology and reaction with stains. We used the heavy elements staining the ultrathin sections, which were easily detected with EDS, as probes for organic matter. On five photographs taken at random at the same magnification for each sample ( $\times 37,000$ ), each representing a surface of 16 µm<sup>2</sup>, we identified on morphological criteria (i) organic particles; (ii) organomineral aggregates; and (iii) mineral particles. We classified as mineral particles those in which organic features were not detectable on paper prints of the photographs at the working magnification. The contours of particles and aggregates were traced manually on the photographs, scanned and we measured their numbers, dimensions and surfaces using the NIH-Image program for Macintosh computers.

The amount of clay dispersed from soil depends both on the clay dispersibility per unit of exposed surface area and on the exposed surface areas of aggregates (Kay and Dexter, 1990). In order to focus on the former, we measured clay dispersibility on 2–4 mm aggregates. Air-dried aggregates (20 g) were immersed in 75 ml of de-ionized water and the vials were shaken end-over-end for 1 h. Dispersible clay at this stage was issued from aggregate disruption at scales > 0.2 mm. Dispersed clay was recovered by three successive sedimentations and freeze dried. The experiments were performed in triplicate for each soil. Particle size fractionation was also performed on sub-samples of 2–4 mm aggregates.

#### Results

#### Characteristics of the $< 2 \mu m$ fractions (Table 3.14.1)

The clay-sized fractions were composed of kaolinite, illite and chlorite, with small amounts of quartz. The comparison of the particle size fractionation with the classical mechanical analyses of the soils indicated that all clay was not recovered in the forest soil and in the soil cultivated for 7 years: very stable microaggregates could not be dispersed with ultrasound and remained in the silt fractions. The C content of the < 2  $\mu$ m fractions decreased with cultivation, but the contribution of C in forest < 2  $\mu$ m to soil C increased with cultivation.

#### Clay dispersion

Clay dispersibility at 1 h was calculated by dividing the amount of dispersed clay by the total clay in 2–4 mm aggregates. It increased with time of cultivation and was related linearly to the C content of dispersible clay

Time of cultivation	Ma: (g 100 g		C cor		C:	N	C	Dispe cla (% F <	ау	C cor dispersi (mg C	ble clay
(years)	Mean	SD	Mean	SD	Mean	SD	(76 GGII	Mean	SD	Mean	SD
0	14.7	1.3	117.8	5.2	15.2	0.2	29%	7.3	(0.4)	89.2	0.4
7	15.5	1.3	77.5	1	13.2	0.2	38%	10.7	(0.2)	65.8	11.4
35	16.8	1.9	48.1	1.4	12.2	0.2	48%	15.3	(0.2)	38.7	2.5

**Table 3.14.1.** Characteristics of the  $< 2 \mu m$  fractions.

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(Table 3.14.1,  $r^2 = 0.998$ ). Dispersible clay had a lower C content than the total  $< 2 \mu m$  fraction.

#### Microstructure

Different forms of organic matter were observed in the < 2  $\mu$ m fractions: micrometre cell wall or plant cell residues (Fig. 3.14.1a), microorganisms or their remnants (Fig. 3.14.1b) and shapeless and structureless organic matter (Fig. 3.14.1c). This organic matter was either free, i.e. not associated with minerals (Fig. 3.14.1a and b), or bound to clay particles. Microaggregates enclosing plant debris were observed (Fig. 3.14.1d) as well as complex clay aggregates in which the OM occurred as thin layers between small stacks of clay sheets (Fig. 3.14.1e). Organic constituents were much more abundant in thin sections from the forest soil than in those from the soils cultivated for 7 or 35 years. Preliminary results from image analyses showed that organomineral aggregates and free organic particles were more abundant in the forest sample than in < 2  $\mu$ m fractions from the cultivated soils (Table 3.14.2).

#### **Discussion and Conclusions**

Cultivation of the humic loamy soils of this area led in a few years to decreased aggregate stability and erosion problems (Le Bissonnais and Arrouays, 1997). The present results show that decreases in aggregate stability on the millimetre scale are accompanied by changes on the micro-scale: clay-organic matter microaggregates were less abundant and clay was more easily dispersed. Increases in clay dispersibility were also found by Curtin *et al.* (1994) and by Fuller *et al.* (1995). In a previous study (Chenu *et al.*, 1998, 2000), we measured contact angles of water on the < 2 µm fractions from the same soils. The wettability of the fraction decreased with the organic matter content on the fraction and with cultivation. Changes in macroaggregate stability in these soils with cultivation were then partly due to modifications of the composition of the clay-sized fractions and hence of their cohesion and wettability.

Balesdent *et al.* (1998) found that soil OM turnover was eight times slower under forest than in cultivated soils. Our electron microscopy observations showed that OM was largely associated with clay minerals in the  $< 2 \, \mu m$  fraction of the forest soil, both as coatings in clay particles and entrapped within very stable microaggregates. With cultivation, organomineral microaggregates were much less abundant. We suggest that OM was physically protected in the forest soil by virtue of its association

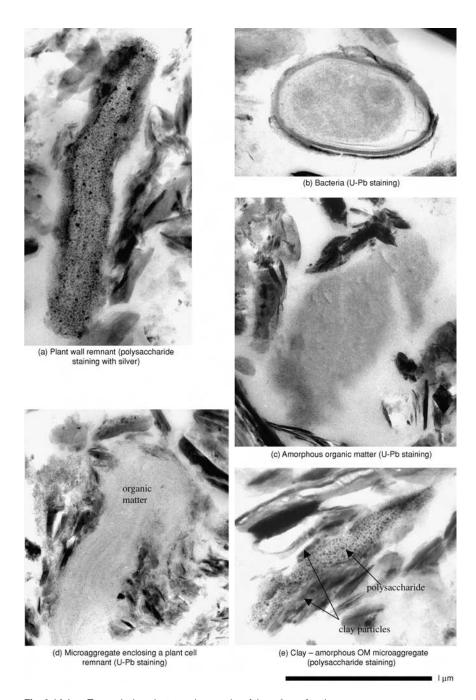


Fig. 3.14.1. Transmission electron micrographs of the < 2  $\mu$ m fractions.

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			9	6 total surfac	ce of particl	es	
	No. of particles – analysed	Organo aggre		U	anic icles	Min parti	
		Mean	SD	Mean	SD	Mean	SD
Forest	219	47.9	13.4	12.0	3.6	40.0	12.4
Cultivated 7 years Cultivated 35 years	422 570	9.4 5.9	8.4 7.3	4.5 6.4	4.3 10.0	89.5 87.7	9.7 16.9

**Table 3.14.2.** Abundance of morphological classes of particles observed with TEM in fraction  $< 2 \mu m$ .

Electron micrographs of the < 2 µm fractions.

with clay minerals and that disruption of the microaggregates by tillage or exposure to rain in cultivated soil caused its de-protection.

#### **Acknowledgements**

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# The Influence of a 3.15 Grass-Clover Mixture on Soil Organic Matter and Aggregation of a Podzolic Loamy Sand Soil

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#### Introduction

Sustainable land use is fundamentally dependent on maintaining adequate concentrations of soil organic matter. Organic matter is known to be essential in the self-regeneration of soil structure, water-stable aggregation, water holding and air capacity following mechanical damage to soils. Two main mechanisms have been proposed to explain the physical protection of organic matter against decomposition: association with clay particles and occlusion within aggregates (Hassink *et al.*, 1993; Golchin *et al.*, 1995). The stabilization of organic matter within soil micro- and macroaggregates is closely related to the water-stable aggregation, which is one of the key indicators of sustainable land use. This parameter has been found to be sensitive to the effects of different tillage systems, crops and organic fertilizers (Ekwue, 1990; Franzluebbers and Arshad, 1997).

At present, only low rates of animal manure are incorporated into arable soils in Russia. An increasing use of perennial vegetation and green manure instead of animal manure is being observed in the current management practices in the northwestern region of Russia. A critical problem is whether soil quality can be maintained with the use of perennial grass—clover mixtures as organic fertilizers.

The objective of the present study was to estimate the effects of a seven-course rotation on the relationships between the size distribution of water-stable aggregates and organic matter content in a light-textured podzolic soil under two diverse tillage treatments. Discussion is restricted to the data for the organic matter content of the whole soil and aggregates and

aggregate size distribution following perennial grass-clover mixture and spring wheat crops.

#### **Materials and Methods**

The experiment started in 1982 at the Agrophysical Research Institute's experimental station located ~60 km southwest of St Petersburg. Plots were stablished on a podzolic loamy sand soil in a seven-course rotation with a grass-clover mixture (Phleum pratense and Trifolium pratense) for 2 years, potato (Solanum tuberosum), spring barley (Hordeum vulgare), winter rye and downy vetch (Secale cereale and Vicia villosa), winter rye (Secale cereale) and spring wheat (Triticum vulgare). All the crops were grown in each year from 1982. The plots were not replicated and there was only one plot for each crop in each year. Tillage treatments consisted of spring mouldboard ploughing (conventional tillage) at a depth of 20-22 cm and spring disking (minimum tillage) at a depth of 10-12 cm. The size of the plots was 30 m × 186 m (for each crop) and 15 m × 186 m (for each tillage treatment). No fertilizers were applied to any of the crops. The grass-clover mixture was used as the only source of nutrients for the crops of all the seven stages of the rotation. The grass-clover mixture was used for hay. Soil samples (0-10 cm) were taken from the plots with grass-clover mixtures (the first and second stages of the rotation) and from the plot with spring wheat (the seventh stage of the rotation) in May and September of 1998. Five samples of soil were taken from each plot and mixed. Three subsamples were taken from these bulked samples and all the soil properties were determined from these and used for analysis. Organic matter contents in the whole soil samples and in the water-stable aggregate size fractions were determined by wet oxidation according to Tjurin's procedure (Kaurichev, 1986). Measurements of aggregate size distribution were carried out by dry and wet sieving with a rotary sieve (Vadjunina and Korchagina, 1986). Sieve sizes were 0.5, 1.0, 2.0, 3.0, 5.0 and 7.0 mm.

#### Results and Discussion

The growing of perennial grass-clover mixtures is known to contribute to a considerable input of plant residues and hydrophobic organic substances into soils. Studies have shown that the percentage of water-stable aggregates increases with increasing content of organic matter originating from grasses (Ekwue, 1990; Broersma *et al.*, 1997). Such changes in wet aggregate stability are very important, especially for light-textured soils, which usually demonstrate both low organic matter content and a low percentage of water-stable aggregates.

The distribution of organic matter contents in the whole soil and in the 0.5–7.0 mm water-stable aggregates is presented in Table 3.15.1. At the end of the 2-year growing period of the grass-clover mixture, the organic matter content of the whole soil was higher than that after the spring wheat and was higher on the plots with the grass-clover mixture after minimum tillage than after conventional tillage.

The data for the spring wheat plot (Table 3.15.1) show that there was a significant (P < 0.05) decrease in organic matter content of the whole soil during the 5-year period of the crop rotation which followed the grass-clover mixture.

The above trends were also observed in most of the fractions of 0.5–7.0 mm water-stable aggregates. However, after the first year of the grass-clover mixture, there was no greater increase in the organic matter of the aggregates compared with that of the whole soil. Only after 2 years of grass-clover mixture was there a higher accumulation of organic matter in the aggregates than in the whole soil. Despite such positive changes in organic matter content in the soil after 2 years of the perennial grass-clover mixture, the increase of organic matter in the aggregates was not large. Subsequent crops did not lead to any accumulation of organic matter in soil aggregates. In contrast, a more or less pronounced decomposition of organic matter in the water-stable aggregates was observed during the

**Table 3.15.1.** Organic carbon content in the whole soil (C), in 0.5–7.0 mm water-stable aggregates (C\*) and total amount of 0.5–7.0 mm water-stable aggregates in the podzolic loamy sand soil affected by crops and tillage systems.

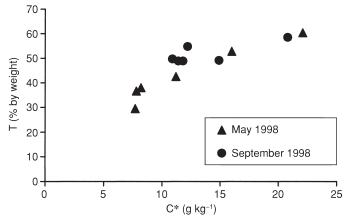
Type of tillage	Crop	Date of soil sampling	C (g kg <sup>-1</sup> )	C* (g kg <sup>-1</sup> )	T (% by weight)	Average diameter of the water-stable aggregates (mm)
Conventional	SW	May 1998	21.2	7.8	36.6	2.1
tillage		September 1998	23.9	11.4	48.9	4.1
	PG1	May 1998	22.3	11.2	42.5	3.3
		September 1998	24.1	12.2	54.8	4.4
	PG2	May 1998	26.8	22.1	60.4	4.0
		September 1998	28.1	14.9	49.1	4.0
Minimum	SW	May 1998	19.5	7.7	29.5	3.3
tillage		September 1998	21.7	10.9	49.7	3.7
	PG1	May 1998	23.9	8.2	38.0	3.2
		September 1998	25.7	11.8	48.9	4.0
	PG2	May 1998	30.9	16.0	52.8	3.2
		September 1998	32.9	20.8	58.5	3.7

SW = spring wheat; PG1 = perennial grass-clover mixture (first year); PG2 = perennial grass-clover mixture (second year).

5-year period of crop rotation following the 2 years of perennial grass-clover mixture. The total amount of 0.5–7.0 mm dry aggregates was equal to 60–74% (spring wheat) and 56–80% (perennial grass–clover mixture). The total amount of dry aggregates usually decreased by 4–20% from May to September, with almost no change under the influence of the conventional and minimum tillage. The conventional and minimum tillage did not significantly affect the total amount of the 0.5–7.0 mm water-stable aggregates, although they did affect the total organic matter content.

The total amount of 0.5–7.0 mm water-stable aggregates was higher after establishment of the grass-clover mixture, especially after the second year, than after spring wheat (Table 3.15.1). This presumably was due to the greater input of organic matter originating from the grass and clover plants in the pasture. An increase in total amount of water-stable aggregates was observed during the growing period on the plots with perennial grass-clover mixture and spring wheat. Data on the average diameter of water-stable aggregates (Table 3.15.1) showed that the amount of large water-stable aggregates rose from May to September. On the one hand, soil wetting and drying and the consolidating action of thin roots and organic matter particles possibly resulted in the observed formation of the large water-stable aggregates in September. Over-winter processes seem to lead to a breakdown of these aggregates into finer water-stable aggregates.

The correlation coefficients for the relationships between total amount of 0.5–7.0 mm water-stable aggregates and organic matter content in these aggregates were equal to 0.97 (in May) and 0.75 (in September) for the grass-clover mixture for both tillage treatments (Fig. 3.15.1). In contrast to the humified organic matter, the fresh organic matter accumulated by the end of the growing period played a minor role in the water-stable



**Fig. 3.15.1.** Relationship between total amount of 0.5–7.0 mm water-stable aggregates (T) and organic carbon content in the water-stable aggregates (C\*).

aggregation. The differences in organic matter stabilization can be explained by the association of humified organic substances with clay particles and by the occlusion of the organic matter within aggregates. However, because of the low amount of clay particles in this soil, the stabilization of organic substances from perennial grass—clover mixture was insufficiently strong to protect this organic matter against decomposition and the water-stable aggregates against breakdown during the further period of the crop rotation. As a consequence of this weak stabilization of organic matter, a decrease in the amount of water-stable aggregate size fractions was observed 5 years after growing of the perennial grass—clover mixture of second year use.

#### **Conclusions**

The growing of a perennial grass-clover mixture for 1 and especially 2 years resulted in increases in organic matter content of both whole soil and aggregates and in water-stable aggregation of podzolic loamy sand soil. Compared with conventional tillage, the use of minimum tillage resulted in (i) increasing total organic carbon content and (ii) decreasing water-stable aggregate organic matter content. Nevertheless, there were no significant differences in the effects of the conventional and minimum tillage on water-stable aggregation. There was an accumulation of total organic carbon in the whole soil and in the water-stable aggregates, and in amount of 0.5–7.0 mm water-stable aggregates from May to September for both types of soil tillage. During the further 5-year period of crop rotation with potato, spring barley, spring wheat, winter rye together with downy vetch and spring wheat, decreases in total and water-stable aggregate organic matter content and amount of water-stable aggregates were observed.

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## The Role of Soil Organic Matter and Manures in Sustainable Nutrient Cycling

4

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#### Introduction

Soil organic matter (SOM) is vital to the sustainable use of soil because of its role in maintaining soil structure, water-holding capacity, the microbial biomass and soil fauna, and in nutrient cycling. The maintenance of SOM through rotational lev-arable farming, and the effective utilization of crop residues and manures, are central to organic farming, but there are increasing indications that good SOM, manure and residue management may also be important for conventional agriculture. For example, for many years yields of wheat on the plots receiving optimum amounts of fertilizers on the Broadbalk Experiment at Rothamsted were as good as, if not better than, those produced with farmyard manure (FYM). Recently, however, the largest yields (and, unfortunately, the largest losses to the environment) have been obtained from the plot given FYM in autumn and extra nitrogen (N) fertilizer in spring (Johnston, 1997). The reasons for this change are not yet clear. The yield differences could be caused by a better supply of nutrients from the manure, e.g. the slow, regular release of mineral N from manure through the growing season better meets the N requirements of the growing crop than the large, single, spring application of fertilizer N currently used on Broadbalk. If so, this could be corrected relatively easily by applying more appropriate types and amounts of fertilizers at the correct times. Alternatively the difference could be due to more complex changes such as improvements in soil structure, water-holding capacity and the root environment caused by manure, or even to the impact of FYM on weeds and below-ground pathogens.

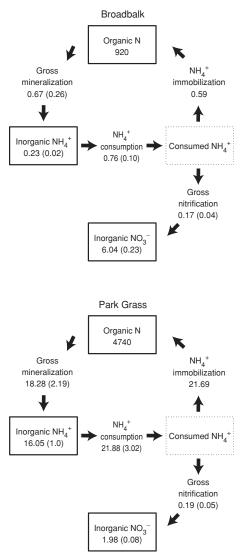
This chapter discusses some further evidence for the importance of good organic matter management in maintaining nutrient supplies, and the conflict between this and minimizing losses to the environment. It does this by moving progressively through three areas of research that currently are of great international interest and which dominate the chapters in this part of the book. First, it reviews developments in the measurement of gross N transformations, especially mineralization; such measurements enable the release of N from the breakdown of SOM, crop residues and manures to be better studied, and highlight the central role of carbon (C) in N cycling. Secondly, it considers the size and significance of the soluble organic N pool in soils. This pool is proving to be central to rapid C and N turnover, but also a potential organic source of N loss to the environment. Thirdly, it reviews some recent research showing the greater effectiveness of manures over fertilizers for supplying phosphorus (P) and potassium (K).

#### **Gross N Transformations**

If N is to be used efficiently in agroecosystems, the factors and mechanisms that influence its supply must be understood. In the past, it was difficult to determine the relative importance of key processes in N turnover – ammonification, nitrification and immobilization – because they could not be separated. Isotopic pool dilution with <sup>15</sup>N enables gross rates of N transformations to be determined (Powlson and Barraclough, 1992). Measurements made on soils from a range of land uses at Rothamsted and elsewhere show how these techniques are identifying key processes and linking process rates to land use, management and other controlling factors, especially C (Murphy *et al.*, 1998, 1999a).

Soils were collected from a plot of the Broadbalk Experiment, which has received no fertilizer since its inception in 1843 and has been under a 5-year rotation of fallow, potatoes, wheat, wheat, wheat since 1986, and from the unlimed, unfertilized plot (3) of the Park Grass Continuous Hay Experiment. Soil samples were taken from each plot, sieved to < 5 mm, bulked to give a representative sample, and stored at 4°C in the dark until used. Isotopic pool dilution experiments were conducted on three replicates from each bulk sample, as described by Willison *et al.* (1998), to measure gross rates of ammonification, ammonium consumption and nitrification (Fig. 4.1). Ammonium immobilization was estimated as the difference between ammonium consumption and gross nitrification.

Net rates of mineralization for the two experiments are of the same order of magnitude: we measured them to be 0.13 and 0.55 mg N kg<sup>-1</sup> day<sup>-1</sup> for Broadbalk and Park Grass, respectively. However, gross N transformation rates (between arrows in Fig. 4.1) and pool sizes (in boxes in Fig. 4.1) are strongly affected by land use and differ by at least one order



**Fig. 4.1.** Gross rates of N transformations (mg kg<sup>-1</sup> day<sup>-1</sup>) and pool sizes (mg kg<sup>-1</sup>) in soils from unfertilized plots of the Broadbalk and Park Grass experiments at Rothamsted (Tlustos *et al.*, 1998).

of magnitude. This particular Broadbalk plot is not typical of conventional arable land since it does not receive any N fertilizer, only 35–40 kg N ha<sup>-1</sup> year<sup>-1</sup> from the atmosphere (Goulding *et al.*, 1998). The soil has a slow rate of gross mineralization and a very slow rate of gross nitrification. Immobilization dominates over nitrification. The soil is N limited, with

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		Pools IN kg	<sup>-1</sup> )		Proce (mg N kg			
Site	Land use	Organic N		NO <sub>3</sub> <sup>-</sup>	Gross mineralization	NH <sub>4</sub> <sup>+</sup> consumption	Gross nitrification	Immobilizationa
Broadbalk	Arable	920	0.2	6.0	0.7	0.8	0.2	0.6
Park Grass	Grass	4,740	16.1	2.0	18.3	21.9	0.2	21.7
Knott Wood	Wood	3,590	8.9	35.5	2.5	3.1	1.2	1.9
Lakenheath	Wood	30,950	6.4	53.1	3.5	4.7	4.9	$-0.2^{b}$
Lakenheath	Arable	29,390	2.0	19.1	3.6	4.2	1.9	2.3

**Table 4.1.** Gross rates of N transformations and pool sizes in soils from Rothamsted and Lakenheath.

yields of only 1 t ha<sup>-1</sup> year<sup>-1</sup> grain in continuous wheat and 2 t ha<sup>-1</sup> year<sup>-1</sup> in rotation, and unlikely to lose much N. This is confirmed by measurements which show that, on average, only 12 kg N ha<sup>-1</sup> year<sup>-1</sup> are leached from this plot. Park Grass has a rapid rate of gross mineralization but a relatively slow rate of nitrification. This soil is unlikely to lose N because it is cycling so rapidly between mineralization and immobilization. We thus have two N-limited soils, which have very different rates of gross mineralization–immobilization turnover (MIT).

To begin to explore the links between land use and MIT, gross N transformations were also measured on soil from Knott Wood (300-year-old coppiced deciduous woodland at Rothamsted) and from drained fenland peat at Lakenheath in Suffolk, which had been under either intensive arable cropping for the last 80 years or a poplar wood for the last 30 years (Willison *et al.*, 1998) The pool sizes and process rates for N transformations in all the soils are shown in Table 4.1.

The dominance of immobilization over nitrification on the Park Grass plot could be because its acidity (pH currently 5.2 in water) suppresses nitrifiers. However, Knott Wood soil is also acid, but N cycling there is not dominated by immobilization: the soil is N-saturated and losing N. Generally, for immobilization to dominate over nitrification and for N to be conserved, sufficient C is needed to drive the heterotrophic organisms that immobilize ammonium. Thus C and N availability determine the rate and balance of MIT and the supply of nitrate available for plant uptake or loss to the environment. Exudates from the dense root mass on Park Grass could be the source of the C needed to cause immobilization to dominate. This would explain why such rapid rates of MIT were measured there but not on the Broadbalk 'Nil' plot with its similar atmospheric input of N.

<sup>&</sup>lt;sup>a</sup>By difference; <sup>b</sup>i.e. zero.

The soil under Knott Wood has a rate of gross nitrification (1.2 mg N kg<sup>-1</sup> day<sup>-1</sup>) approaching that of gross immobilization (1.9 mg N kg<sup>-1</sup> day<sup>-1</sup>). Thus ~60% of the soil N supply is recycled whilst the remainder potentially is lost from the system after nitrification into the large nitrate pool (35.6 mg kg<sup>-1</sup>). Knott Wood appears to be N-saturated through atmospheric inputs of ~100 kg N ha<sup>-1</sup> compared with requirements by the trees of ~10 kg N ha<sup>-1</sup> year<sup>-1</sup>. This view is supported by its rapid emission rates of nitrous oxide of 0.7 kg N<sub>2</sub>O ha<sup>-1</sup> year<sup>-1</sup> (Goulding *et al.*, 1998).

The Lakenheath soils have the same rates of gross mineralization (3.6 mg N kg<sup>-1</sup> day<sup>-1</sup> for arable, 3.5 mg N kg<sup>-1</sup> day<sup>-1</sup> for woodland) and ammonium consumption (4.2 mg N kg<sup>-1</sup> day<sup>-1</sup> for arable, 4.7 mg N kg<sup>-1</sup> day<sup>-1</sup> for woodland), and their organic N pools are both 30 g kg<sup>-1</sup>. Thus the mineralization rate of the peat has not changed with the change in land use. This rate presumably depends on the size and composition of the mineralizable fraction of the SOM, which remains the same under arable farming and woodland. The main differences between the soils are in the gross nitrification rates (1.9 under arable, 4.9 under woodland) and gross immobilization rates (obtained by difference, 2.3 under arable, -0.2, i.e. zero, under woodland). We suggest that the extra atmospheric N scavenged by woodland causes an imbalance in the C: N ratio and MIT and thus the large N losses from Knott Wood and the Lakenheath woodland.

## The Size and Significance of Soluble Organic N and Dissolved Organic N in Soils

The forms of N present in the soil and lost in drainage have been researched for many years. Lawes and Gilbert (1881) made gravimetric analyses of the organic and inorganic contents of drainage waters after laboriously distilling a > 100 l sample. They reported that the amount of dissolved organic matter was small and highly nitrogenous, with a mean C: N ratio of 2.6: 1. We have been measuring both soluble organic N (SON, i.e. N extracted from soil with water or a salt solution, in this case KCl), and dissolved organic N (DON, i.e. N in drainage water samples) collected from the same experiments (Murphy *et al.*, 1999b). Our aim is to quantify amounts of SON in a range of soils under various management systems and how much of this is lost to surface waters as DON.

Recent studies have confirmed that as much SON as mineral N can exist in soil under agricultural cropping systems. Jensen *et al.* (1997) showed that 0.5 M K<sub>2</sub>SO<sub>4</sub>-extractable SON in the top 15 cm of soil varied seasonally between 8 and 20 kg SON-N ha<sup>-1</sup> in a coarse sand and between 15 and 30 kg SON-N ha<sup>-1</sup> in a sandy loam; the minimum occurred during winter and the maximum in late summer. McNeill *et al.* (1998) showed that, in the top 10 cm of a loamy sand, SON comprised 55–66% of the total

soluble N (TSN) under wheat (18 kg SON-N ha<sup>-1</sup>) and pasture (28 kg SON-N ha<sup>-1</sup>). Our data (Table 4.2) show, for 'snapshot' samples from a wide range of soil types (0–30 cm), a KCl-extractable SON content of 20–30 kg N ha<sup>-1</sup> and a surprisingly constant ratio between mineral N and SON in arable soils, with SON comprising ~40–50% TSN. In a range of soils on an organic farm, SON in the 0–25 cm layer accounted for 80% of TSN and ranged from 24 to 46 kg SON-N ha<sup>-1</sup>, increasing with the number of previous years under grass–clover ley (data not shown). Clearly, SON is a significant pool within agricultural soils, but how much is leached? Table 4.3 shows some data recently collected from the Broadbalk Continuous Wheat Experiment at Rothamsted: DON in samples collected

**Table 4.2.** Mineral nitrogen (SMN) and soluble organic nitrogen (SON) pools extracted in 2 M KCl from the 0–30 cm layer of 12 soils from a range of sites in England under arable cropping in spring 1999. Values in parentheses are standard errors of the mean of three replicates.

Soil type	Previous crop	Bulk density (g cm <sup>-3</sup> )	SMN (kg N ha <sup>-1</sup> )	SON (kg N ha <sup>-1</sup> )
Sandy loam	Winter wheat	1.01	26 (6)	29 (8)
Silty loam	Winter wheat	1.44	35 (3)	33 (3)
Sandy clay loam	Winter rape	1.42	37 (3)	22 (4)
Sandy clay loam	Winter wheat	1.27	81 (3)	30 (1)
Silty clay loam	Celery	1.31	26 (1)	34 (1)
Clay loams (two soils)	Winter wheat	1.13-1.41	25–32 (2)	24–28 (3)
Clay loam	Field peas	1.31	21 (3)	33 (1)
Clays (four soils)	Winter wheat	0.97-1.36	27–33 (6)	23–33 (3)

**Table 4.3.** Mineral N and soluble organic N (SON) in a 2 M KCl extract from the 0–75 cm soil layer (samples collected on 7 December 1998), and dissolved organic N (DON) in the drainage waters collected from tile drains at 65 cm during September–November 1998, on the Broadbalk Experiment. Values in parentheses are standard errors of three replicates. There were no replicates of drainage water samples.

		N applied (kg ha <sup>-1</sup> year <sup>-1</sup> )						
			144	288	240 as FYM			
Source	Analysis		N conten	t (kg ha <sup>-1</sup> )				
Soil extract:	Mineral N DON	19.3 (1.3) 46.9 (8.9)	35.9 (1.3) 23.7 (8.7)	49.3 (2.2) 55.3 (11.7)	82.9 (3.0) 60.5 (11.9)			
Drainage water:	Mineral N DON	9.9	6.3	29.0	52.0 7.0			

from the drains is compared with SON measured in 2 M KCl soil extracts. (The comparison is semi-quantitative because the DON and SON were not sampled at the same times.) The data show that the DON pool is equivalent to only 2-10% of the SON pool, increasing in the order nil N plot < N fertilizer plots < FYM plot. However, ~10% of the N leached from drains is organic, and significant amounts of DON are leached from plots receiving FYM.

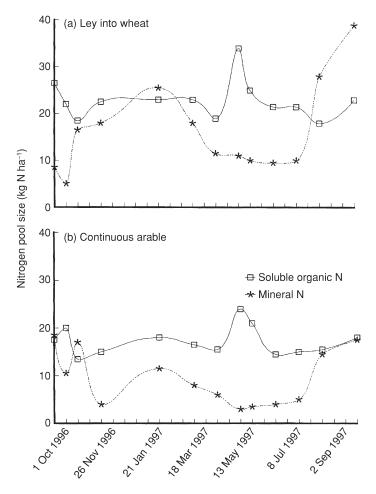
Thus the amount of N that is likely to be leached in organic forms appears to be much less than that contained in soluble organic forms within the soil. This raises the question of how much SON is mineralized to ammonium and nitrate prior to leaching or whether it is taken up directly by crops (Murphy *et al.*, 1999b). Its contribution to N loss from arable agriculture would seem to be small, except from manured land.

We have also measured the dynamics of mineral N and SON through an agricultural year in the plough layer (0-23 cm) of the Ley-Arable Experiment at Woburn Farm (Fig. 4.2). SON is not as dynamic as mineral N, but it changes through the year. In particular, the quantity of SON increases during periods of plant uptake and mineralization-immobilization. However, we have no conclusive evidence of a causal link. Under continuous arable cultivation, the size of the SON pool is relatively constant at ~15-20 kg SON-N ha<sup>-1</sup>, decreasing with leaching in early winter and increasing markedly during the period of rapid root growth in spring (Fig. 4.2b). Under ploughed-out grass, the SON pool size is larger at ~20–25 kg SON-N ha<sup>-1</sup> and the changes are larger than under continuous arable, but occur at the same times of the year and in the same way (Fig. 4.2a). The dynamics of the SON and mineral N pool in the plough layer were reflected in sub-soil down to 90 cm (data not shown). The period of rapid plant growth both above and below ground appears to play an important role in SON dynamics. In this soil at least, there is a relatively constant pool of SON (related to SOM content and soil texture) and a more dynamic pool of SON which reflects current plant dynamics. We are studying the chemical composition of these pools.

#### Improving the Efficiency of Use of P and K Fertilizers

Field experiments around the world, applying P fertilizer with manure on contrasting weakly and strongly P-fixing soils, are testing the hypothesis that, if P can be kept in organic forms, fixation is reduced and availability and crop yields can be increased. Results are encouraging (Greenland, 1997; Sanchez *et al.*, 1997). Table 4.4 shows yields of maize from the 23-year-old Kabete Experiment at the Kenyan Agricultural Research Institute (KARI). As with the Broadbalk Experiment, at the beginning of the Kabete Experiment, NP fertilizer gave as good if not better yields than those

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**Fig. 4.2.** The seasonal dynamics from October 1996 to September 1997 of mineral N and soluble organic N in the surface soil (0–23 cm) under (a) an 8-year grass ley ploughed immediately after the first sampling and planted with wheat, and (b) a continuous arable plot ploughed at the same time.

obtained with FYM; recent yields show that the trend has reversed and FYM, with or without fertilizer or residues, gives the best yields. However, the data in Table 4.4 suggest that this is not because there is more 'available P' in this plot; available P levels are quite low in the FYM-treated plot.

It is possible that, where manure was added, the inorganic P was converted into labile organic and microbial forms rather than fixed as inorganic P. Also, organic matter sorbs on to P-fixing surfaces, blocking fixation. The cause would appear to be linked to SOM because the organic carbon content has been maintained more effectively where manures were

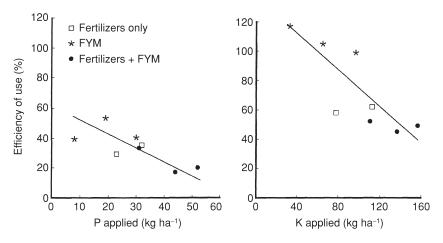
applied (Table 4.4). However, soil carbon levels have decreased under cultivation even on manured plots.

The efficiency of manures and fertilizers for supplying P and K have been compared in three long-term experiments in Europe. Blake *et al.* (2000a,b) measured P and K balances in the plough layers of the Broadbalk Continuous Wheat Experiment at Rothamsted, a 97-year-old rotation experiment at Bad Lauchstaedt in Germany, and a 76-year-old experiment at Skierniewice in Poland on rotations and continuous rye. The P and K were applied as fertilizers, FYM and a combination of both.

Figure 4.3 shows the efficiency of use of applied P and K (i.e. that removed in the crop); that not used was fixed in the soil or leached below the sampling depth (plough layer). Efficiencies of > 100% for K indicate

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Kabete Exp	eriment at the Kenyan Agricultural Researc	h Institute (KARI).	
Table 4.2.	Organic carbon contents, available P cont	ents and maize yie	elds in the 23-year-old

	Organic carbon (%)		Available P	Maize yield (t ha <sup>-1</sup> )	
Treatment	1976	1998	(mg kg <sup>-1</sup> ) 1998	1976	1998
Nil	2.8	1.62	13.1	3.8	1.2
NP		1.63	88.0	4.3	2.3
FYM		1.99	38.2	3.9	3.2
FYM + NP		2.07	129.1	4.0	2.9
FYM + NP + crop residues		2.15	107.2	4.0	3.0



**Fig. 4.3.** Efficiency of phosphorus and potassium applied to three soils from the UK, Germany and Poland when applied as fertilizers, manures (FYM) or a combination of the two.

that K was released from non-exchangeable reserves or taken up from below the sampling depth; this could also have occurred where efficiencies were < 100%. The soils contain ample P and K after > 70 years of application, and so the efficiency of use decreases with the amount applied. The results do show, however, that FYM produced the most efficient use of P and K, and a combination of FYM and fertilizers was generally the least efficient. However, efficiency of use is only one way of judging the effectiveness of manures and fertilizers. Combinations of FYM and mineral K produced the largest yields and offtakes.

Plant availability and the utilization of P and K were controlled by the nature and extent of the sorption surfaces in the soils, both mineral and organic, and the capacity for fixation. Fertilizers were least effective in the most strongly P- and K-fixing soil at Rothamsted, and most effective in the sandy loam soil at Skierniewice, which held the nutrients in an available form without fixing them. Also, where FYM increased the cation exchange capacity (CEC) of soils by adding humic materials (Skierniewice; Blake et al., 2000a) or blocked K-fixing sites through adsorption onto clay surfaces (Rothamsted; Goulding and Talibudeen, 1984), this improved K utilization.

Deficiencies of other nutrients decreased the effectiveness of applied P and K. FYM was generally more effective than fertilizers because these nutrients were supplied in the manure. However, the release of N and P from organic manures is strongly dependent on the mineralization rate. This is obviously not likely to be a problem in East Africa, but at Skierniewice the cold climate greatly reduced the rate of mineralization and caused N and P deficiencies when only FYM was applied.

#### Conclusions

Researchers, farmers and advisors are faced with a dilemma. Organic manures and crop residues should be recycled and used effectively to maintain SOM levels, supply nutrients and make farming more sustainable. In addition, C plays a central role in controlling the rate of N cycling. Soils in which C supply matches N input cycle N tightly, keeping it within the system; N-saturated soils deficient in C are more likely to lose N to the environment. Carbon supply from manures and residues is therefore beneficial. Manures also supply a broad range of nutrients, and they can be more effective than fertilizers, especially in soils with a range of nutrient deficiencies or which fix P or K or both. However, rates of mineralization of N and P from SOM and manures are difficult to predict and even harder to control. Rates of release are determined by environmental conditions and are not necessarily well synchronized with crop uptake. This can lead to

larger leaching losses, especially of N and including soluble organic forms of N.

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## Can Concepts of N Saturation Developed for Forest Systems be Applied in Arable Soils?

4.1

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#### Introduction

Nitrogen (N) fluxes in soils are dependent on the supply of, and demand for, mineral N. Both are controlled by interacting biological, chemical and physical factors. Mineral N can be supplied to the soil as fertilizers, or returned in animal excreta, as well as through atmospheric deposition and from ammonification (commonly termed N mineralization) of soil organic matter (SOM) and crop residues. The development of <sup>15</sup>N isotopic dilution techniques has provided an opportunity to obtain a much clearer understanding of soil N cycling. The principal advantage of these techniques is that the actual rate of ammonification (termed gross N mineralization) can be estimated without the confounding influence of immobilization. Similarly, gross rates of nitrification (the total production of nitrate without any influence of immobilization or gaseous losses of nitrate (NO<sub>3</sub><sup>-</sup>) during the incubation) can be measured without addition of ammonium  $(NH_4^+)$ , which stimulates the process. The rates of specific pathways of the soil N cycle can therefore be quantified. The gross N mineralization rate determines the supply of mineral N from organic matter, while both immobilization and nitrification rates represent microbial demands for N within the system (Fig. 4.1.1).

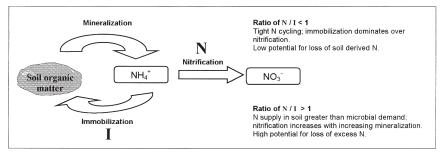
Concepts and measurements of potential N loss have been used for some years to assess the 'N saturation' status of forest systems (Tietema and Wessel, 1992; Tietema, 1998). In forest ecosystems and undisturbed

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grasslands, the N cycle was thought to be 'highly conservative', where high rates of mineralization were balanced by high rates of immobilization, with minimal net N mineralization or nitrification. However, additions of N in atmospheric deposition can disrupt the soil N cycle.

Stimulation of the process of nitrification is of key importance, since  $NO_3^-$  is vulnerable to loss by leaching or denitrification. The ratio of gross nitrification to  $NH_4^+$  immobilization has been used to assess the absolute potential of the soil to exchange N with the wider environment (Fig. 4.1.1; Tietema and Wessel, 1992). The greater the ratio, the more likely a soil is to lose N via leaching or denitrification. The lower the ratio, the more likely the soil is to retain N within the internal cycling of mineralization—immobilization turnover (MIT). This ratio can be used to assess the 'N saturation' of forested soils and, along with measurements of  $NH_4^+$  supply, can be used to examine the potential for N loss.

In arable soils, it is generally assumed that nitrification occurs rapidly, when NH<sub>4</sub><sup>+</sup> supply permits. A large pool of NH<sub>4</sub><sup>+</sup> is rarely maintained in arable soils. Nitrifiers respond rapidly to additions of NH<sub>4</sub><sup>+</sup> in fertilizer or manure, where NH<sub>4</sub><sup>+</sup> supply is in excess of requirements for maintenance and growth of the heterotrophic microbial population, or when this is restricted by carbon availability. In contrast to forests, N uptake by crops can also be rapid in agricultural systems, representing a further sink for 'surplus N'. It is therefore only during periods when crop uptake is small that the ratio of gross nitrification to NH<sub>4</sub><sup>+</sup> immobilization forms an index of potential N loss. In humid temperate climates, coincidence of low crop uptake and high effective rainfall over winter may mean that indices of potential N loss can be applied in agricultural systems, as well as forests. This was evaluated preliminarily by Goulding *et al.* (1998). In this study, we further evaluated these indices in a range of arable agricultural soils,



**Fig. 4.1.1.** The soil internal N cycle. The ratio of gross nitrification to NH<sub>4</sub><sup>+</sup> immobilization can be used to indicate the potential for loss of N.

where we have applied <sup>15</sup>N isotopic dilution techniques to examine the internal cycling of N.

#### **Materials and Methods**

Agricultural soils were sampled from the plough layer (0–30 cm) of 12 sites throughout England with varying textures in arable or ley-arable rotations (Table 4.1.1). The <sup>15</sup>N isotopic dilution technique requires labelled <sup>15</sup>N to be distributed throughout the soil so that the unlabelled soil NH<sub>4</sub><sup>+</sup> pool (or, in the case of nitrification, the NO<sub>3</sub><sup>-</sup> pool) becomes uniformly enriched with <sup>15</sup>N. This was achieved by applying 20 ml of (<sup>15</sup>NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> at 60.9 atom % or K<sup>15</sup>NO<sub>3</sub> at 61.2 atom % at a rate equivalent to 1.5 mg N kg<sup>-1</sup> dry soil to pre-incubated soil (300 g of soil at field capacity). Solutions were added dropwise while rotating the incubation vessels (see Willison et al., 1998). A nitrification inhibitor (± acetylene treatments) was added to the headspace above soil labelled with (15NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> to minimize problems associated with preferential nitrification of the applied versus indigenous NH<sub>4</sub><sup>+</sup>. Soil was removed from the incubation vessels on days 1, 3 and 7 for <sup>15</sup>NH<sub>4</sub><sup>+</sup>-labelled soil and days 1, 7 and 14 for <sup>15</sup>NO<sub>3</sub><sup>-</sup>-labelled soil. At each sampling, 80 g of soil was removed for mineral N extraction in 160 ml of 2 M KCl. Values are presented on a dry weight basis. NH<sub>4</sub>+-N and

**Table 4.1.1.** Site characteristics, gross mineralization and nitrification rates and N: I ratios for ten arable soils sampled in spring 1999.

Soil texture	Previous crop	Current crop	Gross mineralization rate (mg N kg <sup>-1</sup> day <sup>-1</sup> )	Gross nitrification rate (mg N kg <sup>-1</sup> day <sup>-1</sup> )	Ratio N : I
Sandy loam	W. wheat	W. wheat	0.76	0.44	0.50
Sandy clay loam	W. OSR	W. wheat	0.56	0.24	0.67
Silty loam	Celery	W. wheat	0.57	0.28	0.74
Clay	W. wheat	W. wheat	1.66	0.45	0.27
Clay loam	W. wheat	W. wheat	1.05	0.30	0.44
Clay loam	W. barley	W. OSR	0.96	0.46	0.92
Silty loam	W. wheat	W. OSR	1.28	0.51	1.42
Clay loam	Dried peas	W. wheat	1.05	0.61	2.10
Silty clay loam	W. wheat	W. barley	0.80	0.40	0.75
Clay loam	W. wheat	W. OSR	0.88	0.35	0.66

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NO<sub>3</sub><sup>-</sup>-N concentrations were determined colorimetrically by flow injection analysis (Skalar SAN<sup>PLUS</sup>). The <sup>15</sup>N : <sup>14</sup>N isotopic ratio of the NH<sub>4</sub><sup>+</sup>-N or NO<sub>3</sub><sup>-</sup>-N within soil extracts, after preparation by diffusion, was determined by mass spectrometry (Europa Tracermass). Gross N mineralization and gross nitrification rates were calculated from the change in pool size, and the decline in atom % <sup>15</sup>N excess, of the <sup>15</sup>N-labelled NH<sub>4</sub><sup>+</sup> or NO<sub>3</sub><sup>-</sup> pool, as soil-derived organic <sup>14</sup>N was ammonified or indigenous <sup>14</sup>NH<sub>4</sub><sup>+</sup> was nitrified (Kirkham and Bartholomew, 1954; Barraclough, 1991). The rate of microbial consumption of NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup> was also calculated. Ammonium immobilization was estimated as the difference between total NH<sub>4</sub><sup>+</sup> consumption, estimated from the dilution of <sup>15</sup>NH<sub>4</sub><sup>+</sup>, and gross nitrification, estimated from the dilution of <sup>15</sup>NO<sub>3</sub><sup>-</sup>. Although this is not a direct measurement of immobilization, it does enable the relative importance of the two major microbial NH<sub>4</sub><sup>+</sup> consumptive processes to be examined.

Calculated gross N transformation rates were used to describe the internal cycling of N in each soil. The ratio of gross nitrification to  $NH_4^+$  immobilization (N : I) was then used to determine the potential for N loss from each soil.

#### **Results and Discussion**

Gross mineralization rates ranged from 0.56 to 1.66 mg N kg<sup>-1</sup> day<sup>-1</sup>, and gross nitrification rates from 0.24 to 0.61 mg N kg<sup>-1</sup> day<sup>-1</sup> for sites sampled in spring 1999 (Table 4.1.1). There were no clear patterns with soil type or with current or previous crop. When ratios of gross nitrification to NH<sub>4</sub><sup>+</sup> immobilization were calculated, these were dominantly < 1 (average 0.7), except where a cereal followed peas (2.1) or where oilseed rape was grown (average of 1.0 for three sites). Where the same soil was sampled before and after cultivation in autumn 1997, the ratio of gross nitrification to NH<sub>4</sub><sup>+</sup> immobilization increased rapidly (within 1 day) (Fig. 4.1.2), and before NO<sub>3</sub><sup>-</sup> leaching was detected, illustrating that the processes which control soil N dynamics can respond rapidly to management practices (Goulding *et al.*, 1998). In all these cases, soil N supply (mineralization) was stimulated above the immediate heterotrophic microbial demand (immobilization), thus increasing the amount of NH<sub>4</sub><sup>+</sup> available for nitrification.

In these cases, however, increases in soil N supply through cultivation or incorporation of low C: N residue crop residues or leaf litter are well known to increase soil N supply, and the use of the complex measurement of the 'index of potential N loss' was probably not worthwhile. However, these data show that the index can be applied to arable soils, if careful interpretation is also used.

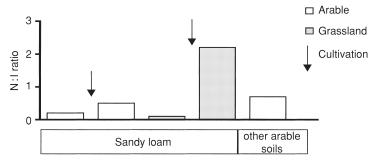


Fig. 4.1.2. Ratio of gross nitrification/NH $_4$ \* immobilization for soils sampled in autumn 1997; the sandy loam soils were sampled before and after cultivation.

#### Conclusion

Gross N transformation data can be used to examine specific pathways of the soil N cycle in greater detail than was previously possible. Such data can also be used to study the dominant processes 'consuming'  $NH_4^+$  in soil. This may lead to an indicator of potential N loss or degree of 'N saturation'. This indicator seems to be appropriate for use in arable soils, with careful interpretation, as well as in forested systems. However, the range of values for this ratio indicating low, medium or high potentials for N loss (or 'N saturation') still needs to be defined. To achieve this, additional data sets are being obtained from our ongoing research.

Apart from the relative dominance of each N pathway in a soil, the actual amount of N loss will also depend on the size of the NO<sub>3</sub><sup>-</sup> pool, soil physical properties, climate and any direct losses from fertilizer or manure. Integrated use of measurements, indices and modelling approaches in the prediction of N loss is likely to give the best results, with the actual combination of measurements and models determined by the specific questions to be answered.

#### Acknowledgements

We would like to thank Maureen Birdsey and Wendy Wilmer for <sup>15</sup>N analysis. This work was partially funded by the UK Ministry of Agriculture, Fisheries and Food, the German Ministry for the Environment and the Open University Research Fund. IACR-Rothamsted receives grant-aided support from the Biotechnological and Biological Sciences Research Council.

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# Nitrogen Mineralization Under Bare Soils After the Destruction of Grazed Pastures

4.2

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#### Introduction

The proportion of herbage grazed by cows in intensive livestock systems in Brittany (France) currently is increasing, as a result of the reduced cost of production (compared with maize plus concentrate-based systems). Moreover, the frequency of mixed grass—clover pasture is increasing, in order to reduce N fertilizer applications and to obtain a better distribution of production throughout the season (particularly in summer). Most of these grasslands are regularly destroyed (after 4–7 years) to grow silage maize or winter wheat, releasing large amounts of nitrogen into the soil and thus increasing the risk of nitrate leaching. This risk must be taken into account when comparing the sustainability of different systems. The work presented here had two aims: (i) to determine the amount and kinetics of nitrogen mineralization following the destruction of grassland; and (ii) to determine the factors which affect mineralization, i.e. previous N management (fertilization rate and timing), type of grassland (pure grass or grass—clover) and soil organic matter content.

#### Materials and Methods

The experiments were conducted on two sites in Brittany during the period 1990–1998. The first site, Kerlavic, was a well-drained sandy loam soil of 0.8 m depth, with 6.5% organic matter content and a pH of 5.8. Four treatments were set up: one was clover–grass sward receiving

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50 kg N ha<sup>-1</sup> year<sup>-1</sup> (KL-CG50N); the other three were pure ryegrass given 0, 200 and 400 kg N ha<sup>-1</sup> year<sup>-1</sup> (respectively KL0N, 200N and 400N). The second site (Kerbernez) was also a well-drained sandy loam soil of 0.8 m depth, with 4.5% organic matter content and a pH of 5.8. Two treatments were established: clover–grass receiving no N fertilization (KZ-CG0N) and pure ryegrass receiving 250 kg N ha<sup>-1</sup> year<sup>-1</sup> (KZ250N).

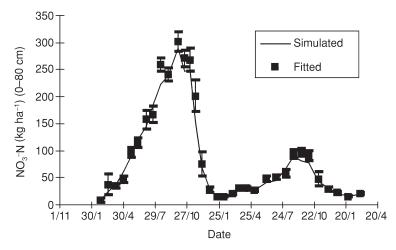
At both sites, grasslands were grazed for 6 years by heifers or young beef cattle. During that period, growth and N accumulated by grass were measured as well as nitrate leaching (Vertès *et al.*, 1997; Laurent *et al.*, 1999). The N balance established over the 6 years varied from –200 (KL0N) to +1200 kg N ha<sup>-1</sup> (KL400N). The grasslands were destroyed in mid-February 1997 by glyphosate application. The amount and biochemical characteristics of grass were determined just before destruction on all sites.

After destruction, the soil was kept bare (using herbicides) in all treatments for 2 years (1997–1999). Soil cores were taken by auger sampling every 3 weeks up to 0.8 m depth (in three layers). Water and mineral N (NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>) contents were determined in all soil samples. These measurements were used in the LIXIM model (Mary *et al.*, 1999) in order to calculate water (evaporation and drainage) and nitrogen fluxes (mineralization and leaching). This model searches for the best fit between measured and simulated water and nitrate contents in each layer, and calculates mineralization and leaching rates. Lysimeters (Kerbernez) or porous cups (Kerlavic) were used to determine the amounts of drained water and nitrate leached.

An incubation experiment was conducted simultaneously (Laurent et al., 1998). Soils from each site and treatment were sampled (0–25 cm depth) just before grass destruction. Aerial parts and roots were separated from soils, cut into 2–5 mm fragments and added to fresh sieved soil (sieved on 5-mm mesh). Grass residues were added in amounts similar to field conditions. Non-treated soils were also incubated. Incubation was performed at 15°C and at constant soil water content (80–90% field capacity) for 1.5 years. Mineralized carbon was monitored continuously by CO<sub>2</sub> trapping. Mineralized nitrogen was determined at regular intervals on soil samples.

#### In situ N dynamics and nitrate leaching

Just before destruction, grass biomass was similar in both sites, ~10 t DM ha<sup>-1</sup> for pure ryegrass and 6.5 t DM ha<sup>-1</sup> for clover–grass, corresponding to 140–190 kg N ha<sup>-1</sup> and 110–130 kg N ha<sup>-1</sup> accumulated in grass residues, respectively. The C: N ratio of the residues was higher in the unfertilized treatment (C: N = 30) than in the fertilized grasslands (C: N = 18–24).



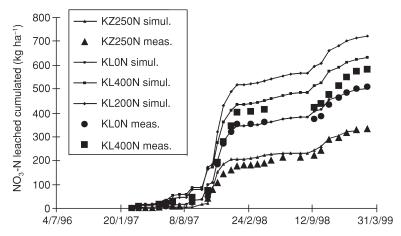
**Fig. 4.2.1.** Changes in mineral nitrogen in soil (0–80 cm) during 2 years at Kerlavic (treatment 0N): measured values (soil cores) are compared with simulated data (LIXIM).

Inorganic N in soils showed clear differences between the two sites: the maximum soil mineral N (observed in late summer) was 300–380 kg N ha<sup>-1</sup> at Kerlavic and 120–150 kg N ha<sup>-1</sup> at Kerbernez. Half of the mineralized N occurred within 3 months after glyphosate application, and would have been available for a growing crop in late May. The LIXIM model was able to reproduce the evolution of mineral N in the whole soil profile (Fig. 4.2.1). The kinetics of water drainage simulated by the model were also in good agreement with the measured kinetics, on both sites (data not shown), which suggested that the model was able to predict N mineralization and leaching well.

The leaching losses, calculated and measured from the time of grass destruction to the end of the second winter, are reported in Fig. 4.2.2 for pure grass treatments. For a similar annual drainage (700–800 mm), the amount of leached N during the first winter was about twice that during the second winter. N leached at the Kerlavic site during the first year (360–420 kg N ha<sup>-1</sup>) was much higher for all treatments than N leached at the Kerbernez site (170–180 kg N ha<sup>-1</sup>). At the first site, a slight significant difference was observed only between the non-fertilized treatment (KL0N) and the three others. No statistical test can be easily used to test model results (Mary *et al.*, 1999).

The LIXIM model gave slightly higher estimates of leaching losses than direct measurements did during the first year (Table 4.2.1) but total leached N for the 2 years was estimated accurately by the model for most treatments (Fig. 4.2.3).

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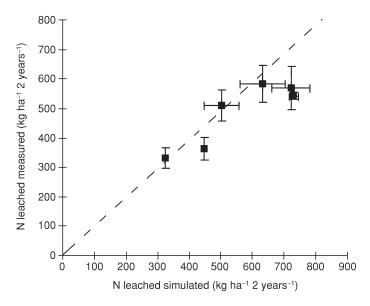
**Fig. 4.2.2.** Leached N (solid lines) measured with porous cups (KL) or lysimeters (KZ) and simulated by the LIXIM model in fallow soil, after grass destruction, for six treatments.

**Table 4.2.1.** Nitrate N (kg ha<sup>-1</sup>) leached under bare soil during the two winters following grass destruction. The 'measured' values are derived from lysimeters in the KZ site or porous cups in the KL site: mean (sp). The 'calculated' values are LIXIM outputs.

	199	7/98	199	8/99
	Calculated	Calculated Measured		Measured
KL PG-0N	348	355 (64)	154	155 (54)
KL PG-200N	522	380 (1)	200	162 (13)
KL PG-400N	439	405 (48)	192	180 (60)
KL GC-50N	539	420 (51)	190	150 (70)
KZ PG-250N	209	181 (23)	115	151 (17)
KZ GC	310	191 (31)	135	172 (15)

#### N mineralization rates in situ and under controlled conditions

The kinetics of N mineralization calculated by the model are shown at Fig. 4.2.4. N mineralization rates ranged from 1.1 (KZ) to 2.4 (KL) kg N ha<sup>-1</sup> nday<sup>-1</sup> during the period 0–100 (KZ) or 0–200 (KL) normalized days (1 nday = 1 day at 15°C and optimal water content; Mary *et al.*, 1999). In laboratory incubations, initial nitrogen mineralization rates were 1.8–2.0 kg N ha<sup>-1</sup> day<sup>-1</sup>; lower rates of 1.4 kg N ha<sup>-1</sup> day<sup>-1</sup> were measured for the 0N treatment (KL site). N mineralization in control soils (data not shown) was 85% of the mineralization rate observed in treated soils. This suggests that soils which were not treated with fresh organic residues contained large amounts of easily mineralizable OM. Net N immobilization did not occur,



**Fig. 4.2.3.** Comparison of measured and simulated leached N (total for 2 years after destruction).

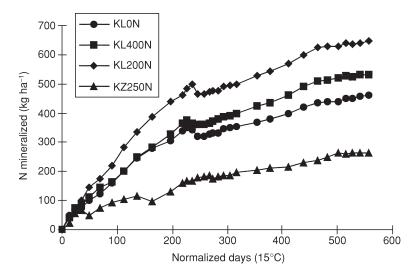


Fig. 4.2.4. Mineralization kinetics simulated by LIXIM for KL treatments.

due to the low C: N ratio of the added residues. No significant differences were observed between sites. After  $\sim 100$  days of incubation, a second phase occurred with lower mineralization rates of  $\sim 0.5$  kg N ha<sup>-1</sup> day<sup>-1</sup>. This lower mineralization rate is assumed to correspond to the 'basal'

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**Table 4.2.2.** N mineralization rate *in situ* (LIXIM calculation) and under controlled conditions (incubations) expressed as kg N ha<sup>-1</sup> nday<sup>-1</sup> (nday = normalised day for residues in treated soils kept bare. The first period lasts 202 and 100 normalized days for the KL and KZ sites, respectively; then the second period takes place until the end of the observations (March 1999).

	Vp 1s	st period	Vp 2nd period		
	LIXIM	Incubation	LIXIM	Incubation	
KL PG-0N	1.61	1.41	0.46	0.55	
KL PG-200N	2.27	1.92	0.63		
KL PG-400N	1.73	1.89	0.62		
KL G/WC	2.21	2.0	0.61		
KZ PG-250N	1.06	1.85	0.45	0.66	
KZ G/WC	1.11	1.86			

mineralization of soil OM, while the high rates observed during the first 100 days would correspond to the decomposition of the easily decomposable OM accumulated during the grass period (8 years). This hypothesis is supported by the kinetics of C mineralization.

Laboratory incubation and *in situ* estimates of net mineralization gave a similar range of net mineralization rates (using the normalized days as a common unit, Table 4.2.2). However, differences between sites did not appear in controlled conditions, and the period of intense mineralization lasted longer in field conditions than under controlled conditions (180 versus 100 normalized days). The differences observed might result from differences in decomposition conditions (Mary *et al.*, 1996). Soil–residue contact was favoured in laboratory incubation by cutting residues and mixing them with sieved soil, whereas *in situ* decomposition occurred at the soil surface. Laboratory incubation conditions may have stimulated decomposition compared with field conditions, as has already been observed for rapeseed residues (Dejoux *et al.*, 2000; Trinsoutrot *et al.*, 2000).

#### Discussion and Conclusion

The study confirms that a high rate of N release follows grassland destruction, even without soil tillage: ~2 kg N ha<sup>-1</sup> normalized day<sup>-1</sup> in bare soil during a first period corresponding to the decomposition of fresh residues. The rate was almost independent of the previous grassland management. Thus no relationship was observed between N balance during the grazing period and N mineralization after destruction. Unlike the results of Hogh-Jensen (1996), no significant difference was observed between mineralization of pure grass versus grass-clover, but incorporated N and C

were lower in grass-clover swards: 6.5 t DM ha<sup>-1</sup> versus 10–11 t ha<sup>-1</sup>, with a similar C: N ratio (~20). This low C: N ratio explains the lack of nitrogen immobilization observed under controlled conditions. Moreover, although total mineralization was not higher in grass-clover treatments, the percentage of nitrogen mineralized from residues appeared higher for grass-clover plants (results not shown), consistent with the findings of Hauggaard-Nielsen *et al.* (1998). To confirm and better understand these results, a new experiment is in progress, which aims at separating soil from plant residue effects.

After decomposition of fresh residues, which lasted ~100 days under controlled conditions, mineralization greatly decreased (0.5 kg N ha<sup>-1</sup> day<sup>-1</sup>), and remained stable for > 1.5 year after grass destruction. This 'basal' level is still high, and may be related to the high organic matter content of the soils studied. This high mineralization rate induced a high potential for nitrate leaching: for the 2 years, leaching under bare soils varied between 320 and 600 kg N ha<sup>-1</sup>. Such a leaching risk must be taken into account when selecting production systems: in livestock systems, the proportion of grasslands (and among them mixed stands) is increasing, but most of them are destroyed regularly. After a late winter destruction, few summer crops are able to use such large amounts of mineral nitrogen, except fodder beet or maize + undersown Italian ryegrass, which are both able to recover 300–350 kg N ha<sup>-1</sup>. They should be considered as suitable crops in fodder systems, but are not often cultivated (and not encouraged by current CAP policy).

#### **Acknowledgements**

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# Dissolved Organic Nitrogen in a Peaty Podzol: Influence of Temperature and Vegetation Cover

4.3

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#### Introduction

In most soils, > 90% of the nitrogen is present in organic forms, and recent studies have shown that dissolved organic nitrogen (DON) compounds are often the predominant form of N in soil solutions and leachates (e.g. Qualls and Haines, 1991). Relatively little information, however, is available on either the origin or processes controlling DON in soil water. One hypothesis is that DON serves as an intermediate N pool between inorganic N and 'insoluble' organic N fractions of the soil during mineralization of organic matter and immobilization of inorganic N, respectively. However, as observed for dissolved organic carbon (DOC), the availability and mobility of DON within the soil are likely to be controlled by sorption processes. A major source of dissolved organic matter (DOM) to the soil is the decomposition of plant material and the release of soluble organic compounds, such as DON. Another source of DON is direct release from plant roots into the rhizosphere. The detection of free amino acids in root exudates was reported many years ago (Smith, 1969), but the dynamics of this N were not followed mainly because of problems associated with extraction and analysis.

The objective of this study was to determine the influence of temperature and vegetation cover on DON in a peaty podzol. Firstly, we followed the temporal changes in extractable DON and mineral N in soil cores incubated at 5, 10 and 15°C, to determine whether DON was depleted when net mineralization of N occurred. Secondly, intact cores of the same soil, with and without vegetation, were leached with artificial rain for

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6 weeks at 6.5 and 15°C, to examine whether vegetation cover was a major source of DON in soil leachate.

#### **Materials and Methods**

#### Site description

Soil samples were collected from Glensaugh in NE Scotland. The soil is a peaty podzol of the Gaerlie series (Stagni-Placic Haplic Podzol: FAO/UNESCO, 1994) which is underlain by quartz-mica schists of Dalradian age. Associated with this soil type is acid moorland vegetation dominated by Calluna vulgaris L. and Vaccinium myrtillus interspersed with Deschampsia flexuosa, Nardus stricta, Vaccinium vitis-idea and Erica cineria and which is grazed by sheep. In our study, the vegetation present on the soil cores was dominated by the moss Rhytidiadephus squarrosus and the grasses Agrostis capillaris and Anthoxanthum odoratum. The presence of the latter indicated that the area had been improved agriculturally in the past.

#### Soil incubation experiment

Forty-five soil cores (diameter 7.8 cm, length 10 cm) were sampled at random locations, within an area of 400 m², using PVC tubes. In the laboratory, the soil cores were removed from the tubes and the vegetation and surface litter separated from each core. The cores were trimmed, from the base, to a length of 6 cm and weighed. Fifteen cores were placed inside each of three incubators set at 5, 10 and 15°C. Three replicate cores were harvested at each temperature after incubation for 0, 7, 14, 28 and 56 days. The soils were sieved (< 2 mm) to remove stones and roots and subsampled for pH and water and 0.5 M K<sub>2</sub>SO<sub>4</sub>-extractable inorganic and organic N determinations. Soil moisture content was determined on day 0. The dried soil was then milled and analysed for total C and N.

#### Soil leachate experiment

Twelve intact soil cores were sampled by inserting hollow PVC tubes (diameter 15 cm, length 10 cm) into the soil at random locations. At six sites, the surface vegetation and root mat (2–3 cm thick) were removed prior to inserting the PVC tubes. In the laboratory, three cores with and three cores without vegetation were placed inside each of two controlled environment cabinets set at 6.5 and 15°C, a relative humidity of 65% and a day length of 12 h. After 48 h, the soil cores were leached with artificial rain

having a chemical composition based upon the mean annual composition of weekly bulked precipitation samples collected in the area (Chapman *et al.*, 1997). The NH<sub>4</sub><sup>+</sup> concentration was 0.55 mg N l<sup>-1</sup> and the NO<sub>3</sub><sup>-</sup> concentration was 0.33 mg N l<sup>-1</sup>. Each core received 400 cm<sup>3</sup> of artificial rain twice a week for 6 weeks. The leachate from each core was collected 24 h after the rain was applied and analysed for pH, N fractions and DOC.

#### Methods of extraction and chemical analysis

Ammonium,  $NO_3^-$  and DON were extracted from the soil by shaking 10 g fresh weight of soil for 2 h with 50 ml of distilled water or 50 ml of 0.5 M  $K_2SO_4$ . The suspensions were first filtered through Whatman No. 42 paper before passing through 0.45 µm membrane filters. The soil acidity was determined by measuring pH in suspension of fresh soil in 0.01 M  $CaCl_2$  at a sample : solution ratio of 1 : 10 (w/v). Total dissolved N (TDN) in the water and  $K_2SO_4$  extracts and soil leachates was measured as  $NO_3^-$  after oxidation with alkaline potassium persulphate (Williams *et al.*, 1995). Ammonium and  $NO_3^-$  were measured using standard colorimetric methods. DON was calculated by difference: DON = TDN – ( $NH_4^+ + NO_3^-$ ). Concentrations of DOC were determined using a soluble carbon analyser (OI Analytical Model 700).

#### Statistical analyses

Analyses of variance to compare treatments, temperatures and depth were carried out using the software Genstat (Genstat 5, 1993). A repeated measures technique was used to test for treatment effects in the leachate experiment. Skewed data were transformed logarithmically for analysis of variance, and the arithmetic means of the untransformed data are presented.

#### Results

#### Soil characteristics

The soil used in both experiments contained on average 278 mg C kg<sup>-1</sup> ( $\pm$  11) and 16.3 mg N kg<sup>-1</sup> ( $\pm$  1.2), and the mean C : N ratio was 17.4. At sampling, the mean moisture content was 44.42% ( $\pm$  0.78) of fresh weight and the average pH (CaCl<sub>2</sub>) was 4.04 ( $\pm$  0.04).

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#### Soil incubation experiment

Net mineralization occurred at all temperatures, with larger rates observed at higher temperatures (Fig. 4.3.1a); the average daily rates were 6.7, 25.3 and 62.7 mg N m<sup>-2</sup> day<sup>-1</sup> at 5, 10 and 15°C, respectively. In contrast, water-extractable DON displayed no clear trend over the incubation period, although the amount extracted on day 28 was significantly (P < 0.001) smaller than at other times, and showed little response to temperature (Fig. 4.3.1b). Additional DON, equivalent to 2 g N m<sup>-2</sup>, extracted with 0.5 M K<sub>2</sub>SO<sub>4</sub>, also showed little sensitivity to changes in time or temperature.

#### Soil leachate experiment

Concentrations of NH<sub>4</sub><sup>+</sup> in leachates from all the soil cores were small (ranging typically between 0.25 and 0.5 mg N l<sup>-1</sup>) and varied little with time or between each other. Concentrations of NO<sub>3</sub><sup>-</sup> in leachates from the soil cores without vegetation steadily increased over the 42 days (Fig. 4.3.2a), and the rate of increase was greater for the cores at 15°C than at 6.5°C. In contrast, concentrations of NO<sub>3</sub><sup>-</sup> in leachates from the soil cores with vegetation initially increased before decreasing after day 10 at 15°C and after day 21 at 6.5°C (Fig. 4.3.2a). Concentrations, however, were consistently larger in leachates from the cores at 15°C than at 6.5°C. At 15°C, concentrations of NO<sub>3</sub><sup>-</sup> initially were larger in leachates from cores with vegetation than without, until after day 12 when concentrations of NO<sub>3</sub><sup>-</sup> were larger in leachates from cores without vegetation (Fig. 4.3.2a). At 6.5°C, there was little difference in the concentration of NO<sub>3</sub><sup>-</sup> between cores with or without vegetation until after day 21, when concentrations

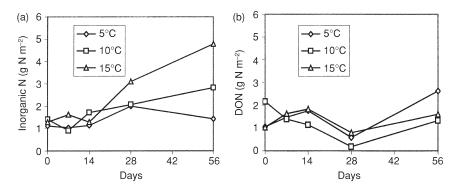
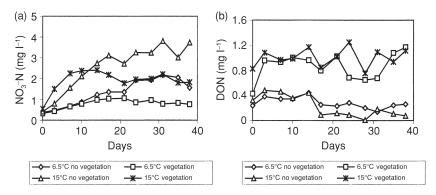


Fig. 4.3.1. Changes in (a)  $0.5 \text{ M K}_2\text{SO}_4$ -extractable inorganic N and (b) water-extractable DON during incubation of a peaty podzol at 5, 10 and 15°C.



**Fig. 4.3.2.** Concentrations of (a) NO<sub>3</sub><sup>-</sup> and (b) DON in leachates collected from soil cores.

were larger in leachates from the cores without vegetation. Analysis of variance using a repeated measures technique indicated that temperature and the presence or absence of vegetation interacted to have a significant (P < 0.01) effect on the concentration of  $NO_3^-$  in soil leachate at all time points.

Concentrations of DON and DOC were significantly (P < 0.001) larger in leachates from cores with than without vegetation at all times, whereas temperature had little effect on either DON or DOC concentrations (Fig. 4.3.2b). On average, DON concentrations in leachates from vegetated cores were over three times larger at  $6.5^{\circ}$ C and almost five times larger at  $15^{\circ}$ C, than those from cores without vegetation. In contrast, DOC concentrations were twice as high in leachates from vegetated cores at both temperatures. The average C: N ratio in leachates was significantly (P < 0.01) greater from cores without vegetation (mean = 27) than from cores with vegetation (mean = 19).

Over the 42-day leaching period, all the cores received 149 mg N m<sup>-2</sup> as NH<sub>4</sub><sup>+</sup> and 89.5 mg N m<sup>-2</sup> as NO<sub>3</sub><sup>-</sup> from the artificial rain. The rain contained no DON. The flux of NH<sub>4</sub><sup>+</sup> from all the cores was very small (Table 4.3.1), whereas all the cores leached more NO<sub>3</sub><sup>-</sup> than they received in the rain (Table 4.3.1). However, when the total inorganic N input and output fluxes are compared, it can be seen that at 6.5°C the cores with vegetation received more inorganic N from the rain than they leached, while the cores without vegetation leached 62.5 mg N m<sup>-2</sup> more inorganic N than they received (Table 4.3.1). At 15°C, both the cores with and without vegetation leached a greater quantity of inorganic N, predominantly as NO<sub>3</sub><sup>-</sup>, than they received, although a larger flux was observed from the cores without vegetation (Table 4.3.1). The difference in amount of inorganic N leached from the cores with and without vegetation probably represents that taken up by the vegetation, which accounted for 92.8 and 127 mg N m<sup>-2</sup> at 6.5 and 15°C, respectively (Table 4.3.1).

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	Input	in rain	Outp	out in lead	chate	Net ret	ention/lo	ss of N	
Cores ± vegetation <sup>1</sup>	NH <sub>4</sub> <sup>+</sup>	NO <sub>3</sub> <sup>-</sup>	NH <sub>4</sub> <sup>+</sup>	NO <sub>3</sub> <sup>-</sup>	DON	NH <sub>4</sub> <sup>+</sup>	NO <sub>3</sub> <sup>-</sup>	Min. N <sup>2</sup>	Net TDN leached <sup>3</sup>
6.5°C+	149	90	10 <sup>a</sup>	198 <sup>a</sup>	237ª	-139ª	108ª	-31ª	206ª
6.5°C-	149	90	11 <sup>a</sup>	290a	63 <sup>b</sup>	$-138^{a}$	200a	62 <sup>b</sup>	125 <sup>a</sup>
15°C+	149	90	17 <sup>a</sup>	419 <sup>b</sup>	230a	-132a	329 <sup>b</sup>	197°	427 <sup>b</sup>
15°C-	149	90	8ª	555 <sup>b</sup>	52 <sup>b</sup>	-141 <sup>a</sup>	465 <sup>b</sup>	324 <sup>d</sup>	376 <sup>b</sup>

**Table 4.3.1.** Nitrogen input–output budget (mg N m<sup>-2</sup>) for the leached soil cores.

Means within the same column followed by a common letter are not significantly different (P < 0.05).

The DON flux from the soil cores displayed a contrasting pattern to the inorganic N flux. The DON flux was approximately four times larger from the cores with than without vegetation, and temperature appeared to have no influence upon the flux (Table 4.3.1). Thus, although the net inorganic N flux was larger from the cores without vegetation, the net TDN flux was larger from the cores with vegetation at both temperatures. In addition, the amount of DON, and DOC, leached from the cores with vegetation was similar regardless of whether net N immobilization or net N mineralization was occurring within the soil (Table 4.3.1).

#### Discussion

#### **Origin of DON**

In the incubation experiment, the relative sizes of the DON and mineral N pools initially were very similar, yet during incubation net mineralization was observed at all temperatures, whereas the DON pool size varied little over the 8-week incubation experiment and showed little response to temperature. Thus, there was no apparent accumulation or depletion of DON over the course of the incubation study. Other studies have found that DON, extracted with 0.01 M CaCl<sub>2</sub>, remained relatively constant during incubation while mineral N increased linearly (e.g. Appel and Mengel, 1990). For this to occur, the DON pool may be maintained by and in equilibrium with a large reserve of organic N. The rate of transfer between these pools would need to be comparable with the sum of the rates of

 $<sup>^{1}+</sup>$  = with vegetation and - = no vegetation.

 $<sup>^{2}</sup>$ Min = mineral N (NH<sub>4</sub><sup>+</sup> + NO<sub>3</sub><sup>-</sup>).

<sup>&</sup>lt;sup>3</sup>TDN = total dissolved nitrogen.

ammonification, nitrification and assimilation into the microbial biomass. The location of such a reserve of organic N is not clear, but it could occupy an adsorbed phase on surfaces in the soil matrix, which can be released partly by salt solutions (Reemtsma *et al.*, 1999). This would be consistent with the greater amounts of DON released from soil by 0.5 M K<sub>2</sub>SO<sub>4</sub> compared with water.

In the soil leachate experiment, concentrations of DON were significantly larger in leachates from cores with vegetation. The total amount of DON leached from the cores with vegetation was 0.23 g N m<sup>-2</sup>, which was four times greater than that leached from cores without vegetation and 20% less than the DON extracted with water from the soil cores in the incubation experiment. Thus, removing the vegetation cover and root mat eliminated an important source of DON. In the cores without vegetation, some root material remained, the decay of which would account for the larger concentration of DON leached over the first 14 days compared with subsequent days. The greater leaching from cores with vegetation suggests that much of the DON was leached from foliage and plant residues in the root mat and/or exuded from roots. In forest soils, it has been suggested that the major source of DON in soil water was water-soluble organic material leached from freshly shed leaf litter (e.g. Qualls and Haines, 1991). Plant roots are also known to release a complex mixture of organic compounds that include sugars, amino acids and organic acids into the rhizosphere as part of normal growth. If the DON in the leachates was derived predominantly from the plant roots and litter, then the water-extractable DON pool in the soil contributed very little to the DON in leachates.

#### Role of DON in N mineralization

One of the hypotheses regarding the DON pool is that it is an intermediate in the mineralization process and a source of readily mineralized organic N (Appel and Mengel, 1990). In sandy soils, DON extractable with 0.01 M CaCl<sub>2</sub> correlated positively with plant uptake and soil inorganic N (Appel and Mengel, 1993), possibly because the sandy soils contained < 50 mg C kg<sup>-1</sup> dry soil. The podzol studied here contained 278 mg C kg<sup>-1</sup> dry soil and the relationship between the readily mineralized N pool and the mineral N pool possibly was obscured by the quantity of DON. Thus the DON pool may contain a smaller active fraction that is turning over rapidly. For example, microbial metabolites, such as amino acids derived from the large microbial N pool, have a rapid turnover rate in some soils (Jones, 1999). Alternatively, the organic N mineralized may have been replaced by DON desorbed from a larger pool of adsorbed organic matter.

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#### Contribution of DON to N in surface waters

Concentrations of DON in the soil leachate ranged from < 0.01 to 1.8 mg N l<sup>-1</sup>, with larger concentrations in leachates from the core with vegetation (mean =  $0.93 \text{ mg N } l^{-1}$ ) than without vegetation (mean =  $0.26 \text{ mg N } l^{-1}$ ). These are generally larger than concentrations of DON observed in drainage waters from peatland in northern Scotland, which varied with season and were largest, up to  $0.56 \text{ mg N l}^{-1}$ , in the autumn (Anderson et al., 1991). In a study of upland streams in the UK, concentrations of DON ranged between < 0.01 and 0.78 mg N l<sup>-1</sup>, although the majority of stream samples (80%) contained between 0.05 and 0.325 mg N l<sup>-1</sup> (Chapman et al., 1998). Chapman et al. (1998) also reported that DON contributed to between 14 and 69% of TDN in upland streams, with the proportion increasing in upland areas dominated by peat soils. The lower concentrations of DON in stream waters compared with soil leachates may be explained by a combination of factors such as: (i) dilution with different source waters of a lower DON content; and/or (ii) mineralization of DON within the stream; and/or (iii) uptake of DON by stream biota.

#### **Acknowledgements**

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# Dissolved Organic Nutrients (N, P and S) in Shallow Forest Soils: Fluxes and Spectroscopic Characterization

4.4

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#### Introduction

Dissolved organic matter (DOM) contains important nutrients such as N, P and S. There is little vertical loss of the organic nutrients from deeply developed forest soils under cool and temperate climates due to effective retention of DOM in the sub-soil (McDowell and Likens, 1988; Qualls and Haines, 1991). In contrast to deeply developed soils, the lower horizons of shallow soils sorb relatively little DOM (Kaiser *et al.*, 1996). Thus, shallow forest soils might release large amounts of dissolved organic carbon (DOC) and associated dissolved organic nutrients such as N, P and S (DON, DOP and DOS) into aquatic environments. We tested this hypothesis in a field experiment.

#### Materials and Methods

#### **Experimental site**

The experimental site was located in NE Bavaria, Germany. The site was on a flat hilltop covered with a 90-year-old European beech (*Fagus sylvatica* L.) forest. The soils were Lithic Rendolls derived from Upper Kimmeridgian dolomite. The basic properties of the soils are summarized in Table 4.4.1. Due to the loamy texture and the large organic carbon (OC) content, the soils were strongly aggregated. In addition, there were flow channels throughout the whole profile resulting from preferential

Horizon	Depth (cm)	pH CaCl <sub>2</sub>	CEC <sup>a</sup> (mmol <sub>c</sub> kg <sup>-1</sup> )	CO <sub>3</sub> -C <sup>b</sup> (g kg <sup>-1</sup> )		Clay <sup>d</sup> (g kg <sup>-1</sup> )	O A	Fe <sub>Ox</sub> e (g kg <sup>-1</sup> )	Fe <sub>DCB</sub> <sup>f</sup> (g kg <sup>-1</sup> )
A1	0–10	7.1	422	4	112	230	5.3	3.6	8.4
A2	10-25	7.3	284	24	69	210	4.7	3.3	7.7
С	25–95	7.6	63	70	11	70	1.4	1.6	5.0

**Table 4.4.1.** Basic properties of the soils at the experimental site.

weathering along cracks in the bedrock. The organic forest floor layer was mull-type.

#### Soil solution sampling

At the experimental sites three plots were each equipped with four bulk precipitation and throughfall collectors, and with zero-tension (beneath the forest floor and at 90 cm depth) tension lysimeters (at 15 and 30 cm depth), suction cups (at 90 cm depth) and tensiometers (at 15, 30 and 90 cm depth). The tension at the tension lysimeters and the suction cups was regulated according to the soil water tension. Rain and soil water were collected at 7-day intervals during the growing season and at 14-day intervals during the dormant season. The samples were filtered through  $0.45\,\mu m$  membrane filters and stored at  $-18^{\circ}C$ . The results presented here refer to the sampling period from December 1997 to November 1998.

#### **Chemical analyses**

The water samples were fractionated into hydrophilic and hydrophobic compounds using the XAD-8 resin method (Aiken and Leenheer, 1993). The hydrophobic fraction is dominated by lignin-derived compounds whereas the hydrophilic fraction represents mainly carbohydrates of both plant and microbial origin (Guggenberger *et al.*, 1994). The original samples and the hydrophilic fractions were analysed with a Shimadzu

 $<sup>^{\</sup>mathrm{a}}$ Cation exchange capacity, measured with 1 M NH $_{4}^{+}$  acetate at pH 7.0 (Avery and Bascomb, 1974).

<sup>&</sup>lt;sup>b</sup>Carbonate carbon, measured with a calcimeter according to Scheibler (Schlichting and Blume, 1966).

Organic carbon, calculated by the difference between total carbon measured with a CHNS analyser (Vario EL, Elementar Analysensysteme GmbH, Hanau, Germany) and carbonate carbon.

dEstimated by using the sieve-pipette method (Avery and Bascomb, 1974).

eOxalate-extractable Al and Fe (Schwertmann, 1964).

<sup>&</sup>lt;sup>f</sup>Dithionite-citrate-bicarbonate-extractable Fe (Mehra and Jackson, 1960).

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TOC-5050 analyser for DOC and dissolved inorganic C, with an Abimed TN-05 for total N, with ICP-OES (GBC Integra XMP) for total P and S, and with ion chromatography (Dionex DX-100) for NH<sub>4</sub><sup>+</sup>, NO<sub>3</sub><sup>-</sup>, NO<sub>2</sub><sup>-</sup>, H<sub>2</sub>PO<sub>4</sub><sup>-</sup> and SO<sub>4</sub><sup>2-</sup>. DOC, DON, DOP and DOS were calculated by the difference between the total amounts and the sum of the inorganic forms. Elements in the hydrophobic fraction were calculated by the difference between the amounts in the original sample and the hydrophobic fraction.

Sub-samples of soil water collected over each quarter of the year were pooled. The samples were treated with an acidic cation exchange resin (BioRad AG MP-50) to remove all cations other than H<sup>+</sup>, and freeze dried. Chemical structures of C and P were estimated by liquid-state <sup>13</sup>C- and <sup>31</sup>P-nuclear magnetic resonance (NMR) spectroscopy. For analytical conditions and designation of signals, see Kaiser *et al.* (1997) and Sumann *et al.* (1998).

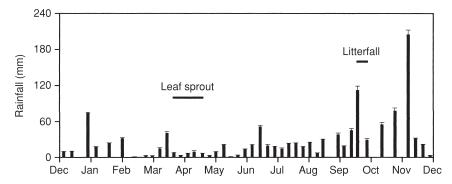
#### Calculation of fluxes

We calculated fluxes of the DOM released from the forest floor into the mineral soil from the amounts of seepage water sampled using zero-tension lysimeters beneath the forest floor layer and concentrations therein. Fluxes in the mineral soil were calculated from porewater concentrations and the simulated water fluxes. Water flux simulation was carried out using a water transport model (WHNSIM; Huwe, 1992). Input variables were meteorological data of the site and the water content—tension relationship of the soils. Validation of the model was achieved to measured soil water tensions. Seepage water in sub-soil at rapid flow conditions was sampled using zero-tension lysimeters. Fluxes were calculated from the amount of seepage water and concentrations therein.

#### Results and Discussion

The sampling period was characterized by unusually high rainfall during autumn 1998 compared with former years. Bulk precipitation peaked at the beginning of November 1998 with maximum of 204 mm in 10 days (Fig. 4.4.1). The rainfall during September to November 1998 comprised 50% of the annual precipitation.

The forest canopy was a minor source of DOM. Exceptions were the times of leaf sprout and before leaf fall (results not shown here). The major source of DOC, DON, DOP and DOS was the forest floor (Table 4.4.2). Organic nutrient forms represented up to 85% of the total nutrient content of the forest floor seepage water and were mainly in the hydrophilic DOM fraction. This result accords well with the observation of Qualls and Haines



**Fig. 4.4.1.** Bulk precipitation at the experimental site during the period December 1997–November 1998.

**Table 4.4.2.** Mean annual concentrations of dissolved organic carbon, nitrogen, phosphorus and sulphur (DOC, DON, DOP and DOS) in bulk precipitation (BP), canopy throughfall (TF), forest floor leachate (FF) and mineral soil porewater at 15, 30 and 90 cm depth in a European beech (*Fagus sylvatica* L.) forest with Lithic Rendolls during the period December 1997–November 1998. In addition, the mean proportions of organic carbon and nutrients in the hydrophilic fraction (hiDOC, hiDOP and hiDOS) are given. Values in parentheses indicate the standard deviation.

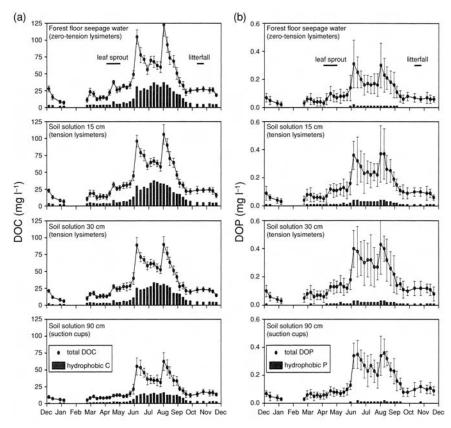
Compartment	DOC (mg l <sup>-1</sup> )	hiDOC (%)	DON (mg l <sup>-1</sup> )	hiDON (%)	DOP (mg l <sup>-1</sup> )	hiDOP (%)	DOS (mg l <sup>-1</sup> )	hiDOS (%)
BP	3.73 (3.22)	nd	0.12 (0.05)	nd	0.01 (0.01)	nd	0.04 (0.02)	nd
TF	12.05 (10.19)	77	0.42 (0.11)	86	0.03 (0.02)	94	0.12 (0.06)	70
FF	30.26 (24.57)	68	1.32 (0.22)	80	0.09 (0.07)	97	0.31 (0.09)	92
15 cm	28.50 (23.33)	67	1.31 (0.24)	82	0.11 (0.09)	88	0.34 (0.11)	86
30 cm	25.69 (20.75)	67	1.30 (0.21)	83	0.13 (0.10)	92	0.34 (0.10)	89
90 cm	16.70 (14.04)	73	0.98 (0.20)	94	0.11 (0.09)	96	0.29 (0.10)	91

nd = Not determined.

(1991). The concentrations of DOC and organic nutrient forms were significantly (P < 0.001) higher during the growing season (April–September 1998) than during winter 1997 and autumn 1998 (e.g. DOC and DOP; Fig. 4.4.2).

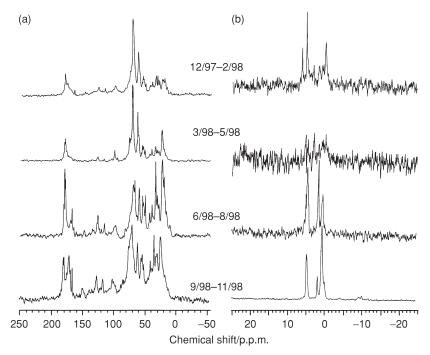
The concentrations in the forest floor seepage water and the mineral soil porewater peaked at heavy rain events during the first warmer weeks in spring and after rain storms following drier periods in summer. In autumn 1998, the concentrations reached a more or less constant level clearly below the concentrations during spring and summer. One reason for the constant concentrations could be the large amount of rainfall during that time resulting in permanent leaching of the forest floor material.

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**Fig. 4.4.2.** Dissolved organic carbon (DOC) and phosphorus (DOP) concentrations in forest floor seepage water and mineral soil porewater of Lithic Rendolls covered with a European beech (*Fagus sylvatica* L.) forest during the period December 1997–November 1998. The error bars indicate the standard deviation of at least three samples.

Larger proportions of lignin-derived hydrophobic DOC (Table 4.4.2) and of signals of alkyl C (0–50 p.p.m.) and aromatic C (110–160 p.p.m.) in the <sup>13</sup>C-NMR spectra of DOM in the forest floor seepage water in summer and autumn indicated enhanced decomposition (Fig. 4.4.2a). The DOC in winter and spring was concentrated in the hydrophilic fraction (Fig. 4.4.2a) and consisted mainly of O-alkyl C (50–110 p.p.m.) representing carbohydrates. We assume that DOM occurring during the winter and spring is due to leaching of soluble material from fresh beech litter and microbial debris. The <sup>31</sup>P-NMR spectra of forest floor leachates (Fig. 4.4.3b) in winter indicated that inorganic P was mainly as orthophosphate (~6.2 p.p.m.) as there was little orthophosphate monoester P (4.1–5.6 p.p.m.). The P concentrations in spring were too low for recording spectra. During summer and autumn 1998, signals of organic P forms dominated the spectra.



**Fig. 4.4.3.** Liquid-state <sup>13</sup>C- and <sup>31</sup>P-NMR spectra of organic carbon (a) and phosphorus (b) in forest floor seepage waters of Lithic Rendolls covered with an European beech (*Fagus sylvatica* L.) forest in winter 1997/98 and spring, summer and autumn 1998.

Monoester and sugar diester P (0.0–3.0 p.p.m.) again indicated release of DOM due to enhanced decomposition. Sugar diesters, e.g. teichoic acids, are assumed to originate from microbial biomass (Condron *et al.*, 1990), and so their large signals, especially in the summer sample, underline the microbial contribution to the DOM.

Retention of DOC and dissolved organic nutrients in the upper mineral horizons was limited (Table 4.4.2). Neither the concentrations of DOC nor those of dissolved organic nutrients in the soil solution decreased. Instead, the concentrations of DOP tended to increase during the passage of the topsoil. During the further passage through the sub-soil, the total DOC concentration decreased by ~45%; that of hydrophobic DOC by 53%. Compared with total DOC, all nutrients in organic forms were leached preferentially from soil, especially DOP and DOS. The enhanced mobility of dissolved organic nutrients could be due to their enrichment in the mobile hydrophilic DOM fraction (e.g. Qualls and Haines, 1991; Anderson et al., 1999).

Heavy rain events in autumn 1998 (September 11–16, October 24–November 6) induced rapid flow through macropores. As a result, the

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**Table 4.4.3.** Fluxes of dissolved organic carbon, nitrogen, phosphorus and sulphur (DOC, DON, DOP and DOS) entering the mineral soil (forest floor seepage water) and in the sub-soil (seepage water at 90 cm depth) of Lithic Rendolls covered with a European beech (*Fagus sylvatica* L.) forest during two heavy rain events in autumn 1998.

Compartment	Period	Water budget (mm)	DOC (g m <sup>-2</sup> )	DON (g m <sup>-2</sup> )	DOP (g m <sup>-2</sup> )	DOS (g m <sup>-2</sup> )
Seepage water	September 6–11 October 24–November 6 September 6–11 October 24–November 6	73	1.81 4.71 1.60 4.06	0.095 0.203 0.080 0.180	0.005 0.013 0.005 0.012	0.021 0.040 0.018 0.034

water budget measured with zero-tension lysimeters directly beneath the forest floor layer and at 90 cm depth were nearly equal (Table 4.4.3). Also the concentrations of DOC and dissolved organic nutrients in forest floor and sub-soil seepage water were similar, while the concentrations in porewater (sampled with suction cups) were reduced by 29–48% compared with the forest floor seepage water. Water budgets calculated with WHNSIM were ~65% of those measured with zero-tension lysimeters. Thus, calculation of fluxes from porewater data underestimated the fluxes by 56–70% during heavy rain events. The measured sub-soil fluxes of DOC during the two rain events exceeded the annual fluxes estimated for sites with developed soils (Guggenberger *et al.*, 1994).

In summary, shallow forest soils released large amounts of DOC and nutrients from the forest floor and the upper mineral soil horizons. In contrast to deeply developed soils, DOM was hardly retained in the sub-soils and, thus, organic nutrients were exported from the solum. Loss of DOM from the soil was greatest during rain storm events during the growing season and in autumn.

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### Dissolved Organic Carbon Losses from Grazed Grasslands

4.5

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#### Introduction

Soil organic matter (SOM) is a complex mixture of senescent residues of plants, animals and microorganisms through to highly humified material hundreds of years old. These varied materials are broken down simultaneously, but at different rates, with most of the products of degradation being more susceptible to dissolution in soil water than the original material. Dissolved organic matter (DOM) provides an important energy substrate for microbial communities to undertake transformation processes (e.g. for denitrification; Burford and Bremner, 1975) and also controls the availability of nutrients and trace metals to plants and microorganisms through formation of complexes (Stevenson, 1994). Environmental implications of DOM are associated with the transport of elements and compounds that are potential pollutants. For example, DOM has been shown to contribute to the mobility of some metals (e.g. Temminghoff et al., 1997), polycyclic aromatic hydrocarbons (e.g. Johnson and Amy, 1995) and nutrients such as N and P (Qualls and Haines, 1991). Additionally, knowledge of the potential fluxes of soluble organic C in different soils is essential to improve global C budgets (Han and Thompson, 1999).

Several studies have reported on DOM leaching from forest soils (e.g. McDowell and Likens, 1988) and on DOM in surface waters (see Thurman, 1985 for references). Hope *et al.* (1994) calculated that, for most catchments, the annual flux of organic C (OC) in streams varied from 10 to 100 kg C ha<sup>-1</sup> year<sup>-1</sup>. However, it is not known whether exports of OC from managed grasslands fall within this range and the extent of variation

with hydrological conditions and management. Here, we report on the amount and composition of DOC exported from grazed grasslands, and discuss factors that modify exports, such as fertilizer management and drainage regime.

#### Site Description

Most of the work reported here was carried out on the 1 ha lysimeter plots of the Rowden Moor Drainage Experiment (Tyson *et al.*, 1992) located at the Institute of Grassland and Environmental Research, North Wyke, Devon (NGR: SX 650995). The soil is a clayey non-calcareous pelostagnogley of the Hallsworth series (Dystic Gleysol, FAO) (see Armstrong and Garwood, 1991).

The experiment comprises 14 lysimeter plots of ~1 ha, grazed with steers. Seven plots have 55 cm deep mole drains crossing 85 cm deep permanent pipe drains, and are termed *drained*, while the remaining seven plots are termed *undrained*. All plots have V-notch weirs for the surface plus interflow to 30 cm depth that runs into perimeter ditches. The drained plots have additional weirs for water that is dispersed through the drainage system. A range of management strategies is applied to these plots, and details of the treatments reported here are shown in Table 4.5.1 together with treatment notation.

#### Quantities of DOC Exported from Grazed Grassland

In order to determine gross exports of DOC from the Rowden plots, grab samples were taken from the weirs during the first 2 months of drainage, i.e.

J					
	N application (kg	g N ha <sup>-1</sup> year <sup>-1</sup> )			Number of
Treatment	NH <sub>4</sub> NO <sub>3</sub>	Slurry <sup>a</sup>	Drainage	Notation	Treatments
High N	280	65	Undrained	HNU	2
Grass-clover	0	65	Undrained	GCU	2
Zero N	0	0	Undrained	ZNU	1
High N	280	65	Drained	HND	2
Grass-clover	0	65	Drained	GCD	2

Drained

ZND

1

**Table 4.5.1.** Drainage regime, N application and number of treatments of 1 ha lysimeters, together with treatment notation.

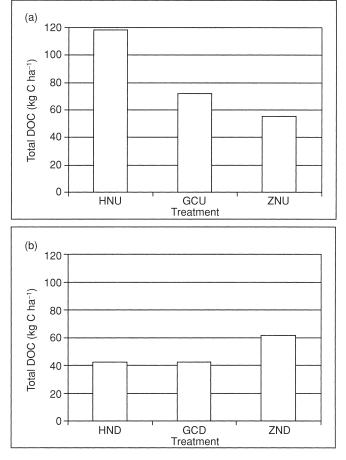
Zero N

<sup>&</sup>lt;sup>a</sup>Slurry application ceased 1 year before the start of the current study.

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from 18 November 1997 until 23 January 1998 when flow stopped due to a lack of rainfall. Samples were stored (2–4°C) until analysis, which was usually within 48 h. DOC was measured, after filtering (0.45  $\mu$ m), using a total OC analyser (model CA-10, Skalar Analytical B.V., De Breda, The Netherlands). Total losses of DOC were calculated as the sum of the products of weekly drainage volume and mean DOC concentrations.

The largest losses during this period were from the HNU plots, which exported ~120 kg C ha<sup>-1</sup> (Fig. 4.5.1a). This is greater than the range reported by Hope *et al.* (1994) for annual OC export (i.e. 10–100 kg C ha<sup>-1</sup> year<sup>-1</sup>), and which included both DOC and particulate OC. No animal manures were applied during the year preceding this work, although several



**Fig. 4.5.1.** Mean total DOC export from undrained (a) and drained (b) plots over 2 months. See Table 4.5.1 for notation.

of the plots had received slurry prior to that (Table 4.5.1). Losses of DOC would be expected to be greater when animal wastes are applied. Even with knowledge of the quantities of manures applied, it would be difficult to predict accurately the resulting increase in DOC export, as this would be governed in large part by the timing of application in relation to rain events.

To give an indication of amounts which may be expected following slurry application to heavy soils, an example is given from other work on the same soil type. Twenty-four 30 m<sup>2</sup> plots receive either nothing, slurry, fertilizer on a nutrient (N, P or K) equivalent basis, or a slurry-fertilizer mix. Slurry application rates are within limits recommended by the Ministry of Agriculture Fisheries and Foods (UK). The first application was after the first silage cut when local farmers were also applying slurry. Shortly after application, heavy rainfall resulted in the equivalent of 180 kg C ha<sup>-1</sup> being lost from the slurry-treated plots in only 8 days. This is almost twice as high as the upper end of the range reported by Hope et al. (1994) for annual losses and one and a half times the exports measured from the HNU plots of the Rowden experiment during the first 2 months of drainage. This was from a single slurry application and generally there are four or five applications during the year. There is therefore clear, but as yet unquantified, potential for large losses of C (and associated N and P) from intensively managed grasslands.

#### Factors Affecting DOC Export

As with all large-scale field experiments carried out with finite resources, a compromise had to be reached between the size of each plot, the number of treatments and the degree of replication. The small degree of replication made it desirable to bulk fertilizer rates within drained and undrained treatments to maximize the number of degrees of freedom, and, similarly, drainage treatments were bulked within 'fertilizer N rate'. Thus analysis of variance (SAS, 1996) could be performed on the quantity of DOC exported with 9 degrees of freedom, of which 1, 2 and 6 were allocated to drainage, fertilizer N rate and residual, respectively.

#### **Fertilizer**

An analysis of variance across the fertilizer treatments presented here (drained and undrained plots considered together) showed no significant effect of fertilizer N application rate on total DOC export over the 2-month period. However, for the undrained plots, there was a significant ( $r^2 = 0.78$ ; P = 0.048) positive correlation between fertilizer N application rate and total DOC export. As grassland dry matter productivity is proportional to

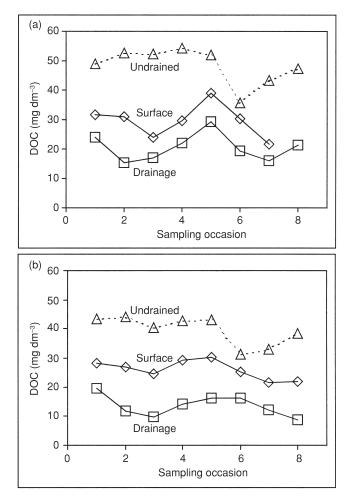
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N application (Morrison *et al.*, 1980), and increased productivity leads to greater returns of OM to the soil via faeces, and leaf and root death (Parsons *et al.*, 1991), DOC production, and subsequent loss, appear to be a function of productivity.

#### Sub-soil drainage

Drainage did not seem to affect exports of DOC from the Zero N plots (Fig. 4.5.1). However, when values of total DOC export from all plots were grouped within either drained or undrained treatments, an analysis of variance showed that there was a significant difference (F = 6.69; P = 0.032), with drained plots exporting significantly less DOC than the undrained plots. Either less DOC was produced in the soils of the drained plots, or DOC was lost between the point of production and the drainage weirs. One major source of DOC would be from the applied manures, but both drained and undrained treatments received equal amounts. As no manures were applied during the present year, residual levels of mobile C are likely to have been low. The other two major sources of DOC are fresh organic remains (mainly plant derived) and older SOM. Levels of productivity are slightly higher on the drained plots (Tyson et al., 1992) so there should be at least as much fresh organic remains for DOC formation as on the undrained plots. Soils from the undrained plots had higher C concentrations (up to 30% higher) in the top 4 cm (~9% C) compared with the drained plots (~7% C) although, below 4 cm, differences were not significant. As DOC export from HNU was nearly three times that from HND (Fig. 4.5.1), the differences in soil C content could not account for all of the difference in export.

The inference, therefore, is that a large proportion of DOC is removed from soil water as it moves into drainage. Several workers have demonstrated that large amounts of DOM can be sorbed from soil solution to mineral surfaces (e.g. Greenland, 1971; Jardine et al., 1989). Evidence for this occurring on our plots is presented in Fig. 4.5.2, where DOC concentrations in water that has passed through to the drainage system is shown to be substantially lower than that of the surface water from the same plots. This suggests that the DOC has been removed by the soil during percolation. Additionally, as draining the soil reduces water content, and hence increases air content, the soil of the drained plots may have been more oxidized, with more Fe(III) oxides available for DOC sorption than was the case for the undrained plots. Both sets of values are lower than those observed at the same time from the HNU plots (Fig. 4.5.2). However, considered together with the difference in soil C content, sorption of DOM may explain the differences in DOC export between the two treatments.

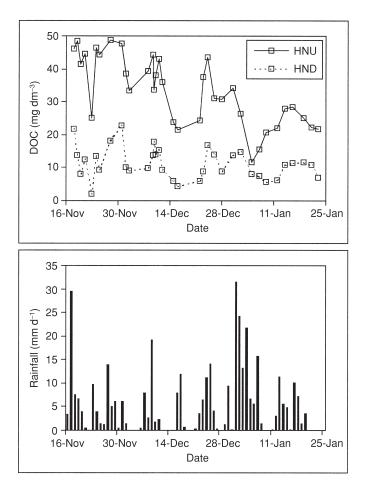


**Fig. 4.5.2.** DOC concentrations in drainage water and composite surface—topsoil discharge from two replicate HND plots (a and b). DOC concentrations in runoff from HNU plots are also shown for comparison (n = 2). See Table 4.5.1 for notation.

### Rainfall intensity

A comparison of the temporal measurements of DOC concentration in the waters leaving the plots with daily levels of rainfall (Fig. 4.5.3) shows that rainfall intensity and hence the rate of passage of water either through or over the soil strongly influences the amount of DOC exported. In general, for both the drained and undrained plots, higher concentrations of DOC occurred at times of high rainfall. However, regressing DOC concentration

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**Fig. 4.5.3.** DOC concentrations in runoff and drainage waters with daily rainfall. Note the response of DOC to rainfall intensity. See Table 4.5.1 for notation.

in exported water with daily levels of rainfall over the whole 2-month period did not show any significant relationships. Explanations for this include: (i) grab samples were taken only once per day and flow rates at the time of sampling may not have reflected the amount of rainfall measured for that day (e.g. heavy rain during an evening, night or weekend may have gone through the system before the next sampling took place); and (ii) concentrations of DOC in runoff from the undrained plots gradually declined with time (see Fig. 4.5.3 for example).

A subsequent intensive (sampling every hour over 12 h) study of drain flow rates and the organic composition of the runoff and drainage waters

during a storm event did allow meaningful comparisons, and produced some contrasting results (McTiernan *et al.*, 1999). The drained plots had positive logarithmic correlations (P < 0.001,  $r^2 = 0.77-0.95$ , depending on fertilizer treatment) between drain flow rates and DOC concentrations. The undrained plots, however, showed negative linear correlations (P < 0.05,  $r^2 = 0.40-0.44$ ) between drain flow rates and DOC concentrations.

For the drained plots, there was a direct correlation between the rate of passage of water through the soil and the concentration of DOC, tending towards an upper limit. The magnitude of this upper limit was dependent on fertilizer treatment. Isotopic and other analyses of the drainage DOM indicated that an increasing amount of OM from the surface of the plots was carried to the drains as rates of flow increased (McTiernan et al., 1999). The size and number of soil channels through which water can pass limit flow to the drainage system and, as the capacity of these channels is approached, an increasing amount of water travels through the topsoil and into surface runoff weirs. As the soils of the drained plots have lower water contents than the soils of the undrained plots, the drained soil may have been more aggregated. This may have resulted in the more extensive formation of macropore channels, permitting rapid flow of water through the upper soil profile to the drainage system.

A large proportion of the storm rainfall on the undrained plots flowed directly over the surface of the soil, giving much higher flow rates than for the drained plots. As there was a dilution of the DOC content of the runoff at the higher rates of drain flow, it would appear that the amount of C available for dissolution was limited, leading to a negative correlation between flow rate and DOC concentration.

We can speculate that there are three stages in the relationship between flow rate and DOC concentration in runoff from the undrained plots. At low rates, most rainfall passes through preferential channels and the soil matrix. As these soils have a high clay content, matrix flow is slow, so even small increases in rainfall intensity result in an increasing proportion of the rain water moving close to and then over the surface. Water moving through the soil profile loses DOM through sorption, and hence any increase in the proportion of water movement over the surface will result in an increase in the concentration of DOC in the discharged water. This could give a positive correlation between drain flow rates and DOC concentration. Secondly, as rainfall intensity increases, the additional water flowing over the surface of the soil removes a proportionate amount of C and there is no relationship between flow rate and DOC concentration. Finally, the amount of C that is available for dissolution and transport in water flowing over the surface of the soil is limited and, as this limit is exceeded, dilution occurs.

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### Acknowledgements

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## Effects of Sterilization and Incubation Temperature on Formation and Quality of Dissolved Organic Matter in Soils

### 4.6

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### Introduction

Dissolved organic matter (DOM) in soils can contribute to a number of important soil processes. These include the solubilization and complexation or sorption of metals and hydrophobic organic compounds which may thus affect plant nutrition, soil genesis or the bioavailability, toxicity and translocation of pollutants (Zsolnay, 1996). Furthermore, pedogenic DOM can act as substrate for microorganisms in the soil solution or in groundwater, enabling denitrification or other redox-relevant activities which influence solution chemistry (Qualls and Haines, 1992; Møller *et al.*, 1999).

Still, little is known about the processes that are involved in the formation of DOM or about the relevance of these processes to DOM quality. Generally, litter leachates, root exudates and microbial degradation products are regarded as important DOM sources (Zsolnay, 1996). However, drying–rewetting cycles, temperature and changes in solution chemistry also affect DOM release (Powlson and Jenkinson, 1976; Tipping and Woof, 1990; Gödde et al., 1996; Lundquist et al., 1999). This may occur either directly through physical or chemical processes or indirectly through the stimulation or inhibition of microbial activity. Our aim was therefore to investigate the role of microorganisms in the release or formation of DOM and on some of its ecologically relevant properties in response to different temperatures and incubation periods. In order to exclude microbial activity, soils were exposed to

 $\gamma$ -irradiation which effectively kills all active and dormant microorganisms but may still allow for some enzyme activity (Powlson and Jenkinson, 1976).

### Materials and Methods

The study was conducted with the sandy–loamy Ap horizon of a Fluvisol from the Oderbruch area in Eastern Brandenburg, Germany. After sample collection in autumn 1997, the soil was air-dried and passed through a 2 mm mesh and then stored in the dark. It had the following properties: 61% sand, 24% clay; pH 7.2; 6.4%  $C_{\rm org}$ ; CEC<sub>p</sub> 227 mmol<sub>c</sub> kg<sup>-1</sup>.

For every incubation experiment, samples were moistened to 60% water-holding capacity (15% w/w) and filled into open steel cylinders (100 ml, 5 cm diameter) and pre-incubated at 5°C for 24 h. A sub-set of these samples was then sterilized by γ-irradiation (25–29 kGy), and a further sub-set of these sterilized samples was re-inoculated by replacing 10% of the soil with non-sterile soil. For the temperature experiment, all samples were then placed into sealed vessels of an incubation apparatus with continuous CO<sub>2</sub> monitoring (Respicond, Nordgren SA) and incubated for 2 weeks at 5, 20 or 35°C. Extracts from before and after incubation were obtained from separate samples. For the long-term experiment, non-sterile and re-inoculated samples were stored in sealed containers with ample headspace to allow for aerobic conditions and extracted repeatedly at 0, 2, 4, 8 and 12 weeks.

For the extraction, the cylinders were placed into a percolation apparatus, where 250 ml of 1 mM CaCl<sub>2</sub> solution was drip-irrigated on the samples at ~40 ml h<sup>-1</sup> and the leachate collected at the base at −60 hPa, thus allowing unsaturated flow in the soil sample. In the 0.45 μm membrane-filtered solutions, total DOC (Shimadzu 5050) and UV absorbance at 280 nm were determined. The Cu complexation ability of DOM was determined by measuring the solubility enhancement of CuO in phosphate buffer solutions (pH 5.5) in the presence of DOM in comparison with a DOM-free control. DOM degradability was assessed by supplementing the extracts with N and P and inoculating with 1% fresh soil solution. After a 5-day incubation period, the remaining DOC was determined. Samples with glucose served as a control for the viability of the inoculum.

All assays were conducted with six replicates, but extracts from two samples were combined to composite samples in order to have sufficient solution for the following fractionation and analyses.

### Results

With increasing incubation temperature, DOC extractability from non-sterile soil samples is greatly reduced (Table 4.6.1). There was a reduction of 89% in DOC following incubation for 2 weeks at 35°C. A similar temperature dependence was found in the re-inoculated samples (not shown) and, for both treatments, the decrease in DOC is clearly related to the increased respiration rates at higher temperatures (Fig. 4.6.1). The slope of the fitted regression shows that  $CO_2$  evolution exceeds the change in extractable DOC by a factor of 1.85. Assuming that all DOC lost during incubation was completely mineralized, 54% of the total soil respiration could be attributed to this process. In the extracts from  $\gamma$ -irradiated samples, initial DOC values were higher than from non-irradiated samples (Table 4.6.1). Temperature effects on DOC extractability were also observed, but declines were only significant at 35°C and still amounted to ~60% of DOC extractable before incubation.

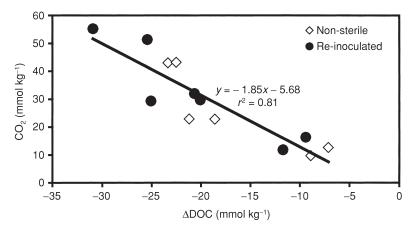
With decreasing DOC extractability in the non-sterile samples, their molar absorptivity increases significantly at incubation temperatures above  $20^{\circ}$ C. The opposite is the case in the sterile samples, where the initially lower  $\epsilon_{280}$  values are reduced further during incubation, with a minimum at  $20^{\circ}$ C.

The Cu complexation ability of DOM was affected by incubation and temperature only in the non-sterile samples. Compared with initial extracts and the sterile samples, DOM from biologically active soils incubated at

**Table 4.6.1.** DOC extractability and DOM characteristics in extracts from sterilized and non-sterile soil samples before and after incubation at different temperatures. For each soil treatment, significant effects of incubation temperature on solution parameters are indicated by different letters following the values in the columns (Duncan test).

	DOC (mmol kg <sup>-1</sup> )	${\epsilon_{280}}^1$ (I m $^{-1}$ mmol $^{-1}$ )	Cu complexation (mmol mol <sup>-1</sup> DOC)	DOM degradability (%)
Non-sterile				
Before	30.9 <sup>a</sup>	25.6a	4.4 <sup>a</sup>	15 <sup>a</sup>
5°C	21.3 <sup>b</sup>	26.2a	5.7 <sup>b</sup>	11 <sup>a</sup>
20°C	7.0°	31.0°	12.3 <sup>d</sup>	63 <sup>b</sup>
35°C	3.4 <sup>d</sup>	29.8 <sup>b</sup>	8.1°	80 <sup>b</sup>
Sterile				
Before	46.5 <sup>a</sup>	18.4 <sup>a</sup>	4.7 <sup>a</sup>	24 <sup>ab</sup>
5°C	46.6a	14.6 <sup>b</sup>	4.7 <sup>a</sup>	18 <sup>b</sup>
20°C	35.8 <sup>ab</sup>	11.8°	4.5 <sup>a</sup>	32ª
35°C	27.3 <sup>b</sup>	12.8 <sup>bc</sup>	5.3ª	19 <sup>b</sup>

<sup>&</sup>lt;sup>1</sup>UV absorption at 280 nm.



**Fig. 4.6.1.** Relationship between the change in DOC extractability and respiration during incubation of non-sterile and re-inoculated soil samples at different temperatures.

20°C can bind almost three times more Cu. At 35°C, Cu complexation ability is lower than at 20°C but still elevated in comparison with 5°C.

Since the reduced DOM extractability at higher temperatures in the non-sterile soils is attributable to increased mineralization activity (Fig. 4.6.1), one would expect an accumulation of recalcitrant soluble compounds. However, our data indicate that a much larger proportion of DOM extractable after incubation at 20 and 35°C is utilizable by soil microorganisms, reaching almost 80% at 35°C (Table 4.6.1). DOM from sterile samples also shows some incubation effects, but they were small and not consistent with temperature.

When biologically active soil samples were incubated over a longer time period, DOC extractability declined rapidly but, even after five extractions in 12 weeks, soluble organic compounds were not depleted but remained at a similar level to that in the third extract (Table 4.6.2). DOM quality, as determined by its molar absorptivity, was affected mainly during the first 4 weeks, when  $\epsilon_{280}$  increased by ~60% and later only declined slightly. In contrast to the fairly stable DOC concentrations and  $\epsilon_{280}$  values after the 4th week, the Cu complexation ability of DOM continued to increase with incubation time to reach a maximum in the 12th week that was almost three times higher than in the initial extracts (Table 4.6.2).

### Discussion

Soil sterilization by  $\gamma$ -irradiation proved to be an effective measure in eliminating microbial activity without interfering greatly with other properties of the soluble organic matter fraction. DOM extractability and

parameters are maloated by american following the values in the obtaining (Duncam test).								
Incubation time (weeks)	DOC (mmol kg <sup>-1</sup> )	${\epsilon_{280}}^1$ (I m $^{-1}$ mmol $^{-1}$ )	Cu complexation (mmol mol <sup>-1</sup> DOC)					
0	26.1ª	26.8 <sup>a</sup>	7.7 <sup>a</sup>					
2	8.0 <sup>b</sup>	35.5 <sup>b</sup>	7.9 <sup>a</sup>					
4	5.7°	42.8 <sup>d</sup>	13.8 <sup>b</sup>					
8	3.3 <sup>d</sup>	39.9 <sup>bc</sup>	15.9 <sup>b</sup>					
12	4.0 <sup>e</sup>	37.1 <sup>cd</sup>	20.5 <sup>c</sup>					

**Table 4.6.2.** DOC extractability and DOM characteristics in repeated extracts from non-sterile soil samples during a 12-week incubation at 20°C. Significant effects of incubation time on solution parameters are indicated by different letters following the values in the columns (Duncan test).

DOM degradability were slightly increased by the treatment, as observed by Powlson and Jenkinson (1976), which may be related to the lower specific UV absorptivity of these substances since this indicates lower aromaticity (Chin et al., 1994). As temperature increases, DOM extractability and some DOM properties are altered in the sterile samples, which is probably due to the remaining enzyme activity (Powlson and Jenkinson, 1976). However, as the effects are much more pronounced in the non-sterile samples, certain changes in DOM properties can clearly be attributed to microbial activity. Contrary to our expectations and reports from Gödde et al. (1996), increasing temperatures did not enhance the net release of soluble organic matter but decreased it, since DOM present at the beginning of the incubation was utilized as substrate. Apparently, the physical effect of rewetting the air-dry samples for the experiments on DOM release was much more pronounced than subsequent microbial activity. This effect is commonly observed and attributed to the disruption of aggregates and the release of otherwise non-available organic matter (Lundquist et al., 1999).

The changes in DOM quality with increasing incubation temperature show that microorganisms either preferentially degrade certain DOM fractions or actually release compounds with different properties from those present initially in the soil. The increase in UV absorptivity in the remaining DOM fraction indicates an expected accumulation of more recalcitrant substances (Chin *et al.*, 1994). However, this is inconsistent with the greatly elevated biodegradability of DOM at 20 and 35°C. Together with the increased Cu complexation ability of DOM at higher temperatures and longer incubation period, we regard this as evidence for an increased contribution of microbial metabolic products to the soluble organic matter fraction. These products could consist of low molecular weight organic acids which have a high metal complexation ability and are

<sup>&</sup>lt;sup>1</sup>UV absorption at 280 nm.

easily degradable (Jones, 1998). It remains unclear, however, why such products should accumulate at higher temperatures and are not mineralized in the soil.

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### Nitrogen Fluxes Through Sustainable Farming Systems in the Mid-hills of Nepal

4.7

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### Introduction

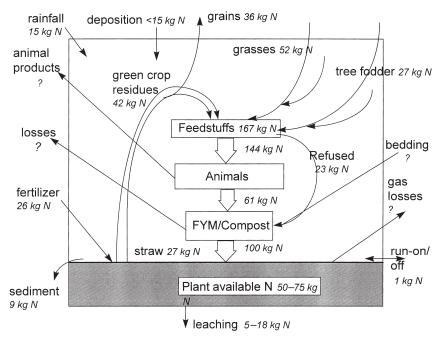
Traditional subsistence farming systems in the mid-hills of Nepal derive their sustainability from the close integration of forestry, livestock husbandry and crop production, in which nutrients, particularly N, are cycled within an apparently closed system (Jodha, 1990). With intensification of crop production to feed a burgeoning population and exposure to external market pressures, the linkages between elements of the traditional system weaken, potentially causing the system to become increasingly unsustainable, with deleterious consequences for the environment as the system becomes increasingly leaky with respect to N (Jodha, 1990). Creating a nitrogen budget for a hypothetical household in the mid-hills of Nepal provides a tool for analysing these two very different scenarios, their implications for sustainability and their possible impact on soil organic matter levels.

This chapter first uses data gathered from the literature to identify the fluxes of N within a hypothetical household, and by quantifying them to indicate their relative importance. It then uses experimental data from the field to validate two pathways for the flow of N within the household. The first considers the flow of N into cropped land in manure and fertilizer and out of cropped land in grains. This is a major flux pathway. The second provides estimates of the losses of N through leaching, which are anticipated to be large in a monsoon climate, and yet are poorly quantified. This may reflect the rate of mineralization of soil organic matter and so the sustainability of the system.

### **Estimation of N Fluxes for Hypothetical Households**

Pilbeam et al. (2000) synthesize literature values to define the characteristics of a hypothetical household in the mid-hills of Nepal. This household owns 1 ha of land divided in a 2:1 ratio between rain-fed bench terraces (bari land) and irrigated lowland (khet land). Characteristically, two crops are grown per year on each land type; maize-millet is the dominant rotation on bari land, while rice-wheat rotations dominate khet land. All households own a number of both large and small ruminants, and some poultry. An indeterminate number of trees are grown on the land owned by a household. These together with crop residues and grasses growing on terrace risers and wasteland provide fodder for the livestock. The proportion of animal feed coming from within the household boundary relative to that from outside is not known.

Figure 4.7.1 presents the size of N pools and the fluxes of N across the boundary of the household (which in the figure is indicated by the box) in a single year. Some fluxes have not been quantified, either because they are not known currently (e.g. gaseous N losses from soil), or because the relevant data were not readily available (e.g. losses of N in wool, milk and meat). Of the quantified annual fluxes, the more significant inputs of N



**Fig. 4.7.1.** Magnitude of N fluxes over a year for a hypothetical household owning 1 ha of land in the mid-hills of Nepal. Arrows represent fluxes. ? represent unknown quantities.

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occur in fertilizer, rainfall or animal feed, either as grasses or as tree fodder. The most significant loss of N from the household occurs in cereal grains rather than in leachate, eroded sediment or runoff. The largest flux of N occurs in the passage of fodder into the animal and so into the manure, which is returned to the land. Although this particular flux pathway is relatively conservative of N (apparently little N is lost from or added to this cycle), the household is not completely self-sustaining with respect to N. The household N budget depends upon the input of N particularly as fodder from areas of land external to the household.

### **Experimental Validation of Two N Fluxes**

### Fluxes in grain and straw

Two experiments located at ARS-Pakhribas, Dhankuta, Nepal provide crop yield data and tissue N concentrations. Seven experimental treatments were applied to a maize-millet rotation on *bari* land and a rice-wheat rotation on *khet* land. The treatments were zero inputs, and inputs of N at two rates as manure alone, fertilizer alone or manure plus fertilizer (FYM) in combination. The higher rate of N represented farmer-recommended practice for a particular crop, while the lower rate was half of the recommended dose. If these N inputs to *bari* and *khet* land are scaled to represent the area of each land type in an hypothetical household (i.e. 0.66 and 0.33 ha, respectively), then the N input as FYM and fertilizer for each experimental treatment can be compared with that for the hypothetical household (see Table 4.7.1). More fertilizer is applied in the experiments

**Table 4.7.1.** Amounts of N (kg) in fertilizer and manure applied in each of six experimental treatments (see text for details) to maize–millet and rice–wheat rotations expressed on the basis of a hypothetical household owning 1 ha divided in a 2 : 1 ratio between *bari* and *khet* land.

	Maize-millet (bari)		Rice-whe	eat (khet)	Total ho	Total household		
Treatmenta	Fertilizer	Manure	Fertilizer	Manure	Fertilizer	Manure		
HF	60	0	43.3	0	103.3	0		
HM	0	60	0	43.3	0	103.3		
HFM	30	30	21.7	21.7	51.7	51.7		
LF	30	30	21.7	0	51.7	0		
LM	0	30	0	21.7	0	51.7		
LFM	15	15	10.8	10.8	25.8	25.8		
Fig. 4.7.1					26	100		

<sup>&</sup>lt;sup>a</sup>Treatment symbols: H, high N rate; L, low N rate; F, fertilizer; M, manure.

than in the household, while relatively more manure is applied in the household than in the experiments, which suggests that a household is relatively independent of external sources of N. Overall, the total N inputs in a hypothetical household are greater than those in the experiment.

Table 4.7.2 shows the average crop yields in two seasons for all seven treatments from the two experiments at ARS-Pakhribas compared with the yields used to generate the data in Fig. 4.7.1. In most cases, grain and straw yields were greater in the experimental data than in the data for the household. Similarly, the tissue N concentrations were generally greater for the experimental data than the 10 or 5 mg g<sup>-1</sup> N used for grain and straw, respectively, in the calculations for the hypothetical household. Consequently, fluxes of N in grain (i.e. crossing the household boundary) or in straw (i.e. cycling internally as animal fodder) are greater in the experimental conditions than predicted from the literature.

### Fluxes in leachate

Ceramic cup solution samplers were installed at three experimental sites (Pakhribas, Dordor Gaun and Dordor Tar) on *bari* land in the mid-hills of Nepal in 1998. They were installed to a depth of 75 or 90 cm in three of the seven experimental treatments described above, namely zero input, manure only and fertilizer only, both applied at the higher rate of N.

Table 4.7.3 shows the average concentration of nitrate in the soil solution collected between the end of June and mid-October in the ceramic cups at 50 kPa suction applied for 24 h. There seems to be little difference

**Table 4.7.2.** Average grain and straw yields (kg ha<sup>-1</sup>) and tissue N concentrations (mg g<sup>-1</sup>) from experiments including four crops at ARS-Pakhribas in 1997 and 1998, and grain and straw yields used in the hypothetical household.

		Hypothetical	Experime	ental yield	Experimental tiss	sue concentration
		household	1997	1998	1997	1998
Maize	Grain	2000	3590	1980	14.6	14.2
	Straw	3000	3513	ND	7	8.1
Millet	Grain	1160	1937	2033	10.8	7.9
	Straw	1740	3391	2347	5.7	8.2
Rice	Grain	2000	1835	2593	12.2	12.8
	Straw	3000	4811	4333	8.7	6.8
Wheat	Grain	2500	2437	3148	25.1	ND
	Straw	3750	3717	4760	3.5	ND

ND denotes not determined.

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**Table 4.7.3.** Average concentrations (p.p.m.) and amounts (kg N ha $^{-1}$ ) of nitrate N collected at depths > 75 cm under three experimental treatments at three sites in the mid-hills of Nepal in 1998. Values in parentheses represent a standard error of the mean.

	Pakhribas		Dordo	or Tar	Dordor Gaun		
Treatment	p.p.m.	kg N ha <sup>-1</sup>	p.p.m.	kg N ha <sup>-1</sup>	p.p.m.	kg N ha <sup>-1</sup>	
Zero input	1.18 (0.176)	8.9	2.61 (0.280)	19.6	0.14 (0.021)	1.1	
Fertilizer	3.28 (1.011)	24.6	2.25 (0.185)	16.9	0.15 (0.021)	1.1	
Manure	1.11 (0.179)	8.3	1.87 (0.219)	14.0	0.14 (0.023)	1.1	

in the concentration of nitrate in the soil solution in the three treatments, at least at Dordor Tar and Dordor Gaun. The apparently greater value for the fertilized treatment at Pakhribas is not significantly different from the other two treatments. It may however reflect the impact of top-dressing N to maize which gave initially high nitrate concentrations in the soil solution that subsequently declined to values similar to the other treatments, thus giving a larger mean and error than found for other treatments. The absence of any treatment differences indicates a constant rate of mineralization of soil N irrespective of nutrient inputs to the surface. Applications of <sup>15</sup>N-labelled fertilizer show that much of the applied N fertilizer is immobilized rapidly in the soil (C.J. Pilbeam, unpublished data). Assuming a drainage loss of 750 mm, which is approximately half of the annual rainfall, then losses of nitrate N by leaching vary considerably between sites, but in 1998 were always < 25 kg N ha<sup>-1</sup>, often considerably so. These data broadly support the range of values used in Fig. 4.7.1.

### **Conclusions**

N budgets have two purposes. The first is to identify gaps in knowledge which are amenable to measurement. The second is to facilitate the efficient management of N inputs, and thereby control outputs of N from the system, particularly losses to the environment. N inputs to and outputs from a hypothetical household in the mid-hills of Nepal appear to be roughly in balance, and so, at the level of the household, productivity is sustainable. However, this requires the importation of N into the household, perhaps as fertilizer, but more commonly from the forest areas in tree fodder and grass, with the attendant degradation of the forest resource base in the mid-hills of Nepal. In other developing countries, particularly in Africa, similar degradation occurs where fodder is removed from communal areas, especially forests, and carried to the homestead. Deduced

values for particular N fluxes largely agree with those measured by experiment in the field, which indicates the robustness of the original balance derived from the literature, and its reliability for analysing possible perturbations to the farming system.

### **Acknowledgements**

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### N, P and K Budgets for Some 4.8 UK Organic Farming Systems – Implications for Sustainability

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### Introduction

The economic and environmental sustainability of organic farming is dependent on the efficient use of nitrogen (N), phosphorus (P) and potassium (K). Organic farmers aim to maintain and increase the long-term fertility of soils while minimizing nutrient losses (International Federation of Organic Agriculture Movements (IFOAM)). The emphasis of organic farming is to encourage the efficient cycling of nutrients rather than rely on fertilizers and purchased manure (Lampkin, 1990). On many conventional farms, large inputs of fertilizers and animal feeds, at levels above those removed in outputs, have led to an accumulation of nutrients, such as P, to a degree that many soils have been classified as over-fertilized (Sharpley and Smith, 1989; Mäder et al., 1999). N is brought into organic farming systems through the inclusion of N-fixing crops in the rotation and, as a result, N balances for organic farms are usually positive (Nguyen et al., 1995). The potential for large losses of N when leguminous leys are ploughed led the Royal Commission on Environmental Pollution (1996) to conclude that 'organic systems are inherently liable to nitrogen leaching'. The often reduced inputs of P and K to organic farming systems are believed to lead to negative P and K balances. Johnston (1991) suggested that organic systems are not inherently more sustainable than conventional farming, especially with regard to the long-term availability of P and K.

However, lower production levels, the efficient use of manures and composts and the use of permitted fertilizers, where P and K levels are deficient, may lead to a balance or surplus of nutrients in organic systems.

Nutrient budgets allow a breakdown of nutrient inputs and outputs and, in more comprehensive studies, a quantification of internal flows in farming systems (Watson and Atkinson, 1999). Nutrient budgets have been compiled at a variety of scales and using various methodological approaches across the world (Scoones and Toulmin, 1998; Jarvis, 1999). It is important that for cropping systems based on crop rotation or mixed systems including livestock, such as organic farms, nutrient budgets are considered at the level of the whole farm system (Watson and Stockdale, 1999), so that all potential inputs and outputs are included. Here we will consider both nutrient budgets, compiled as farm gate budgets, and the results of some limited soil analysis to examine whether organic farming in the UK can be sustainable with regard to nutrient cycling.

### Methods

### Soil indices

Soils in England and Wales are analysed routinely for available P and K (MAFF, 1986). Soils can be grouped into categories, the soil P and K index system, where an index of 0 or 1 indicates a possible crop deficiency (MAFF, 1994). A sequential Balzer P extraction (Balzer and Balzer-Graf, 1984) determines soil reserve, plant-available and water-soluble P, respectively, and is carried out by the Organic Advisory Service (OAS) in addition to routine analysis (EFRC, 1999). Results from soil analyses submitted to the OAS during 1997 have been collated and examined using an Access database. The results have also been compared with data on the P and K status of UK arable soils (Skinner *et al.*, 1992). Traditionally, soil tests have been used to assess nutrient availability for crop growth. However, no simple method exists for measuring the potentially available N content of the soil or the longer term release of other nutrients into available forms.

### **Nutrient budgets**

Farm gate budgets are among the simplest form of budgets. These consider only flows that transfer nutrients over the farm boundary, e.g. N fixed, feed, stock, seed, milk and grain (Jarvis, 1999). If the inputs and outputs balance, the farming system is considered to maintain soil fertility. A surplus of nutrients can provide an indication of the potential for losses of nutrients,

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and a negative balance might indicate that the system is unsustainable. For this study, three model organic farms have been derived, which represent the breadth of organic systems in the UK: a typical upland/hill farm with sheep and suckler beef, a lowland dairy farm and a stockless arable/horticultural farm (Shepherd *et al.*, 1999). Farm gate budgets for N, P and K have been compiled from farm records, measurements and standard tables of nutrient contents.

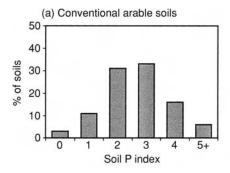
### Results

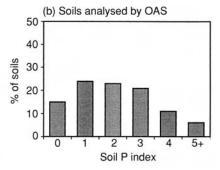
### Soil indices

Data from organic farms showed that 39% of soils were at a P index of 0 or 1, with 23% at index 2 (Fig. 4.8.1b). In conventional arable soils, only 14% were recorded at a P index of 0 or 1 (Fig. 4.8.1a); while in grassland 40% of soils were are at a P index of 0 or 1 (Skinner *et al.*, 1992). In conventional arable soils, 28% of the soils tested were at a K index of 0 or 1 and 48% of grassland (Skinner *et al.*, 1992). Double lactate extraction of P and K, as part of the Balzer P extraction, showed that 86% of soils were deficient in available P and 36% were deficient in available K.

### **Nutrient budgets**

Tables 4.8.1, 4.8.2 and 4.8.3 display nutrient budgets for three model organic farms (upland/hill farm, lowland dairy farm and stockless); to aid comparison, all values are given in kg ha<sup>-1</sup> averaged over the whole farm area. Animal feed and straw are major sources of P and K in the livestock-based systems. In the stockless system, additions of compost are the main source of P and K. In all the systems, symbiotic fixation of N by leguminous plants is important. In the extensive upland system, deposition





**Fig. 4.8.1.** Frequency distribution of the soil P index (MAFF, 1986).

21.3

0.4

Total

Balance

Inputs	N (kg ha <sup>-1</sup> )	P (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )	Outputs	N (kg ha <sup>-1</sup> )	P (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )					
Fixation	6.2	0	0	Stock sales								
Deposition	12.5	0.1	3.3	Cattle	1.2	0.4	0.1					
Stock purchases	~0	~0	~0	Sheep	1.3	0.2	0.1					
Bought-in feed	2.4	0.2	0.5	Wool	0.4	~0	0.1					
Seed	~0	~0	~0									
Straw	0.2	0.1	1.1	Burning	0.1							
				NH <sub>3</sub> volatilization	1.8							

**Table 4.8.1.** Estimates of N, P and K inputs and outputs for a model organic upland/hill farm with sheep and suckler beef (total area 390 ha).

**Table 4.8.2.** Estimates of N, P and K inputs and outputs for a model organic lowland dairy farm (total area 56 ha).

4.8

+16.5

0.6

-0.2

0.3

+4.6

4.9

Inputs	N (kg ha <sup>-1</sup> )	P (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )	Outputs	N (kg ha <sup>-1</sup> )	P (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )
Fixation	117			Stock sales	4	1.0	0.3
Deposition	20	0.2	3.3	Milk	33	6.0	9.0
Bought-in feed	18	2.0	5.0	NH <sub>3</sub> volatilization	14		
Seed	1	0.1	0.2				
Straw	3	0.9	7.0				
Total Balance	159	3.2	15.5		51 +108	7.0 -3.8	9.3 +6.2

**Table 4.8.3.** Estimates of N, P and K inputs and outputs for a model organic stockless arable/horticultural farm.

Inputs	$N = (kg ha^{-1})$	P (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )	Outputs	N (kg ha <sup>-1</sup> )	P (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )
Fixation	79			Crop sales	58	10.0	64
Deposition	20	0.2	3				
Seed	5	0.7	3	NH <sub>3</sub> volatilization	~0		
Compost	50	11.0	38				
Total Balance	154	11.9	44		58 +96	10.0 +1.9	64 20

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also represents a major component of the N input. Main outputs are through crop and animal sales.

### Discussion

The limited data compiled suggest that the soil P index is reduced in soils in organic farming systems, while the levels of available K in soils are less affected. However, this may result from the samples used. Samples from the organic farms were those submitted for analysis rather than sampled routinely, and would have included grass as well as arable fields. Studies have shown that levels of available P and K in soil have both increased (Clark et al., 1998) and decreased (Penfold et al., 1995) on conversion to organic farming. Loes and Ogaard (1997) found reductions in available P and K in soils with previously high levels following conversion to organic farming as a result of the reduction in net imports. Clark et al. (1998) showed that most changes in soil P and K could be predicted from nutrient budgets for the systems.

The nutrient budgets for the model farms show both surpluses and deficits for P and K. Management of nutrients within a stockless rotation is more difficult than in the other systems considered, the import of compost is essential to maintain crop production, and some use of approved K fertilizers may be necessary. On the lowland dairy farm, on which the model is based, rock phosphate is used occasionally, which will address the small P deficit. Within the farms, there may also be large differences in nutrient status over time, with soil nutrient status rising in areas which receive manure, etc., more frequently and declining on fields that receive less inputs or have larger nutrient offtakes (Bacon *et al.*, 1990). A more detailed look at spatial flows would indicate areas of enrichment and depletion on the farm allowing better allocation of P and K. Where P and K supplies are limited, it may be necessary to target them to the most responsive crops. Temporal flows of nutrients also need to be considered as fertilizers are often applied to the ley phase of the rotation and are later made available to subsequent crops.

Loes and Ogaard (1997) suggested that organic farms may maintain lower soil nutrient levels as an adaptation to lower inputs. Mäder *et al.* (1999) demonstrated that plant nutrient deficiencies were not seen, and hypothesized that the larger microbial biomass in soils farmed organically may have been responsible for controlling the supply of P and K to the soil solution. The less intensive nature of production under organic standards may mean that lower soil P and K levels are acceptable. With careful farm husbandry, P and K can also be managed sustainably in organic farming systems, so that except where large reserves have been built up in soils, organic farmers do not mine the fertility of the soil but maintain economically sustainable production levels.

The nutrient budgets for the model farms show surpluses for nitrogen. While N budgets are usually positive on organic farms (Nguyen et al., 1995), the N surplus commonly is lower than in comparable conventional systems (Halberg et al., 1995). Calculated N surpluses were greater than comparable conventional systems on three pairs of farms in New Zealand (Nguyen et al., 1995). However, low P and S availability in the soils on the organic farms may have limited N fixation, so that it was overestimated in the calculation; crop uptake of N in arable crops may also have been limited by other nutrients. The positive N surplus on organic farms may imply that considerable nitrate leaching is likely. However, nitrate leaching losses measured directly have been found to be significantly lower on organic than similar conventional farms (Eltun and Fugleberg, 1996; Berg et al., 1997). Leaching losses of between 7 and 52 kg N ha<sup>-1</sup> were measured at all the points in a rotation by Philipps and Stopes (1995), with an estimated average of ~20 kg N ha<sup>-1</sup> year<sup>-1</sup>. However, the differences seen between organic and conventional systems are much reduced when leaching is expressed relative to crop production levels (Philipps and Stopes, 1995). Nitrate leaching losses from organic systems cannot be related directly to the magnitude of the N surplus. Management of the grass-clover leys, such as timing of incorporation, weather conditions and N demands of the following crop, need to be considered. Losses of N from manure can also be important; measures to place manure heaps on paved surfaces and under cover can help reduce N losses (Dewes, 1995).

### **Conclusions**

Farm gate budgets are useful in indicating whether the system is balanced at the whole farm scale. The information can also be used for understanding fluxes at a regional level and as a policy tool. For the farmer, a more detailed approach may be required, as average N, P and K balances may mask important differences between fields. More complete information will allow the farmer to optimize management practices and maintain or improve the internal cycling of nutrients. Further work is necessary to understand whether organic farms can be balanced in terms of N, P and K and to allow surpluses and deficits to be interpreted correctly. Additionally, work is required to allow surpluses and deficits to be interpreted alongside measurements of soil nutrient pools.

The number of organic farms in the UK has increased dramatically (Lampkin and Measures, 1999). Limited information exists on the comparative sustainability of organic and conventional agriculture. Direct comparisons between systems are difficult, due to differences in factors such as soils, weeds, pests, diseases, climate and enterprises (Fowler *et al.*, 1993). In addition, the methods of measurement, data recorded and

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conceptual models used can make comparisons problematic (Watson and Atkinson, 1999). A standardized approach is required in order to compare different farming systems appropriately, and the reliability and limitations of the data need to be acknowledged. An approach which combines nutrient budgets and soil measurements, along with other measurements such as economic performance, may increase the reliability of such comparisons.

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4.9

# The Effects of All-arable Organic Rotations on Soil Organic Matter Levels and the Phosphorus and Potassium Status Over the Period 1987–1998

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### Introduction

With the growth of the organic sector in Europe, it is inevitable that there will be an increasing number of conventional specialist arable farmers considering conversion to organic methods of production. Von Fragstein (1996) found that the proportion of stockless farms in Germany varied between 20 and 50%. Similarly, David et al. (1996) and Stopes et al. (1996a) reported that the stockless system is becoming increasingly important in organic farming systems in France and the UK, respectively. It is stated in organic farming principles that organic farming systems should sustain or build soil fertility. In temperate climates such as in Northern Europe, mixed ley-arable rotations are the primary means of maintaining soil fertility, as well as controlling weeds, pests and diseases in organic crop production systems (Lampkin, 1990). On a mixed organic farm, the grass-clover ley is expected to accumulate sufficient N by fixation to support subsequent arable crops. Grass-clover leys typically occupy at least 50% of the mixed farm (National Rivers Authority, 1992), and the manure generated by the livestock can be used to stimulate biological activity and move nutrients both around the farm and within the rotation.

However, for economic viability, all-arable or stockless rotations cannot include a long ley phase to provide a balance between fertility building and exploitative arable crops. Instead, short-term leguminous green manures must be used to accumulate N for the subsequent arable phases of the rotation. Trials on the duration, species composition and management

of leguminous green manures demonstrated that cut and mulched red clover can supply up to 370 kg N ha<sup>-1</sup> year<sup>-1</sup> (Stopes *et al.*, 1996b). Although the green manures may supply adequate nitrogen for the crop rotation, little was known about the maintenance of soil organic matter and the levels of phosphorus and potassium in the absence of either livestock manures or soluble fertilizer inputs. With the increasing interest in stockless farming systems, Elm Farm Research Centre (EFRC) established, in 1987, an organic stockless trial to examine the agronomic and environmental implications for organic all-arable farming systems.

### **Materials and Methods**

The trial site was established at Elm Farm Research Centre (national grid reference, SU414 654) at an altitude of ~60 m, with an average annual rainfall of 710 mm year. The soil type was a clay loam of the Wickham series (Jarvis *et al.*, 1984) with organic matter levels that range from 2.42 to 5.7% across the farm.

The whole farm site was converted to organic production, with Soil Association certification in 1985. The farm operated a 4-year grass-clover ley, 1 year winter wheat and 1 year winter oat rotation. The trial site had previously been in a 5-year white clover–grass ley. In 1987, three 4-year rotations were established in three complete randomized blocks (Table 4.9.1). Every course of every rotation was present in each year, giving a total of 36 plots, each  $20 \times 12$  m.

Ploughing, seedbed preparation, drilling, planting, in-crop weed control and potato harvesting were carried out using standard farm equipment, in accordance with farm practice. Cereal crops and the field beans were harvested using a trial plot combine harvester. Throughout the experiment, and in accordance with Soil Association Standards, rock phosphate was applied to a maximum rate of 180 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> during the green manure phase of the rotations, the rate chosen according to soil analysis and the requirement of the subsequent crops. Lime was also applied to a maximum rate of 2 t ha<sup>-1</sup> of CaO per year. Only crop residues were returned and there

		•		
		Cou	rse	
Rotation	1	2	3	4
Α	Red clover	Winter wheat	Winter wheat	Spring oats
В	Red clover	Potatoes	Winter wheat	Winter oats
С	Red clover	Winter wheat	Winter beans	Winter wheat

**Table 4.9.1.** Stockless rotations in the EFRC replicated experiment 1987–1998.

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were no applications of animal residues or other green waste composts. No applications of potassium fertilizers were made over the lifetime of the trial.

The objective of the experiment was to establish if stockless organic rotations were agronomically viable. The effect on soil fertility was assessed by measuring soil organic matter levels, extractable phosphorus and potassium using the standard ADAS methods (MAFF, 1986). Other parameters measured included crop yields, expressed at a standard 15% moisture content, nutrient offtakes, weed biomass and pest and disease incidence.

In the period 1996–1998, crop yields were measured and the nutrient content for these years was assumed to be an average of the previous 8 years' data.

Statistical analysis (analysis of variance ANOVA) was conducted using Genstat 5.3 and the graphs were produced using 'Excel 97'.

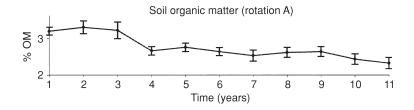
### Results

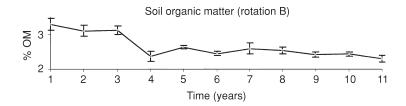
There was a significant decline in soil organic matter levels over time across all three rotations. The most notable change occurred in the first 4 years of the trial, cycle 1, as the management changed from a grassland to an arable system. Between cycle 1 and 2, the soil organic matter percentage fell from 3.02 to 2.58%. The decline between cycle 2 and 3 was less dramatic, with soil organic matter levels falling to 2.42% at the end of the trial. There was no notable difference between rotations. Changes in soil organic matter levels over time are presented in Fig. 4.9.1.

There were no differences between the courses of the rotations although there was a slight decline between course one, the fertility building course, and course four, the furthest point from the green manure crop, as would be expected.

There were no significant changes in the available phosphorus over time, although slow-release rock phosphate had been applied to the green manure crops when soil analysis revealed a deficiency. The available P was not significantly affected by the course of the rotation but there were differences between the rotations. Available P in rotation C, at 14.02 mg P kg<sup>-1</sup>, was significantly lower compared with rotations A (15.42 mg P kg<sup>-1</sup>) and B (15.68 mg P kg<sup>-1</sup>).

Levels of potassium were not affected over the 11 years of the trial, despite no application of supplementary fertilizers or livestock residues. There was no difference in soil potassium levels between rotations A (142.6 mg K kg<sup>-1</sup>) and C (138.0 mg K kg<sup>-1</sup>). However, there was a significant effect between rotation A and rotation B, in which the available K levels were 128.3 mg K kg<sup>-1</sup>. These results are supported by the crop offtake data, presented in Table 4.9.2.





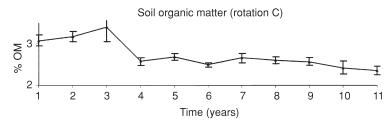


Fig. 4.9.1. Soil organic matter level changes over time.

The lower nutrient offtakes in rotation A were as a result of poorer yields. The potato crop accounted for 46% of nitrogen, 58% of phosphorus and 81% of the potassium exported from rotation B. The field beans exported 150 kg N ha<sup>-1</sup> from an average yield of 3.5 t ha<sup>-1</sup>. This is comparable with the export of N for field beans reported by Sprent and 't Mannetje in 1996. The subsequent wheat crop exported as much N as the first wheat crop in rotation C; therefore, despite the large export of N by the bean crop, it has to be assumed that a significant account of N had been fixed.

### Discussion

It commonly has been reported that during the period of conversion to organic production there is a fall in soil organic matter levels and other soil nutrients (Wander *et al.*, 1994). Similarly, it is well documented within

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**Table 4.9.2.** Nutrient offtakes from cash crops (N, P and K kg<sup>-1</sup> ha<sup>-1</sup> year<sup>-1</sup>). The values in paretheses are the standard error.

		Crop yield (t ha <sup>-1</sup> )		Course o	f rotation of course	fftake per		Mean offtake
Rotation	2	3	4	2	3	4	per course	per rotation
Nitrogen								
Α	4.29 <sup>a</sup> (1.3)	2.64a (1.3)	2.03 <sup>a</sup> (1.3)	61 (2.65)	36 (3.57)	25 (0.18)	41	122
В	29.35 <sup>b</sup> (8.8)	4.29a (1.3)	3.19 <sup>a</sup> (1.3)	87 (0.02)	64 (3.14)	39 (0.14)	63	190
	14.41 <sup>c</sup> (3.6)							
С	3.75 <sup>a</sup> (1.3)	4.10 <sup>a</sup> (1.1)	3.99 <sup>a</sup> (1.3)	56 (3.07)	150 (8.59)	57 (3.12)	88	263
Phosphoru	IS							
Α				25 (1.93)	15 (1.53)	12 (3.60)	17	52
В				61 (4.87)	26 (2.02)	19 (2.06)	35	106
С				23 (1.96)	29 (1.83)	24 (1.76)	25	76
Potassium								
Α				21 (1.04)	14 (1.30)	15 (1.32)	17	50
В				, ,	20 (1.32)	, ,	63	189
С					49 (3.24)		29	88

<sup>&</sup>lt;sup>a</sup>Yields adjusted to standard 15% moisture content.

conventional agricultural systems that a change from grass to arable production will result in losses of soil organic matter. To date, little work has been carried out on the effects of changing from an organic ley–arable system to an organic stockless system. However, it seems likely that the changes in soil organic matter levels between the first 4 years (cycle 1) and the following years (cycles 2 and 3) are a result of the transition from a grass to an arable-based system.

All three rotations presented here were experimental and were never meant to represent organic practice. It is clear from this study that organic rotations with only 25% or less fertility building and only recycling crop residues result in a decline in soil organic matter levels. Changes in the setaside regulations, after this trial was established, have allowed organic farmers to use setaside as a means for fertility building for up to 2 years.

The crop needs for available P appear to be adequately met through the application of rock phosphate to the green manure crops. Cropping sequence appears to have an effect on the availability of P from the slow-release fertilizers (EFRC, 1993).

bTotal yield.

<sup>&</sup>lt;sup>c</sup>Ware (marketable) yield.

It is assumed under organic standards that soils with a clay content of > 20% should, under organic management, be able to derive sufficient K from the mineralization of clay minerals to sustain crop yields. Over the 11 years of this study, it would appear that this was the case, although longer term studies over a range of soil types are needed to substantiate this claim.

### Conclusions

Under experimental conditions, there has been a decline in soil organic matter levels under stockless organic management. This was significant in the first 4 years of the trial. If stockless rotations are to become increasingly widespread within the organic sector, then longer periods of fertility building and the use of composted green waste would be necessary to prevent the decline in organic matter levels under stockless organic management.

It would appear that using rock phosphate may be capable of maintaining soil P levels under stockless management with moderate yields and nutrient offtakes. Similarly, on clay soils, there appears to be no significant decline in the soil K status.

However, organic farming needs to review the potential impacts of organic management on soil fertility particularly under intensive all-arable and horticultural situations. Changes in soil organic matter levels often occur over a long time period and it is important that rotation design evolves as our understanding of soil processes develops.

### Acknowledgements

The hard work and dedication of all the scientific, advisory and support staff and students, in particular H. Bulson, S. Millington, C. Stopes and J. Welsh, who have contributed to this project over its lifetime, are gratefully acknowledged. This work was funded by the Progressive Farming Trust.

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4.10

### Manure Fertilization for Soil Organic Matter Maintenance and its Effects Upon Crops and the Environment, Evaluated in a Long-term Trial

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### Introduction

Soil organic matter (SOM) content and turnover are regarded as important indicators of soil fertility. For crop production, it is of prime interest to know which immediately beneficial effects these indicators have on land use criteria such as crop yield, yield stability and environmental impact. The significance of well-developed soil biological characteristics for successful crop growth and yield is not quite clear, despite numerous studies on this issue. With manure fertilization, increased SOM contents have been observed in several field trials in central and Northern Europe, mostly carried out under site conditions very similar to ours (Asmus et al., 1987; Asmus, 1992; Garz and Stumpe, 1992; Neméth and Tóth, 1992; Kätterer and Andrén, 1999). In some of these trials, crop yield and a variety of soil characteristics have been evaluated. Higher SOM contents have been found to have positive effects on yield and yield components of cereals (Görlitz and Asmus, 1984; Schnieder, 1984; Görlitz, 1986) as well as on soil density, pore volume and maximum water capacity (Asmus et al., 1987). However, in a trial with generally high SOM contents (the lowest was 1.72% C), different humus contents had no significant effects on yield when optimum mineral fertilization was applied (Stumpe et al., 1983).

The aim of the fertilization trial started at our institute in 1980 is to study the long-term effects of manure and mineral fertilization. In particular, the interplay of fertilization, soil fertility and crop yield is being investigated. The results of two crop rotation periods, i.e. the last 8 years,

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are reported here. Attention focuses on the question of whether there are fundamental differences between farmyard manure and mineral fertilization as regards soil fertility indicators, crop yield and the soil's function as a carbon sink or source.

### **Materials and Methods**

A long-term field trial on a sandy orthic Luvisol with 590 mm precipitation per year and 9.5°C annual mean temperature is being carried out to compare three types of fertilizer: (i) composted farmyard manure (CFM); (ii) composted farmyard manure with application of all biodynamic preparations (CFMBD); and (iii) mineral fertilization (MIN, i.e. calcium ammonium nitrate, super phosphate and potassium magnesia). Production and use of the preparations were described by Steiner (1924) for the first time. Further details are given by, for instance, Koepf (1981). Each type of fertilizer is applied at three different levels, corresponding to a total nitrogen content of 60, 100 and  $140 \text{ kg ha}^{-1} \text{ N}$  to wheat and rye and 50, 100 and 150 kg ha<sup>-1</sup> N to potatoes. The nine treatments are implemented identically in four replicates on four fields with four different crops. This gives 36 plots in each of the four fields. Manure is applied before sowing or planting, mineral fertilizer is applied to spring wheat and potatoes before sowing and planting, and to winter rye in spring. To cereals, a part of the nitrogen amount in the medium and high fertilization treatments (20 and 40 kg ha<sup>-1</sup> N) is applied during tillering as liquid manure in CFM and CFMBD or as calcium ammonium nitrate in MIN. The legume crop remains unfertilized in all treatments. The nutrient amounts applied yearly with the mineral and manure treatments are listed in Table 4.10.1.

Straw of the MIN plots remains on the field, but is removed from the CFM and CFMBD plots. Crop rotation is red clover (*Trifolium pratense* L.), alternatively Persian clover (*Trifolium resupinatum* L.), spring wheat (*Triticum aestivum* L.), potatoes (*Solanum tuberosum* L.) and winter rye (*Secale cereale* L.). The trial has been under way with this design since 1985/86. It started in 1980 with the aim of investigating the effects of fertilization on food quality (Abele, 1987). Therefore, from 1980 to 1984, the trial had the same treatments at a higher level of manure fertilization and a different crop rotation. Except for fertilization, all other elements of cultivation are the same in all treatments and follow normal organic farming practices. More details of the trial have been published earlier by Bachinger (1996) and by Raupp (1996).

Where statistical requirements were fulfilled, analyses of variance were calculated, taking type and level of fertilizer as fixed effects and replicates and years as random. In the tables, mean values of the main effects with different letters are significantly different (P < 0.05). The least significant

difference (LSD) stated in Table 4.10.2 refers to the interaction between fertilization type and level (P < 0.05). With the yield results of different treatments, a bivariate correlation was calculated according to the method described by Sokal and Rohlf (1995).

### **Results and Discussion**

SOM content in topsoil was found to be higher with manure than with mineral fertilization. Table 4.10.2 shows the latest available results of 1998, after 18 years of differentiated fertilization. The highest content was observed in the treatment with biodynamic preparations. This level is equivalent to the 1.05% C<sub>org</sub> analysed by Abele (1987) with a mixed sample of each field at the beginning of the trial in 1980. Thus, only the organic fertilizer with biodynamic preparations was able to maintain SOM content until the present day; applying the same quantities of organic matter but

**Table 4.10.1.** Nutrient amounts (kg ha<sup>-1</sup>) applied yearly with solid and liquid manure or mineral fertilizer (phosphorus, potassium and sulphur amounts in CFM and CFMBD are averages of 5 years; the other values are constant every year).

Low				Medium				High				
Fertilization	N	Р	K	S	N	Р	K	S	N	Р	K	S
CFM												
Solid	60	16	76	9	80	21	101	12	100	26	126	15
Liquid	0	0	0		20	1	33		40	2	66	
CFMBD												
Solid	60	17	81	9	80	23	108	11	100	29	135	14
Liquid	0	0	0		20	1	32		40	2	65	
MIN	60	50	75	73	100	75	100	102	140	100	125	132

**Table 4.10.2.** Organic carbon content (% dry matter) in topsoil after 18 years of manure or mineral fertilization (mean values of four fields in 1998).

Fertilization	CFM	CFMBD	MIN	Average
Low	0.83	0.95	0.79	0.86ª
Medium	0.93	0.99	0.79	0.90 <sup>b</sup>
High	0.98	1.07	0.80	0.95°
Average	0.91 <sup>b</sup>	1.00°	$0.79^a$	$LSD_{05} = 0.05$

<sup>&</sup>lt;sup>a,b,c</sup>Mean value of either type or level of fertilization with different superscript letters are significantly different (P < 0.05).

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omitting the preparations gave lower  $C_{\rm org}$  values. The effect of fertilization levels depends significantly upon their type. Whereas increasing amounts of mineral fertilizer had no effect upon SOM content, higher levels of manure fertilization preserved higher organic carbon contents.  $C_{\rm org}$  differentiation set in during the first years of the trial between 1980 and 1983/84 (values presented by Bachinger, 1996). In all treatments,  $C_{\rm org}$  levels have now been fairly constant for > 10 years.

Bachinger (1996) investigated soil microbial parameters in 1988–1991. As an example, the results of 1989 are shown in Table 4.10.3. The treatments with high and constant humus values also had the higher biological activity. Protease and dehydrogenase activity as well as microbial biomass (chloroform fumigation extraction) were more pronounced in the manure than in the mineral treatments. A further study found higher amino acid contents in the manure compared with the mineral treatments (Scheller *et al.*, 1997). This is an indication of the important role of farmyard manure and amino acids for humus synthesis.

Crop yield (Table 4.10.4) did not show the same pattern as soil parameters. Whereas spring wheat gave the same yield with all types of fertilizer, winter rye had 33% and potatoes 10% higher yields with mineral than with

Table 4.10.3.	Averages of soil microbiological parameters in topsoil by type and level of
fertilization; res	ults of 1989 (Bachinger, 1996).

Fertilization	CFM	CFMBD	MIN	Low	Medium	High
PA <sup>1</sup>	0.27 <sup>b</sup>	0.26 <sup>b</sup>	0.20 <sup>a</sup>	0.25 <sup>a</sup>	0.25 <sup>a</sup>	0.23 <sup>a</sup>
$C_{mic}^2$	34.9 <sup>b</sup>	37.8 <sup>b</sup>	26.1 <sup>a</sup>	30.6ª	34.1 <sup>a</sup>	34.2ª
DHA <sup>3</sup>	109.1 <sup>b</sup>	121.9 <sup>c</sup>	75.9 <sup>a</sup>	94.5ª	103.8a	108.6ª

 $<sup>^{</sup>a,b,c}$ Mean values within a half-row with superscript different letters are significantly different (P < 0.05).

**Table 4.10.4.** Crop yield (dt ha<sup>-1</sup>) by type and level of fertilization, averages of the 1992–1995 and 1996–1999 crop rotation periods.

Fertilization	CFM	CFMBD	MIN	Low	Medium	High
Winter rye	28.3ª	29.8ª	37.7 <sup>b</sup>	28.1ª	31.7 <sup>b</sup>	36.1°
Spring wheat	38.9	39.3	41.0	36.8a	40.2 <sup>b</sup>	42.2c
Potatoes	247 <sup>a</sup>	262 <sup>b</sup>	271 <sup>b</sup>	230 <sup>a</sup>	262 <sup>b</sup>	288 <sup>c</sup>

 $<sup>^{</sup>a,b,c}$ Mean values within a half-row with different superscript letters are significantly different (P < 0.05).

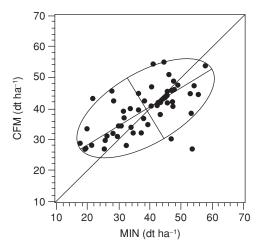
<sup>&</sup>lt;sup>1</sup>Protease activity (mg g<sup>-1</sup> Tyr); method of Ladd and Butler (1972).

<sup>&</sup>lt;sup>2</sup>Microbial biomass (SIR; mg 100 g<sup>-1</sup> C); method of Anderson and Domsch (1978).

<sup>&</sup>lt;sup>3</sup>Dehydrogenase activity ( $\mu g$  10 g<sup>-1</sup> TPF); method of Thalmann (1967).

manure fertilization, compared with CFM. The biodynamic preparations increased potato yields by 15 dt ha<sup>-1</sup> (6%) on average over all years. Higher levels of fertilization influenced yields positively with all crops. Basically, the yields reflect the specific nitrogen demand of each crop at various growth stages in combination with the different nutrient availability of organic or mineral fertilizer in those stages. Spring wheat, cultivated in the year after the legume crop, generally may have the benefit of a favourable position in the crop rotation. This may explain why there was no yield difference between manure and mineral fertilization.

The yields of all crops are not very high and varied considerably from year to year, most likely because of the extreme site conditions: a sandy soil and dry-warm climate with drought from May to July in most years. Irrigation is possible, but only at low levels of 20–60 mm year<sup>-1</sup> because of the limited capacity of our equipment. In cereals, the occurrence of weeds depended on fertilization, but did not explain the yield differences between treatments (Raupp et al., 1998). Severe pest or disease problems only arise with potato late blight in some years, causing more severe infestation in mineral treatments, and by colorado beetle. In spring wheat, yield stability, i.e. fluctuation over the years, differed between organic and mineral fertilization. Figure 4.10.1 shows the correlation between the yields of the mineral and manure treatments (in both cases at a high fertilization level) over the last 14 years. If the yield difference is the same over the entire yield range, the slope of the major axis of the ellipse should be at 45°, i.e. not significantly different from 1. However, the slope is < 1 for the medium and high fertilization level (P < 0.05; Table 4.10.5). Thus, under good growth conditions, minerally fertilized wheat yielded more than manure fertilized wheat. However, yield declined much more in years of poor conditions. In these years, the plant-soil system with manure fertilization seems to be able



**Fig. 4.10.1.** Bivariate correlation between spring wheat yields with the high level of mineral fertilization and of composted farmyard manure (results of four replicates and 14 years, n = 55; confidence ellipse for P < 0.05).

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Table 4.10.5.	Bivariate correlation between spring wheat yields with composted farmyard
manure (Y1) ar	nd mineral fertilization (Y2) at three fertilization levels; results of four replicates and
14 years, $n = 5$	5 ( <i>P</i> < 0.05).

Fertilization	Major axis regression	Confidence limits for the slope b
Low	Y1 = 5.786 + 0.815 Y2	0.596 < b < 1.092
Medium	Y1 = 16.925 + 0.550 Y2	0.333 < b < 0.816
High	Y1 = 15.956 + 0.620 Y2	0.416 < b < 0.868

to compensate for poor environmental conditions and yielded up to 10 dt ha<sup>-1</sup> more than the mineral treatments. Possible compensation mechanisms may include increased root growth with manure fertilization (reported by Bachinger, 1996 for CFMBD) or modified morphological characteristics and yield components (studied by Boemer-Schulte, 1992 in this trial).

With respect to global climate change, currently there is a debate on whether soils can be either a source or a sink of atmospheric carbon dioxide (IPCC, 1996; GACGC, 1998). The issue of which conditions can switch a source to a sink and vice versa is of special interest. A recent study in the USA reported that SOM is increased by organic farming methods (Drinkwater et al., 1998). The authors describe organically managed soils as a substantial carbon sink for carbon dioxide from the atmosphere. The manure treatments in our trial contain 3.6-8.4 t ha<sup>-1</sup> more carbon in the topsoil than the corresponding minerally fertilized plots. These differences are several times higher than those reported in the USA study. However, in contrast to the American experiment, the Corg differences in our trial are the outcome of varying degrees of reduction, not of humus accumulation. Even the soil fertilized with farmyard manure was a carbon source, though to a much lesser extent than the minerally fertilized soil. Carbon losses could only be avoided when farmyard manure application was combined with the biodynamic preparations. Probably the effect of a treatment depends upon the pre-history of the soil. We intend to investigate this matter in more detail, although its relevance to the atmospheric carbon dioxide budget is limited.

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# Effect of Elevated CO<sub>2</sub> and Temperature on Soil C and N Cycling

4.11

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#### Introduction

One potential and highly debated implication of global change is an alteration in the dynamics of soil C in terrestrial ecosystems (Rogers *et al.*, 1999). This potential change in soil C dynamics is important not only because of a possible mitigating effect on rising atmospheric  $CO_2$  concentration, but also because of its influence on quality of soil organic matter. Ultimately, the rate and extent of turnover of organic C produced in an elevated  $CO_2$  environment will control C storage in terrestrial ecosystems (Van Veen *et al.*, 1991).

The decomposition of crop residue inputs is a fundamental component in the turnover of soil organic C which is dependent on several crop, soil, management (i.e. tillage practices) and climatic factors (Potter *et al.*, 1998). Plant factors controlling decomposition such as age, size, chemical composition and residue C: N ratio (Ghidey and Alberts, 1993) may be affected by changing CO<sub>2</sub> level (Torbert *et al.*, 1995; Prior *et al.*, 1997a). Research considering the effect of elevated CO<sub>2</sub> on crop residue decomposition suggests that CO<sub>2</sub>-enriched cropping systems may store more soil C (Torbert *et al.*, 1995, 1998; Prior *et al.*, 1997b). Even if decomposition rates of plant components (produced under elevated CO<sub>2</sub>) are not changed, residue decomposition products may impact soil N dynamics (Torbert *et al.*, 1995, 1996, 1998).

Residue decomposition is a microbial-driven process and soil temperature is an important factor controlling decomposition, thereby influencing soil C dynamics. Thus, potential changes in soil temperature due to global 310 H.A. Torbert et al.

warming could have important effects on soil C cycling (Smith et al., 1999). Studies have shown that increased soil temperature will affect soil nutrient cycling in agroecosystems (Buyanovsky et al., 1986). Also, temperature was shown to be the climatic factor that most closely explained the rate of soil C accumulation due to removal of soil tillage in agroecosystems (Potter et al., 1998). Few studies have examined the interaction of elevated CO<sub>2</sub> with changes in soil temperature. Our objective was to conduct an incubation study to evaluate how changes in soil temperature influence C and N cycling of soil collected from soybean and grain sorghum cropping systems after 5 years of elevated CO<sub>2</sub> treatment.

#### **Materials and Methods**

Soil samples were collected from a 5-year  $CO_2$  enrichment study conducted in an outdoor soil bin at the USDA-ARS National Soil Dynamics Laboratory in Auburn, Alabama, USA. The bin was 2 m deep, 7 m wide and 76 m long, and was filled uniformly with surface soil of a Blanton loamy sand (loamy, siliceous, thermic Grossarenic Paleudult) that had been fallow continuously for > 25 years. Fertilizer and lime additions were used to maintain soil conditions within a normal range for crop production. To ensure adequate plant growth, fertilizer N was broadcast applied at a rate of 34 kg N ha<sup>-1</sup> to the grain sorghum (*Sorghum bicolor* (L.) Moench) and soybean (*Glycine max* (L.) Merr.) crop shortly after planting. An additional 67 kg N ha<sup>-1</sup> was applied to grain sorghum 30 days after planting. All plots were managed under no-tillage conditions.

This study had a split plot design with main plots of two crop species and two CO<sub>2</sub> levels as sub-plots replicated three times. Soybean and grain sorghum were chosen to provide legume and non-legume species that are widely produced in agroecosystems. Open-top field chambers (Rogers *et al.*, 1983) were used to impose CO<sub>2</sub> regimes (365 and 720 µl l<sup>-1</sup>). Harvests consisted of grain sorghum head and soybean pod removal and threshing with a plot combine. Plant stalks were cut (15 cm length) using hedge clippers and uniformly applied to plots.

To determine soil C and N cycling, sieved soil samples were weighed (25 g dry mass basis) and placed in plastic containers; deionized water was then added to adjust soil water content (soil water content equivalent to –20 kPa at a bulk density of 1.3 mg m<sup>-3</sup>). Sample containers were placed in sealed glass jars with 20 ml of water (humidity control) and a 15 ml vial of 1 M NaOH (CO<sub>2</sub> trap), then incubated in the dark at temperatures of 20, 25, and 30°C. Treatment samples were removed after 30 and 60 days. Carbon dioxide in NaOH traps was determined by titrating excess base with 1 M HCl in the presence of BaCl<sub>2</sub>. The cumulative CO<sub>2</sub> emissions after 30 and 60 days incubation were calculated by the difference between

CO<sub>2</sub>-C captured in sample traps and in blanks (glass jars with no soil). The CO<sub>2</sub> emissions divided by total soil organic C were used to calculate C turnover. Soil inorganic N (NO<sub>3</sub>-N and NH<sub>4</sub>-N) was extracted with 2 M KCl and measured by standard colorimetric procedures using a Technicon Autoanalyzer II (Technicon Corp., Tarrytown, New York). The net N mineralization was the difference between the final and initial inorganic N contents for the incubation.

Statistical analyses of data were performed using the mixed procedure of the Statistical Analysis System at an established *a priori* level of  $P \le 0.10$  (SAS, 1996). The term 'trend' is used to designate appreciable, but non-significant, treatment effects which differed at the 0.10 < P < 0.20 level.

#### **Results and Discussion**

For both crop species, elevated  $CO_2$  resulted in a significant increase in the residue mass returned to plots (Torbert *et al.*, 1997), resulting in a significant increase in total C and total N (0–5 cm soil depth) after 5 years (Table 4.11.1). At the 5–10 cm depth, a similar trend for an increase in total N was observed under elevated  $CO_2$ , but not with total C. The increased retention of N in the soil system in the elevated  $CO_2$  treatment resulted in a significant reduction in the C: N ratio for soil under soybeans at the 0–5 cm depth, and for both the sorghum and the soybean at the 5–10 cm depth (Table 4.11.1). The high C: N ratio at both depths and the significant difference between treatments indicated that the soil C was in an unsteady state (Table 4.11.1).

Table 4.11.1.	Effect of plant species and atmospheric CO <sub>2</sub> level on soil total C (g kg <sup>-1</sup> ), total N
(g kg <sup>-1</sup> ) and C:	N ratio <sup>1</sup> .

	Tot	al C	Tota	al N	C : N	ratio
CO <sub>2</sub> level	Sorghum	Soybean	Sorghum	Soybean	Sorghum	Soybean
0–5 cm Ambient Elevated	3.7 <sup>a</sup> 4.2 <sup>b</sup>	4.1ª 5.4 <sup>b</sup>	0.21 <sup>a</sup> 0.24 <sup>b</sup>	0.25 <sup>a</sup> 0.36 <sup>b</sup>	17.3 <sup>a</sup> 17.3 <sup>a</sup>	16.1 <sup>a</sup> 14.9 <sup>b</sup>
5–10 cm Ambient Elevated	2.3 <sup>a</sup> 2.4 <sup>a</sup>	2.4ª 2.2ª	0.13 <sup>a</sup> 0.14 <sup>a</sup>	0.13 <sup>a</sup> 0.14 <sup>a</sup>	18.7 <sup>a</sup> 17.9 <sup>b</sup>	18.2ª 15.7 <sup>b</sup>

<sup>&</sup>lt;sup>1</sup>Values represent means of three replicates. Means within a column followed by the same letter do not differ significantly (0.10 level).

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During the soil incubation,  $CO_2$  emission, N mineralization and C turnover were greatly affected by both time and soil depth (Table 4.11.2). While plant species had little significant effect on  $CO_2$  emission, N mineralization was much higher with soybean compared with sorghum at both time periods and soil depths.

Increasing temperature increased soil  $CO_2$  emission, N mineralization and C turnover at 30 days for both soil depths (Table 4.11.3). Likewise, at 60 days, increasing temperature increased soil  $CO_2$  emission, N mineralization and C turnover at the 0–5 cm depth and the N mineralization at the

**Table 4.11.2.** Effect of plant species and soil depth on CO<sub>2</sub> emission, N mineralization and C turnover<sup>1</sup>.

	C mineralize	ed (mg kg <sup>-1</sup> )	N mineralize	ed (g 100 g <sup>-1</sup> )	C turnover	(g 100 g <sup>-1</sup> )
Crop	0–5 cm	5–10 cm	0–5 cm	5–10 cm	0-5 cm	5–10 cm
30 days Sorghum Soybean	154 <sup>a</sup> 174 <sup>a</sup>	92ª 102ª	4.6 <sup>a</sup> 12.6 <sup>b</sup>	0.8 <sup>a</sup> 1.6 <sup>b</sup>	4.0 <sup>a</sup> 3.8 <sup>a</sup>	3.9 <sup>a</sup> 4.5 <sup>a</sup>
60 days Sorghum Soybean	394ª 401ª	297 <sup>a</sup> 294 <sup>a</sup>	11.4ª 23.0 <sup>b</sup>	2.8 <sup>a</sup> 4.8 <sup>b</sup>	10.1 <sup>a</sup> 8.7 <sup>b</sup>	12.5ª 12.9ª

<sup>&</sup>lt;sup>1</sup>Values represent means of three replicates. Means within a column followed by the same letter do not differ significantly (0.10 level).

**Table 4.11.3.** Effect of temperature and soil depth on CO<sub>2</sub> emission, N mineralization and C turnover<sup>1</sup>.

Temperature	C mineraliz	ed (mg kg <sup>-1</sup> )	N mineralize	ed (g 100 g <sup>-1</sup> )	C turnover	(g 100 g <sup>-1</sup> )
(°C)	0-5 cm	5–10 cm	0-5 cm	5–10 cm	0-5 cm	5-10 cm
30 days						
20	135 <sup>a</sup>	91 <sup>a</sup>	4.8 <sup>a</sup>	0.2ª	3.1 <sup>a</sup>	3.9 <sup>a</sup>
25	159 <sup>b</sup>	92 <sup>a,b</sup>	6.3 <sup>a,b</sup>	1.6 <sup>a,b</sup>	3.8 <sup>b</sup>	4.0 <sup>b</sup>
30	199°	108 <sup>b</sup>	14.7 <sup>b</sup>	1.8 <sup>b</sup>	4.7 <sup>c</sup>	4.7 <sup>c</sup>
60 days						
20	304ª	299 <sup>a</sup>	11.6 <sup>a</sup>	2.4 <sup>a</sup>	7.2 <sup>a</sup>	12.8 <sup>a</sup>
25	360 <sup>b</sup>	308 <sup>a</sup>	14.9 <sup>b</sup>	3.8 <sup>b</sup>	8.4 <sup>b</sup>	13.2a
30	528°	280ª	16.7°	5.1°	12.6 <sup>c</sup>	11.9ª

<sup>&</sup>lt;sup>1</sup>Values represent means of three replicates. Means within a column followed by the same letter do not differ significantly (0.10 level).

5–10 cm depth. No interaction between soil temperature and elevated CO<sub>2</sub> was observed.

No significant difference was observed for N mineralization at the 0-5 cm depth between the CO<sub>2</sub> treatments, but elevated CO<sub>2</sub> resulted in a significant reduction in N mineralization at the 5-10 cm depth for the 30-day period (Table 4.11.4). No significant difference was observed between ambient CO<sub>2</sub> and elevated CO<sub>2</sub> treatments for CO<sub>2</sub> emission at the different time periods and soil depths. However, because of the increased level of soil total C present under elevated CO2, a significant reduction was observed with elevated CO2 for C turnover at both time periods for the 0-5 cm soil depth, compared with ambient CO<sub>2</sub> (Table 4.11.4).

Our findings indicated that the effects of elevated CO<sub>2</sub> on plant decomposition processes observed with isolated plant material (i.e. little difference observed between CO<sub>2</sub> emission, but a reduction in N mineralization with elevated CO<sub>2</sub> (Torbert et al., 1995, 1998)) could be observed with soil samples collected following a 5-year elevated CO<sub>2</sub> field experiment. Nitrogen cycling within the plant-soil system will probably be altered with elevated CO<sub>2</sub> and may be the controlling factor for C storage in these systems.

Results from this study indicate that nutrient cycling may be increased in these agroecosystems with an increase in soil temperature and, since there was no observed temperature by CO<sub>2</sub> treatment interaction, changes in residue quality will not greatly reduce nutrient availability to growing plants and may reduce the impacts predicted from global warming. Furthermore, increased biomass production with elevated CO<sub>2</sub> will probably result in increased soil C storage since no significant increase in CO<sub>2</sub> emission was observed and a significant reduction in C turnover was

turnover <sup>1</sup> .	Effect of atmospheric CO <sub>2</sub> and soil depth on CO <sub>2</sub> emission, in mineralization and					iization and C
	C mineraliz	ed (mg kg <sup>-1</sup> )	N mineralize	ed (g 100 g <sup>-1</sup> )	C turnover	(g 100 g <sup>-1</sup> )
CO <sub>2</sub> level	0–5 cm	5–10 cm	0–5 cm	5–10 cm	0–5 cm	5–10 cm

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	C mineralized (mg kg <sup>-1</sup> )		C mineralized (mg kg $^{-1}$ ) N mineralized (g 100 g $^{-1}$ )		C turnover (g 100 g <sup>-1</sup> )	
CO <sub>2</sub> level	0-5 cm	5-10 cm	0-5 cm	5-10 cm	0-5 cm	5-10 cm
30 days Ambient Elevated	158 <sup>a</sup> 171 <sup>a</sup>	104 <sup>a</sup> 91 <sup>a</sup>	8.6ª 8.5ª	1.5 <sup>a</sup> 0.9 <sup>b</sup>	4.1 <sup>a</sup> 3.6 <sup>b</sup>	4.4 <sup>a</sup> 4.0 <sup>a</sup>
60 days Ambient Elevated	387 <sup>a</sup> 407 <sup>a</sup>	306 <sup>a</sup> 285 <sup>a</sup>	16.7ª 17.7ª	3.9 <sup>a</sup> 3.7 <sup>a</sup>	10.0 <sup>a</sup> 8.8 <sup>b</sup>	12.9ª 12.5ª

<sup>&</sup>lt;sup>1</sup>Values represent means of three replicates. Means within a column followed by the same letter do not differ significantly (0.10 level).

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found under conditions of elevated  $CO_2$ . The potential impact of elevated  $CO_2$  on soils of agroecosystems may be important for the future management and productivity of these systems because small improvements in soil organic C can have important positive influences on soil physical properties such as soil hydraulic conductivity, soil bulk density, soil porosity, soil aggregate stability, soil water retention and rainfall infiltration.

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# Interactions Between 4.12 Elevated CO<sub>2</sub> and N in Soils: Influence on N<sub>2</sub>O Fluxes and Rhizosphere Denitrifier Activity

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#### Introduction

Increasing atmospheric CO<sub>2</sub> concentrations are expected to alter both the quality and quantity of products fixed by photosynthesis, and hence global C and N cycles (Ross et al., 1996). Plants influence soil microbial processes in a variety of ways, largely through modifications of the soil physical and chemical environment. The soil microbial communities likely to be influenced most strongly by such perturbations are the rhizosphere communities (Grayston et al., 1996). One possible effect of an increased rate of CO<sub>2</sub> fixation under elevated CO<sub>2</sub> is the release of more C as exudates and detritus transferred into the rhizosphere soil (Canadell et al., 1996). If some of this C is used by heterotrophic microbes, an effect on soil N transformations, including those leading to N<sub>2</sub>O emissions, would be anticipated (Davidson, 1991). Several consequences are possible. Higher net primary productivity under CO<sub>2</sub> enrichment may result in a greater net removal of water from the rhizosphere and thus create better conditions for  $O_2$  supply promoting the establishment of nitrifying bacteria (Klemedtsson et al., 1987). Alternatively, greater plant water use efficiency, higher root biomass and large microbial activity under elevated CO<sub>2</sub> may lead to a reduction in O<sub>2</sub> availability which, after wetting of soil and surplus of root exudates, subsequently may favour the synthesis of denitrifying enzymes and thus of denitrifier activity and growth (Klemedtsson et al., 1987; Højberg et al., 1996). Root N uptake and enhanced N immobilization by growing soil microbial populations may also diminish the availability of mineral N to  $N_2O$ -producing nitrifiers and denitrifiers (Højberg *et al.*, 1996). The net outcome of the possible effects of elevated  $CO_2$  on those processes controlling  $N_2O$  emissions are therefore complex.

Results from field experiments (Ineson et al., 1998; Kammann et al., 1999) and laboratory incubations (Robinson and Conroy, 1999) have provided the first evidence of a positive feedback between elevated atmospheric  $CO_2$  and greater soil-to-atmosphere  $N_2O$  fluxes. However, more information is necessary in order to elucidate the influence of environmental conditions and to link  $N_2O$  fluxes from the soil surface to below-ground processes. The present work was designed to assess possible interactions between elevated atmospheric  $CO_2$  and N fertilization rates on  $N_2O$  fluxes, plant growth and rhizosphere denitrifier activity under controlled conditions.

#### **Materials and Methods**

#### Soil cores

An arable soil was sampled at Gullane (East Lothian, Scotland, GR 484813) to a depth of 20 cm. The soil was sieved moist (< 10 mm), and stored at 4°C before use. The soil was an imperfectly drained sandy loam (Peffer series), characterized by 62% coarse sand; 30% fine sand; 6.06% silt; 2.07% clay; pH<sub>(H2O)</sub> 8.15; 2.52% organic matter; 0.13% total N; 2.09 µg of available N g<sup>-1</sup>, and 50% water-filled pore space (WFPS) at field capacity. Soil cores (upright section of PVC pipe, 30 cm in height, 10.2 cm in diameter) were prepared by packing dry weight equivalents of field-moist soil to a pre-determined bulk density of 1.3 g cm<sup>-3</sup>. Nitrous oxide emissions from similar soil types have been reported to be significant as WFPS increases from 50 to < 90% (Smith et al., 1998). Thus, pre-packed soil cores were saturated in a modified Hoagland's nutrient solution (excluding any source of C or N) for 16 h, and left to drain gravimetrically for 48 h before the experiment started. The water content was maintained by placing each soil core on an individual water table. WFPS ranged from 65 to  $68 \pm 2.2 - 2.9\%$  in the first 8 cm and from 80 to  $84 \pm 0.8 - 2.8\%$  at lower depths.

#### Plant-soil microcosms

Spring barley (*Hordeum disticum* L.) was grown (one plant per pot) for 25 days in microcosms maintained under controlled conditions and set up to facilitate sequential monitoring of the soil—root atmosphere (soil—root cores were isolated from the above-ground part of the plants).

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#### **Experimental design**

The experiment was established in a growth room (16 h photoperiod, 20–15°C and 80% humidity) as a two-factor experiment (split plot) in a randomized complete block design. The atmospheric CO<sub>2</sub> concentrations (ambient and elevated) were considered as the main plot and the sub-plot N fertilization rates (low and high) were arranged randomly within each main plot. Every CO<sub>2</sub> level was replicated four times, and each N fertilization level was replicated twice within each CO<sub>2</sub> unit.

Treatments were designated as follows:  $N_0(ACO_2)$  = low N input (10 kg N ha<sup>-1</sup>) at ambient  $CO_2$  concentration (369 ± 2 µmol mol<sup>-1</sup>  $CO_2$ );  $N_1(ACO_2)$  = high N input (170 kg N ha<sup>-1</sup>) at ambient  $CO_2$  concentration (369 ± 2 µmol mol<sup>-1</sup>  $CO_2$ );  $N_0(ECO_2)$  = low N input (10 kg N ha<sup>-1</sup>) at elevated  $CO_2$  concentration (716 ± 4 µmol mol<sup>-1</sup>  $CO_2$ ); and  $N_1(ECO_2)$  = high N input (170 kg N ha<sup>-1</sup>) at elevated  $CO_2$  concentration (716 ± 4 µmol mol<sup>-1</sup>  $CO_2$ ).

Total N for high N input was split into two doses of 80 and 90 kg N ha<sup>-1</sup> applied at days 2 and 16, respectively. Total N for low N input was applied once at day 16. Nitrogen was applied as NH<sub>4</sub>NO<sub>3</sub>.

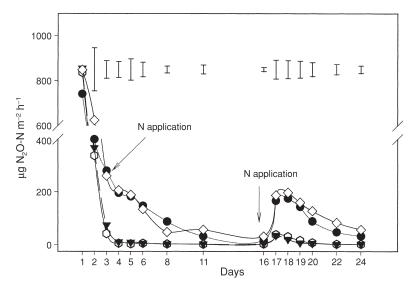
#### **Analysis**

Soil cores were sealed daily to allow gases to accumulate in the enclosed headspace. After 1 h, two 5-ml gas samples were removed from the headspace atmosphere using gas-tight Hamilton syringes. These samples were analysed for  $N_2O$  by gas chromatography. At harvest, plant shoots were excised at the soil surface, and the soil core removed intact from the microcosms. The soil adhering to the roots after shaking was defined as rhizosphere soil, which typically represented a 1 mm thick layer on the root surface. Samples of rhizosphere soil were recovered after washing the roots in distilled water and filtering the combined root washing through a membrane filter (0.45  $\mu$ m). Plant material was analysed for total dry weight and total N uptake. Rhizosphere soil was analysed for total dry weight (oven dry at 105°C), ninhydrin N microbial biomass (Jorgensen and Brookes, 1990) and denitrification enzymatic activity (Smith and Tiedje, 1979).

The data were analysed by two-way ANOVA using the Genstat 4.0 Statistical Package. Significant main effects were separated with paired Student's t-test (P < 0.05).

#### **Results and Discussion**

The very high  $N_2O$  fluxes (741–849  $\mu$ g  $N_2O$ -N m<sup>-2</sup> h<sup>-1</sup>) recorded on day 1 in all treatments (Fig. 4.12.1) were probably associated with the disruption



**Fig. 4.12.1.** Fluxes of  $N_2O$  during the 25 days growing period of spring barley plants under  $N_0(ACO_2)$  ( $\bullet$ ),  $N_1(ACO_2)$  ( $\bullet$ ),  $N_0(ECO_2)$  ( $\blacktriangledown$ ),  $N_1(ECO_2)$  ( $\diamond$ ). Error bars correspond to the standard error of means from the two-way ANOVA analysis.

and wetting of the soil during the establishment of the experiment. Wetting-up of soils has been reported to cause a significant increase in C and N availability as well as a physical impact on  $O_2$  diffusion, resulting in a transient period when NO and  $N_2O$  production may rise by a factor of between 2 and 20 (Davidson, 1991). Gas fluxes in our experiment declined sharply after 24 h.

Fluxes of N<sub>2</sub>O (Fig. 4.12.1) were concentrated in relatively short periods (~5 days) following N fertilization, with significant differences (P < 0.001) between the two N levels applied at all times. The average N<sub>2</sub>O production rates for the entire measurement period after the first N application (days 3–16) were of  $10.3 \pm 2.8$  and  $17.3 \pm 6.4 \,\mu g \, N_2 O - N \, m^{-2} \, h^{-1}$  for N<sub>0</sub>(ACO<sub>2</sub>) and N<sub>0</sub>(ECO<sub>2</sub>), respectively, and of  $143 \pm 19$  and  $140 \pm 22 \,\mu g \, N_2 O - N \, m^{-2} \, h^{-1}$  for N<sub>1</sub>(ACO<sub>2</sub>) and N<sub>1</sub>(ECO<sub>2</sub>), respectively. With the second N application, enriched CO<sub>2</sub> atmosphere caused the rate of N<sub>2</sub>O efflux in high N input to increase gradually from 11 to 74% at the end of the experiment. However, those effects were not statistically significant due to a high variability in the data.

Total  $N_2O$  emissions of 50.1  $\pm$  9.8 and 57.4  $\pm$  16.3 mg  $N_2O$ -N m<sup>-2</sup> for  $N_1(ACO_2)$  and  $N_1(ECO_2)$ , respectively, represented 0.26 and 0.34% of the fertilizer N applied. These results were in the range of  $N_2O$  emissions reported by Clayton *et al.* (1997) for grassland in Scotland fertilized with ammonium nitrate. Ineson *et al.* (1998) in a free air enrichment experiment

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(FACE) in Switzerland recorded a large and immediate increase in  $N_2O$  emissions following the application of ammonium nitrate in solution. This response was 27% greater at an atmospheric  $CO_2$  concentration of 600  $\mu$ mol mol<sup>-1</sup> compared with that at 350  $\mu$ mol mol<sup>-1</sup>. However, in the FACE experiment in low fertilized grassland, Kammann *et al.* (1999) reported that increased  $N_2O$  fluxes between enriched and ambient atmospheric  $CO_2$  sites happened mainly during the summer–autumn vegetation period, and not during the 2 months following fertilizer application. These findings were suggested to be connected with a higher microbial population and to the turnover of this microbial biomass during freeze–thaw periods (Kammann *et al.*, 1999).

Plants grown under elevated  $CO_2$  concentrations appeared to produce more biomass. Statistically significant differences in total dry weight (Table 4.12.1) were found between different levels of  $CO_2$  (P < 0.001) and different levels of N (P < 0.01). Allocation to shoot (Table 4.12.1) was increased statistically both by the N (P < 0.01) and the enriched  $CO_2$  (P < 0.001), the trend among treatments being as follows:  $N_0(ACO_2) < N_1(ACO_2) < N_0(ECO_2) < N_1(ECO_2)$ . However, the largest increment (P < 0.05) in allocation of dry matter to plant roots promoted by elevated  $CO_2$  (Table 4.12.1) was observed when N was limiting (272% increment compared with 72% occurring at the high N fertilization rate). As a result, the influence of doubling the  $CO_2$  concentration in our experiment was to increase the root: shoot (P < 0.05) only at the low rate of N input.

The quantity of rhizosphere soil obtained from spring barley plants (Table 4.12.1) varied significantly (P < 0.001) with CO<sub>2</sub> concentration. The pattern observed followed that obtained for total plant dry matter. However, the ratio of rhizosphere soil to root dry weight (data not shown) did not vary significantly with different levels of CO<sub>2</sub> or N, suggesting that the proportion of C exuded per unit root mass was similar under different CO<sub>2</sub> concentrations. Therefore, the effect of CO<sub>2</sub> concentration on

Table 4.12.1.	Effect of atmospheric CO <sub>2</sub> concentration and N supply on dry weight distribution
and N plant up	ake of spring barley plants grown for 25 days.

	Total plant dry weight <sup>1</sup>	Shoot dry weight <sup>1</sup>	Root dry weight <sup>1</sup>	R : S³ ratio	Rhizosphere soil dry weight <sup>2</sup>	N plant uptake <sup>1</sup>
N <sub>0</sub> (ACO <sub>2</sub> )	279ª	239ª	41.5 <sup>a</sup>	0.18 <sup>a</sup>	3.56 <sup>a</sup>	13 <sup>a</sup>
$N_1(ACO_2)$	437 <sup>b</sup>	380 <sup>b</sup>	56.6a	0.15 <sup>a</sup>	6.45 <sup>b</sup>	24 <sup>b</sup>
$N_0(ECO_2)$	660 <sup>c</sup>	506 <sup>c</sup>	154.2 <sup>c</sup>	0.31 <sup>b</sup>	15.44 <sup>c</sup>	25 <sup>b</sup>
$N_1(ECO_2)$	698°	600 <sup>d</sup>	97.5 <sup>b</sup>	0.16 <sup>a</sup>	13.53 <sup>c</sup>	44 <sup>c</sup>

<sup>&</sup>lt;sup>1</sup>dw and N plant uptake in mg per pot; <sup>2</sup>dw in g per pot; <sup>3</sup>root-to-shoot dw ratio.

Mean values in the same column followed by the same letter do not differ statistically (Student's t-test, P < 0.05).

rhizosphere denitrifier populations may be mediated simply through the greater C flux due to bigger plants, in combination with some other indirect effect (i.e. altered N dynamics).

Doubling the atmospheric  $CO_2$  concentration increased the total N uptake (Table 4.12.1) for the whole plant by 90% in the low N treatment and by 81% in the high N treatment. Therefore, despite an increased flux of C substrate into the rhizosphere under elevated  $CO_2$ , microbial populations may have been limited by the availability of N.

The ninhydrin N microbial biomass (Fig. 4.12.2) in the rhizosphere was significantly (P < 0.05) increased due to the greater C supplied by roots under elevated  $CO_2$ , but was not statistically affected by variations in N availability. Differences in microbial biomass between the enriched and ambient atmosphere were 38 and 14% at low and high N input, respectively.

The overall interaction between N levels and  $CO_2$  concentration was not significant for rhizosphere denitrification activity (Fig. 4.12.3), although the individual effect of different levels of N fertilization was significant (P = 0.01). The denitrification rates observed in our study were within the range (0.05–0.39  $\mu$ g N g<sup>-1</sup> h<sup>-1</sup>) observed by Højberg *et al.* (1996) in young barley rhizosphere soil. These authors also reported much higher potential nitrate reduction activity (tenfold) in the rhizosphere than the potential denitrification rate. Enzyme synthesis for nitrate reduction may take place at slightly higher oxygen concentrations than that for other denitrification enzymes. In our experiment, the greater plant growth under elevated  $CO_2$  may have induced a higher water uptake, leading to a relative increase in the  $O_2$  availability and a decrease in the diffusion of N substrate

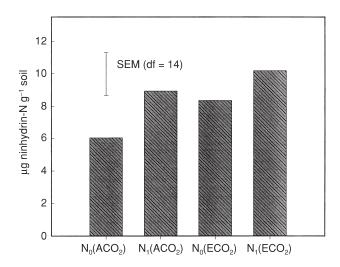


Fig. 4.12.2. Ninhydrin N microbial biomass in the rhizosphere soil.

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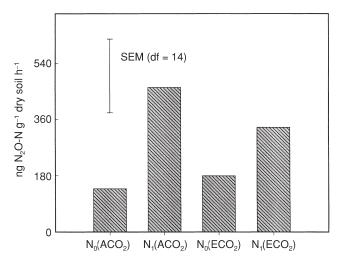


Fig. 4.12.3. Denitrification activity in the rhizosphere soil after harvesting.

to the rhizosphere. All that could have retarded the synthesis of denitrifiers compared with other microbial populations.

The result of our experiment did not indicate a direct plant-mediated effect of elevated CO<sub>2</sub> on N<sub>2</sub>O fluxes or denitrification activity. However, the positive effect on plant growth and microbial biomass has important implications for potential feedback effects between soils and the atmosphere. The turnover of the enlarged microbial biomass, and other processes such as nitrification and nitrate reduction deserve further studies.

#### **Acknowledgements**

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# Carbon Mitigation Options in Agriculture: Improving our Estimates for Kyoto

4.13

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#### Introduction

Soils are only one component of an ecosystem and often respond to global change only indirectly via physiological responses in plants. In response to raised concentrations of  $CO_2$  in the atmosphere, for example, all ecosystem responses are mediated by only three direct effects on plant processes: photosynthesis, respiration and transpiration. Impacts on the soil are mediated through secondary responses in the plant such as changes in water use efficiency, specific leaf area, root: shoot ratio, C: N ratios and many others, so that impacts of raised  $CO_2$  upon the soil can be regarded as tertiary in nature. Figure 4.13.1 demonstrates schematically the complexity of the ecosystem response to raised  $CO_2$ .

Given that soil responses are often so far removed from the direct effects of global change drivers, it is not surprising that soil responses to global change are so difficult to predict. However, it is vital that soils are considered in global change studies since soils represent a major pool of carbon in the biosphere, estimated at  $\sim 1500 \times 10^{15}$  g (Batjes, 1996) globally, about twice that in atmospheric CO<sub>2</sub>. Given that the global stock of soil carbon is so large, it is clear that only small changes in soil carbon stocks can result in significant perturbations to the global carbon cycle. Here we briefly review attempts to quantify European carbon mitigation options in agriculture and examine opportunities and limitations for improving these estimates in the future.

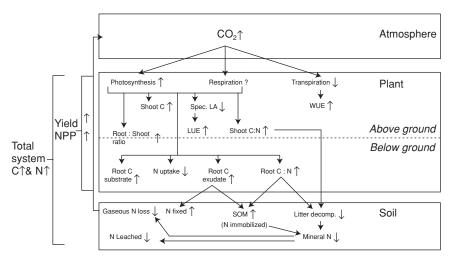


Fig. 4.13.1. Schematic representation of the impacts of raised atmospheric CO<sub>2</sub> on agroecosystems. Arrows indicate the likely direction of the response. See text for further details.

#### Soils and the Kyoto Protocol

The Kyoto protocol (available at: http://www.cop3.de/) was formulated in December 1997 by the 174 parties that ratified the United Nations Framework Convention on Climate Change (UNFCCC). The Kyoto protocol allows carbon emissions to be offset by demonstrable removal of carbon from the atmosphere. Thus, land use/land management change and forestry activities that are shown to reduce atmospheric CO<sub>2</sub> levels can be included in the Kyoto emission reduction targets. These activities include afforestation, reforestation and deforestation (article 3.3 of the Kyoto protocol), and considered for future inclusion are additional activities such as the improved management of agricultural soils (article 3.4).

There are three very important components of the Kyoto protocol. First, it establishes an arbitrary baseline (1990) against which all carbon emissions and carbon mitigation options are compared. Secondly, for the 39 parties listed in Annex B of the Kyoto protocol, it sets quantified emission limitation or reduction commitments for the first time. Thirdly, it establishes a target period for emission reduction or mitigation (the first commitment period; 2008–2012). For the European Union, this means a reduction in CO<sub>2</sub> emissions to 92% of baseline (1990) levels during the first commitment period (2008–2012).

Soils are likely to be most important in the 'additional measures' of Kyoto Article 3.4 where the improved management of agricultural soils is mentioned explicitly as a future possibility for carbon mitigation. It is the opportunities and limitations of the methods to estimate carbon mitigation

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potential in agriculture at the regional level (in this case Europe) that we consider here.

#### Importance of the 1990 Baseline

A number of options for carbon mitigation in European agriculture have been examined including: (i) switching all animal manure use to arable land; (ii) applying all sewage sludge to arable land; (iii) incorporating all surplus cereal straw; (iv) conversion to no-till agriculture; (v) use of surplus arable land to extensify one-third of current intensive crop production (through use of one-third grass—arable rotations); (vi) use of surplus arable land to allow natural woodland regeneration; and (vii) use of surplus arable land for bioenergy crop production. For estimates made before Kyoto, no baseline was in existence.

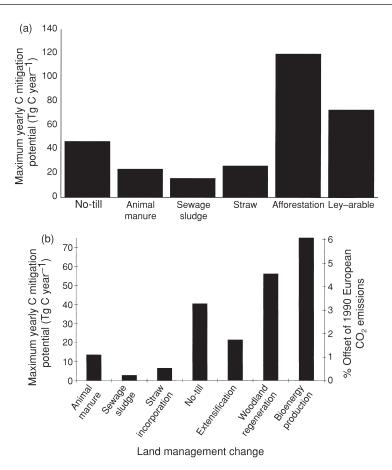
Smith *et al.* (1997a,b, 1998) quantified the carbon mitigation options in agriculture in the European Union and the wider Europe, but these were not set relative to a baseline condition (see Fig. 4.13.2a). The inclusion of a 1990 baseline allows the carbon mitigation potential of each estimate to be assessed more accurately (see Fig. 4.13.2b).

Other changes between pre-Kyoto and post-Kyoto estimates which affect the values presented in Fig. 4.13.2a and 2b include: (i) the use of variable application rates of organic amendments instead of a fixed rate (e.g. animal manure; see also Smith and Powlson, 2000); (ii) a revision of the magnitude of the impact of various management changes on soil organic carbon (SOC; e.g. sewage sludge and natural woodland regeneration); (iii) changes in the agricultural land areas assumed to be available for changes in land use (e.g. a reduction in the predicted level of setaside by 2010 affecting extensification, natural woodland regeneration and bioenergy production scenarios); and (iv) changes in the scenarios themselves (e.g. the inclusion of two separate scenarios for dedicated bioenergy production and natural woodland regeneration, replacing the combined scenario of Smith *et al.*, 1997a,b).

In addition, combined options can be examined. Smith *et al.* (2000) examined combinations of scenarios and showed that there is considerable potential in agriculture for carbon mitigation (see Fig. 4.13.3). A number of the combined scenarios were able to achieve EU emission reduction targets by themselves.

### Improving our Regional Projections of Carbon Mitigation Potential in Agriculture

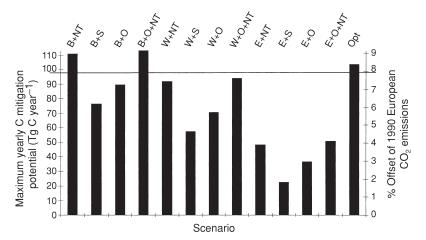
There are many sources of uncertainty in projections such as those presented above. One source of uncertainty arises from the method of



**Fig. 4.13.2.** Carbon mitigation potential in agriculture. (a) The pre-Kyoto estimates based on Smith *et al.* (1997a,b, 1998; Paustian *et al.*, 1997). (b) Post-Kyoto estimates based on revised scenarios including a 1990 baseline condition (Smith *et al.*, 2000).

determining SOC changes under a given management. The simplest approach is to use statistical relationships derived from experiments (as above and Smith et al., 1996). Where uncertainty has been estimated (95% confidence interval), it can be as high as 50% (Smith et al., 1998). Using dynamic simulation models (Smith et al., 1997c) may reduce this error, though this has yet to be demonstrated. The second source of uncertainty arises from the spatial soil–climate data used in the scenarios. In the examples above, differences in soil and climate are subsumed within the statistical relationship derived. Uncertainty would be reduced by including a spatially explicit description of soils and climate either linked to statistical models (e.g. Kern and Johnson, 1993) or, more desirably, linked to dynamic simulation models (e.g. Donigian et al., 1984; Falloon et al., 1998,

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**Fig. 4.13.3.** Carbon mitigation potential of combined changes in agricultural management (Smith *et al.*, 2000). The letter before the first '+' in each scenario indicates the land use employed for 10% surplus arable land; B = bioenergy crops, W = woodland and E = extensification. The letters after the first '+' in each scenario denote the management practice adopted on remaining portions of arable land; NT = no-tillage, S = straw incorporation and O = addition of organic amendments (animal manure and sewage sludge). See Smith *et al.* (2000) for further details.

2000). The greatest source of uncertainty, however, applies to all methods of estimation; the uncertainty associated with establishing the baseline (1990) conditions. Whether using simple statistical models, or simulation models linked to spatial databases, parameters such as the amount of manure available in Europe for agricultural use in 1990 have to be estimated (in the absence of actual census data). Such estimates ultimately are unverifiable and the best that can be done is to make sensible estimates based upon known data, which in this example include head of cattle and pigs in Europe, and equations to estimate the amount of manure produced per head per year. The estimates form the inputs to the scenarios described above or to the set up files of simulation models. Limitations in our ability to reduce uncertainty associated with predictions of regional carbon potential do not, therefore, lie in our technical capability, but in the lack of data upon which we base our baseline estimates.

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### Soils and Soil Organic Matter Along a Transect from Central Taiga to Forest Tundra, Siberia

4.14

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#### Introduction

Permafrost-affected soils cover 13% of the terrestrial surface and 65% of the land surface of the Russian Federation (Bockheim et al., 1994). Despite this large distribution, the periglacial soil landscape of the Siberian taiga has been investigated in small scale only. In addition, results of soil classification according to the Russian taxonomy cannot be related directly to the Canadian or American systems. However, internationally available and comparable data on distribution and classification of permafrost-affected soils in Russia are prerequisites to transfer knowledge on carbon cycling obtained for Northern American soils to the situation in Siberia. One goal of our study, therefore, was to assess the distribution of soils along a South-North transect covering the vegetation belts of the central taiga, the northern taiga and the forest tundra according to USDA Soil Taxonomy. The latitudinal zonation of vegetation and climate provides, furthermore, a good opportunity to study the effects of climate on soil organic matter (SOM) along this transect. Such investigations in these rural areas are of particular importance, because global warming is expected to be most at high latitudes including Siberia. Thus, the second goal of this investigation was to compare the chemical and isotopic composition of SOM along the climatic gradient.

#### Study area

We have chosen our test area near the shores of the Yenisei river in central Siberia between Turukhansk (65°N) and Dudinka (70°N). Mean annual temperature varies between -7.6°C (Turukhansk) and -10.7°C (Dudinka). Corresponding mean summer (July, August and September) temperatures are 10.4 and 5.7°C, respectively; summer precipitation was ~150–200 mm in the whole study area. The permafrost characteristics closely follow the temperature gradient. The depth of the active layer increases from 0.6-2 m in the north to > 3 m in the south (J.G. Karpov and E.L. Baranovskij, 1998, personal communication). Concurrently, the thickness of the permafrost decreases from 100-200 to < 50 m. Parent materials in the study area are Quaternary sediments, primarily derived from the Zyryansk stage (122-59 kyears BP) and the Karginskaya interstage (59-24 kyears BP) of the last glaciation (Sachs, 1948). The soil samples were all taken in watershed areas of the Karginskaya terrace that is of glaciofluvial to glaciolimnic genesis. The Karginskaya terrace follows the Yenisei river from Turukhansk to the mouth of the river. The terrace is located 50 m (south) to 30 m (north) above the Jenisseij, and is composed of homogeneously coarse silty to fine sandy substrate. Only little disturbed forested sites were chosen, with dense Larix sibirica, Picea obovata and Pinus sibirica stands in the south, becoming less dense to the north. L. sibirica represented the only larger tree species at the northern treeline in the forest tundra.

#### **Methods**

Soil pits of  $\sim 2 \text{ m} \times 1 \text{ m} \times 1 \text{ m}$  depth were dug with a spade. Following field description, the profiles were sampled by horizon from three sites of the pit beginning with the deepest horizon. Classification of soils was carried out according to Soil Survey Staff (1998). The samples were oven dried (40°C) within 2 days. In the laboratory, samples were analysed with standard methods for pH, texture, pedogenic Fe- and Al-oxides, and contents and stocks of total organic carbon and nitrogen. SOM was analysed for primary, secondary and tertiary resources. Plant-derived lignins (primary resource) were assessed by the CuO oxidation method according to Hedges and Ertel (1982). VSC is the yield of lignin-derived vanillyl, syringyl and cinnamyl units, and (ac/al)<sub>V</sub> is the acid to aldehyde ratio of the vanillyl units indicating the degree of oxidative lignin degradation. The method of Zhang and Amelung (1996) was used to analyse microbially synthesized amino sugars (secondary resource). The yield is given as the sum of glucosamine, gluN; galactosamine, galN; mannosamine, manN; and muramic acid, mur. The ratio of gluN to mur indicates the relative proportion of fungal and bacterial cell wall residues. Pyrogenic C (tertiary resource) was quantified as benzene

polycarboxylic acids released by HNO<sub>3</sub> oxidation according to Glaser *et al.* (1998). The ratio of pentacarboxylic benzoic acid (b5ca) to hexacarboxylic benzoic acid (b6ca) provides information on the degree of condensation. Stable C and N isotopes of forest floor and mineral soil samples were measured on a Finnigan MAT deltaS instrument, and  $\delta^{13}$ C and  $\delta^{15}$ N values were referenced to PDB and N<sub>2</sub> of air, respectively.

#### Results and Discussion

#### Soil development in the belts of vegetation

The results of our pedological study are summarized in Fig. 4.14.1. All soils showed thick mor-type forest floors. In the southern part of the transect, we found as yet unknown intensive weathering horizons with distinct loamification and enrichment of pedogenic oxides in Bw horizons. To the north, the weathering horizons became less pronounced and disappeared. Concurrently, there is a transition from only weak hydromorphic (pseudogley) properties in the deep active layer of the southern soils to strong gleyic properties in the shallow active layer of the northern pedons. In contrast to the small-scale Soil Map of Russia (1995) which classifies only 'kryotaiga soils' in the study area, our investigation indicates a zonal compartmentation of the soils. With increasing hydromorphy due to the higher permafrost table and decreasing evapotranspiration, there appears to be a transition from well-developed Dystrocryepts (central taiga) to Cryaquepts (northern taiga) to Cryaquepts and Gelisols (forest tundra).

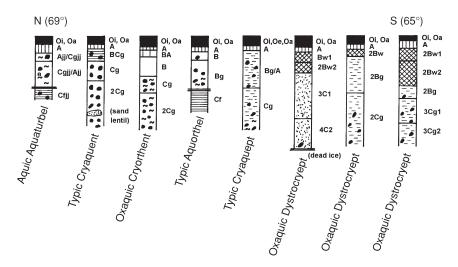


Fig. 4.14.1. Classification of soils in the working area according to Soil Survey Staff (1998).

#### Soil organic matter along the climatic gradient

Carbon stocks ranged from ~100 to ~275 Mg ha<sup>-1</sup> m<sup>-1</sup> soil depth and peaked in soils of the northern taiga (Fig. 4.14.2). This is probably due to a relatively high primary production with a concurrent strong hydromorphy of the soils which restrains OM degradation. In particular in the northern soils, sub-soil horizons contain the most C of the profiles which may be due to slow SOM turnover in the deeper parts of the active layer. The C: N ratios in the A horizons of the soils decrease from 21.7 in the central taiga to 19.7 in the northern taiga, to 14.5 in the forest tundra, suggesting a higher contribution of microbial-derived organic matter in the northerly soils.

The yield of lignin-derived CuO oxidation products in SOM of the mineral soil horizons decreased from south to north (Fig. 4.14.3). Concurrently, the side chain oxidation of the remnant lignin increased, being indicative of a more advanced lignin degradation in the north. We suggest that this is due to the lower root litter input that renews the SOM into the mineral horizons of the northern soils (the root density in mineral soil horizons was lower). In contrast, SOM of the northern soils appeared to be richer in microbial (i.e. bacterial) cell wall residues as is indicated by the amino sugar contents. This can be explained by the more unfavourable conditions (stronger hydromorphy, lower soil temperature) for microbial re-mineralization of the cell wall constituents. Pyrogenic C is a major C

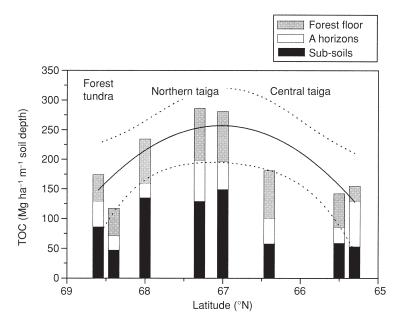
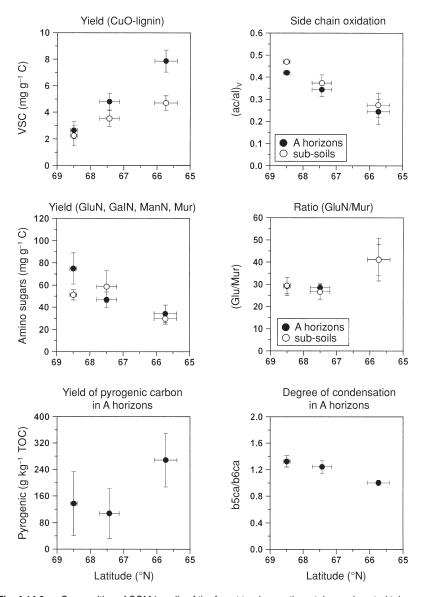


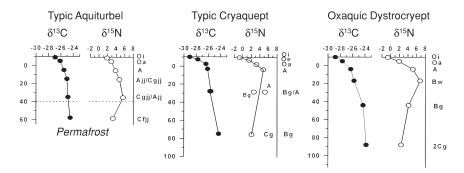
Fig. 4.14.2. Carbon stocks in the soils of the forest tundra, northern taiga and central taiga.

species in all soils, but is highest in the south. Increasing b5ca/b6ca ratios to the north indicate a decreasing degree of condensation.

The profile pattern of  $\delta^{13}C$  and  $\delta^{15}N$  (Fig. 4.14.4) contrasts with the chemical analysis of SOM, if only aerobic decomposition processes are



**Fig. 4.14.3.** Composition of SOM in soils of the forest tundra, northern taiga and central taiga with respect to lignin-derived phenols, microbial-derived amino sugars and pyrogenic C (for details, see Methods).



**Fig. 4.14.4.**  $\delta^{13}$ C and  $\delta^{15}$ N ratios in profiles of a Typic Aquiturbel representing the forest tundra, a Typic Cryaquept representing the northern taiga, and an Oxaquic Dystrocryept representing the central taiga.

considered. Isotopic discrimination is largest in the southern soils, indicating that SOM of these soils is the most processed. However, the strong discrimination observed in mineral surface horizons of the southern soils suggests anaerobic processes, i.e. methanogenesis and denitrification. Both processes result in a pronounced enrichment of the heavier isotope in soil. Reasons for the possibly lower potential of  $CH_4$  and  $N_2O$  formation under forest tundra are less available primary C sources, the longer period when the soils are frozen and the lower soil temperature. The low temperature in the sub-soils close to the permafrost table may also account for the decreasing  $\delta^{15}N$  values in the sub-soils. Yet, we can only speculate about the reason for this observation. It could be due to incorporation of light  $NH_4$  into sub-soil SOM, enrichment of light  $NO_3$  and  $N_2O$  in the sub-soil, or a decoupling of the nitrogen cycle in the sub-soil from that in the surface soil.

In conclusion, this study provides indications that watershed soils of the forest tundra ecotone may be able to store more SOM if the belts of vegetation shift northwards, but emissions of greenhouse gases may also increase.

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## Carbon Dynamics in Upland 4.15 Soils after Serious Fires

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#### Introduction

The estimated global C loss from terrestrial sources to the atmosphere due to fire is 4 g C year<sup>-1</sup> (Andreae, 1991), but this ignores the C loss from soils (Kasischke et al., 1995). The upland soils of the temperate and circumboreal regions contain a vast amount of organic matter because the cool, wet and often acidic soil conditions restrict the rate of decomposition, with the result that organic matter accumulates at the soil surface (Floate, 1977). Such soils are a globally important reservoir of organic C (Post et al., 1985; Eswaran et al., 1993; Howard et al., 1995) and fires are not uncommon in upland areas. Natural vegetation fires are caused predominantly by lightning strikes (Billings, 1964), but in upland areas they are usually the result of human activity. Humans have used fire since stone age times to fashion the landscape, and this has contributed both to the decline of woodland and the maintenance of heath and grass moorland (Tinsley, 1975). In modern times, managed burning is usually restricted to combustion of dry and senescent plant material leaving a layer of ash which is followed by relatively rapid regrowth of the plants with increased vigour (Grant, 1968), and it is thus widely adopted in upland management (Kenworthy, 1963; Gimingham, 1972). Managed burning also reduces the probability of catastrophic fires by removing much of the fuel (Miller et al., 1984). Serious fires occur either completely by accident or when vegetation burning gets out of control, and these can lead to substantial, if localized, loss of soil organic matter either directly by combustion or as a result of erosion from the dried and exposed soil surface. Occasionally, such serious fires lead to 338 S.F.I. Haslam et al.

loss of surface L layers and O horizons, leaving exposed the uppermost mineral horizon of the soil (Radley, 1965; Maltby *et al.*, 1990; Haslam *et al.*, 1998). Destruction of roots and seeds, depletion of the nutrient stock and loss of water-holding capacity of the soil all combine to slow down recovery from severe fires (Haslam *et al.*, 1999).

Although the conditions under which different fires occur are rarely the same, comparison of adjacent sites where fires have occurred at different times allows the investigation of recovery and succession after burning. We believe that conveniently matched fire sites are rare. This study is based on research undertaken at sites of severe fires separated by nearly 20 years in time (1957 and 1976) and 50 m in space in the southern Pennines of northern England (Haslam, 1999), where we have estimated the rates of carbon reaccumulation and investigated details of the recolonization by organisms in the decades since the fires.

#### Carbon Loss and Reaccumulation

The site we have investigated in the Pennines has been described in detail elsewhere (Haslam *et al.*, 1998; Haslam, 1999). It comprises two areas, one in excess of 3 ha and the other nearly 5 ha, where severe fires in 1957 and 1976, respectively, removed all the L layer and O horizon of what was a stagnogley podzol. The total C loss was estimated by determining the amount of C in the L layer and O horizon in the adjacent, unburned soil at 20 randomly selected positions. The rates of reaccumulation were estimated from measurements made in 1996 of C in the L layers and O horizons of soils at 21 randomly selected positions in the sites burnt in both the 1957 and 1976 fires, the underlying assumption being that this C had accumulated since the fires. The justification of this assumption is discussed in more detail by Haslam *et al.* (1998), but essentially it relies on the fires having left the uppermost mineral horizon of the soil exposed.

The total C loss was 30 kg C ha<sup>-1</sup> or 1.47 t C and 0.93 t C as a result of the fires in 1957 and 1976, respectively (Haslam *et al.*, 1998). The amounts of C in the L layers and O horizons of the burned areas when measured in 1996 are shown in Table 4.15.1. Initially the organic matter accumulated in the L layer and consequently there was no recognizable O horizon at the site burned in 1976. From these estimates of C content, the mean rates of C accumulation over the period from 1957 to 1996 and from 1976 to 1996 were estimated. The rate of organic matter accumulation was much slower during the first 19 years after burning than subsequently. During much of the 0–19 year period, the fire sites did not have plant cover and did not, therefore, receive organic matter inputs from plant litter. According to observations in 1984 by one of us (D.W.H.), the 1976 fire site had only ~50% plant cover 8 years after the fire, whilst plant cover on the

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Site	Horizon/layer	Total C content (kg C m <sup>-2</sup> )	C accumulation rate (kg C m <sup>-2</sup> year <sup>-1</sup> )		
1957 fire L	1.67 (1.26, 21)	0.044			
	0	8.42 (5.53, 21)	0.22		
1976 fire	L	0.66 (0.50, 21)	0.035		
	0	Not present	Not present		

**Table 4.15.1.** Total C concentrations in the L layers and O horizons of soils from the 1957 and 1976 fires sites and rates of C accumulation. The mean values are shown with the standard deviations followed by *n* in parentheses. Adapted from Haslam *et al.* (1998).

1957 fire site was complete by 1984 (27 years after the fire). In the later period from 19 to 38 years, the rate of organic matter accumulation was much greater and exceeded the rate of C accumulation for other upland sites that had not been subject to severe fires (between 0.0046 and 0.089 kg C m<sup>-2</sup> year<sup>-1</sup>; Korhola *et al.*, 1995) and lowland mineral soils (between 0.025 and 0.056 kg C m<sup>-2</sup> year<sup>-1</sup>; Jenkinson, 1970).

#### **Recolonization by Organisms**

The slower initial rate of soil organic matter accumulation was consistent with slower recolonization by plants over the first few years after the fires but, by the time we started work at the fire sites in 1996, it was too late to investigate the development of the pioneer communities. However, at the fire sites, we undertook a preliminary investigation of the plants and soil microorganisms as these are, respectively, the major contributors of organic matter to the soils and important decomposer organisms.

The grasses *Deschampsia flexuosa*, *Festuca ovina*, *Agrostis capillaris* and *Nardus stricta*, the rush *Juncus squarrosus* and the moss *Polytrichum commune* established at an early stage (before 20 years) and were replaced over the subsequent 19 years by *Molinia caerulea*. These observations are presented in more detail elsewhere (Haslam *et al.*, 1999).

The sizes of the microbial communities in soils from the burned sites were estimated from biomass C measurements made using the glucose-induced respiration technique (Anderson and Domsch, 1978). In addition to providing information to complement the total C data above, the biomass measurements allowed hypotheses about ecological succession involving soil microorganisms to be tested. Microbial successions in soil have been investigated by Insam and Domsch (1988), Insam and Haselwandter (1989) and Anderson and Domsch (1986), who proposed that the ratio of microbial carbon to total carbon ( $C_{\rm mic}$ :  $C_{\rm total}$ ) increases and that the rate of microbial respiration per unit of biomass (respiratory quotient, qCO<sub>2</sub>) decreases as succession proceeds. However, some of these

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studies have relied on inter-site comparison with only indirect evidence that the sites were at different stages in the same succession. Studies of soil microbial succession with genuine chronosequences are rare. In the case of the post-fire chronosequence, the values of  $C_{mic}$ :  $C_{total}$  and  $qCO_2$  for the L layers were both greater for the 1957 than for the 1976 fire sites (Table 4.15.2). This is the only sensible comparison that can be made in the context of succession as no O horizon had developed at the site of the fire in 1976. These observations offer only partial support for the proposals outlined above of Insam and Domsch (1988), Insam and Haselwandter (1989) and Anderson and Domsch (1986). It is difficult to isolate the influence of time from those of diversity and quality of plant litter inputs and other factors that affect soil microorganisms. For example, Wardle (1993) reported that increases in qCO<sub>2</sub> at late successional sites were due to physiological stress on the organisms and, in a comprehensive critique of the qCO<sub>2</sub> parameter as an indicator of disturbance and ecosystem development, Wardle and Ghani (1995) found no convincing evidence that qCO<sub>2</sub> could be interpreted unambiguously in the context of succession.

There was indirect evidence in the form of the relative concentration of C in larger particle size fractions that soil animals, other important members of the decomposer community, were either less active or less abundant in the burned soils compared with the unburned soil (Haslam *et al.*, 1998). This is not an unexpected observation, but little can be inferred directly and further work will be required to understand the post-fire ecology of soil and litter invertebrates, which both Whelan (1995) and Ahlgren (1994) describe as being poorly understood and the responses of surviving and recolonizing populations as being highly variable.

#### Conclusions

We have provided an estimate for the rates at which organic matter reaccumulates in soils after serious fires. This is important information because one of the predictions of environmental change is increased frequency of hot, dry summers in the temperate and circum-boreal latitudes, which may lead to increased incidence of fires at sites of terrestrial C storage. We have also provided preliminary information about the post-fire succession which suggests that understanding of the successional development of microbial communities is far from complete.

#### **Acknowledgements**

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Site	Horizon/layer	$C_{mic}$ : $C_{total}$	qCO $_2$ ( $\mu g$ CO $_2$ -C $\mu g^{-1}$ biomass C h $^{-1}$ )
1957 fire	L	0.0087 (0.00017)	0.0277 (0.00632)
	0	0.0005 (0.00005)	0.0069 (0.00121)
	Ea, Bh, Bfe and Bs	0.0030 (0.00097)	0.0017 (0.00047)
1976 fire	L	0.0037 (0.00035)	0.0077 (0.00211)
	Α	0.0045 (0.00039)	0.0030 (0.00055)
	Bh, Bfe and Bs	0.0036 (0.00081)	0.0016 (0.00007)

**Table 4.15.2.** Ratio of biomass C to total C ( $C_{mic}$ :  $C_{total}$ ) and biomass specific respiration (qCO<sub>2</sub>) for soil in summer 1997 from the sites of serious fires in 1976 and 1957. Each value is the mean of three replicates and the standard errors are shown in parentheses.

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## Implications of Soil Biodiversity for Sustainable Organic Matter Management

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#### Introduction

Organic matter (OM) plays a pivotal role in the effective functioning of soil systems. It underpins their structural integrity and provides a fundamental basis to soil fertility in that it acts as an energy and nutrient source to soil organisms. The basis of sustainable management of soil OM is one of balance: inputs must be balanced against losses, but there is a need for appropriate dynamics as well, to ensure that nutrient elements are cycled within and between ecosystems. Soil organisms and OM dynamics are inextricably linked, since collectively the biota is the primary agent responsible for the myriad of biochemical transformations that drive elemental cycles and plays an important role in modulating soil structure. Defining biodiversity is not as straightforward as the mass media would wish. It is essentially a concept, and not an entity in its own right, that aims to rationalize a complex set of factors that encompass the basic genetic, taxonomic, trophic and functional components of community and their spatio-temporal dynamics. It should also include the number of different biological forms, entities or units from each of these perspectives, their relative abundance and the degree of interconnectedness between them. Gaston (1996) paraphrases it succinctly and elegantly as 'a biology of numbers and difference'.

Below-ground biodiversity usually exceeds that which prevails above ground by orders of magnitude. From virtually any perspective, the soil biota is exceptionally diverse. Numerically, 100 g of a typical temperate grassland soil contains in the order of 10<sup>11</sup> prokaryotes, 10<sup>4</sup> m of fungal

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hyphae, 10<sup>7</sup> yeasts, 10<sup>6</sup> protozoa, 10<sup>5</sup> nematodes and thousands of assorted worms, mites, collembola and other fauna. In addition, embedded in this numerically huge community are tens of thousands of genetically distinct bacteria and archea, thousands of fungal species and hundreds of protozoan, nematode, worm, insect and arachnid species. The possible *origins* of such diversity are discussed lucidly by Tiedje *et al.* (Chapter 6); here we explore some of the *consequences* in relation to soil organic matter dynamics.

#### Concepts

The functional consequences of biodiversity, and especially of such diversity as prevails in soils, are at present poorly understood from both a theoretical and experimental standpoint. It can be hypothesized that diverse systems are more productive, sustainable and resilient, and that a reduction in biodiversity may result in an impoverishment of ecosystem functions (Giller *et al.*, 1997; Wolters, 1997). The reasoning is based mainly on three mechanisms:

- 1. Repertoire. At the most basic level, for a biologically mediated process to occur, organisms which can carry out that process must be present. A highly diverse system will have a wider repertoire of abilities or a more extensive 'toolkit' that will permit a greater range of functions to be carried out. This is the most basic form of 'functional' diversity and emphasizes the point that if the tool is not available the job will not get done. The diversity of the repertoire in a system is not necessarily related to the taxonomic diversity since one organism may carry out many functions from both a biochemical and physical (e.g. 'ecosystem engineering') perspective.
- 2. Interactions. Organisms influence other organisms to varying degrees, in a positive or negative manner; the more diversity that prevails the more possibilities there are for such interactions to develop. In terms of productivity and sustainability, positive interactions may accelerate nutrient cycling, negative interactions may regulate pathogens. Thus the behaviour of individuals and populations can depend strongly on their biotic context, and community-level controls have been clearly demonstrated in experimental systems (e.g. Janzen et al., 1995; Toyota et al., 1996).
- **3.** Redundancy. The more organisms there are that can carry out a particular process, the more likely it is that if some are incapacitated or removed, the process will not be affected; those that remain simply fill the gap. This is also vaunted as the 'spare wheel' hypothesis (Andrén *et al.*, 1995).

Four main hypotheses linking biodiversity and function currently are postulated; these are summarized in Table 5.1.

Stylized diversity(x)function(y) relationship Hypothesis Basis Referencesa As long as all functional groups are Lawton and Redundancy represented, system functioning is Brown, 1994 independent of diversity Rivet All species make a significant contribution Lawton, to function and that a decrease in diversity 1994 leads to a progressive decline in function Predictable Change in diversity will result in an Mikola and alteration in function, which is predictable Setälä, 1998 change given a priori knowledge of the trophic structure, biomass and principal relationships between the biotic components within a system Idiosyncratic Changes in diversity will result in Lawton. changes in function, but the direction and 1994 magnitude of any response are inherently unpredictable Gas box The biota act as an averaging engine Andrén by-passing the ecological hierarchy; function et al., 1999 is considered as an integrated process

**Table 5.1.** Hypotheses relating to diversity–function relationships.

Initial research claimed to demonstrate benefits to ecosystem function from higher biodiversity (Naeem et al., 1994; Tilman and Downing 1994; Tilman et al., 1997). However, there is a growing body of experimental evidence that the functional characteristics of component species are at least as important as the number of species per se for maintaining critical ecosystem processes (Grime, 1997; Wardle et al., 1997; Bardgett and Shine, 1999). However, it is not known how much biodiversity is needed to ensure continuance of specific soil functions. There is currently little published experimental data which rigorously tests these four hypotheses in below-ground systems, and research from the perspective of soil OM dynamics is particularly sparse.

<sup>&</sup>lt;sup>a</sup>See Mikola and Setälä (1998) for further general discussion.

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#### **Experimental Approaches**

Studies into the relationships between biodiversity and function require the effective experimental control of biodiversity factors, preferably in replicated systems that also permit function to be measured appropriately. This can be achieved in four ways:

- 1. Constructive approaches. Sterile systems are established and prescribed levels of biodiversity built up by the addition of appropriate organisms. The inocula may be comprised of individual species with precisely known characteristics, or may be a less well characterized community of individuals derived from natural habitats. Problems with this approach include the difficulty of establishing slow-growing K strategists, since R strategists will tend to prevail. Building up 'realistic' levels of diversity, particularly in the microbial pool, is also very difficult due to the apparent non-cultivability of many soil prokaryotes and the naturally high levels of diversity in this component.
- 2. Deconstructive approaches. An alternative way to alter diversity is to treat soils in such a way that prescribed groups are selectively removed or destroyed. This can be achieved by application of selective biocides, or based on size exclusion by filtration, sieving or mesh-lined containers.
- 3. Correlative approaches. Natural gradients in diversity can be exploited by taking soils that have inherently different levels of diversity and measuring their functional characteristics. This approach has the drawback that correlation does not necessarily imply cause, and does not offer much possibility of gaining a mechanistic understanding for observed effects. It also represents poor experimental design in that there are likely to be many additional covariates that may confound any diversity effects.
- 4. Modelling. Mathematical models offer a supplementary way of exploring the functional consequences of biodiversity. Models are an experimental tool and require real data for their calibration and validation as well as the concepts that underpin their formulation. Models are best developed in tandem with rigorous experimentation. Most soil-based models that do include diversity elements deal with trophic-level diversity and do not explicitly incorporate intra-group diversity. Indeed, many process models have predicted SOM dynamics successfully without any recourse to the inclusion of species diversity or abundance dynamics. Andrén et al. (1999) review this field and argue that the soil biota can often be regarded as an 'averaging engine', making the analogy of the ecosystem as a gas container. Thus average processes are observed or measured and the contributions of the individual organismal components are filtered out. They also discuss circumstances when this approach may be insufficient.

Some selected examples of the application of these approaches to study the effects of soil biodiversity upon OM dynamics are described below.

## Inputs: Soil Biodiversity Effects Upon Primary Productivity

There are generally no significant direct effects of the soil biota upon primary productivity since the majority of soil organisms are not carbon-fixing autotrophs; an exception may be algal mats in some environments, but they are beyond the scope of this discussion. However, there are many reported effects of trophic diversity upon plant production that are mediated by secondary interactions below ground. These predominantly take the form of interactions between trophic groups in the decomposer food chain, resulting in enhanced nutrient cycling and thus provision of plant-available nutrients. Plant growth has been shown to be enhanced by the presence of grazing nematodes or protozoa in sterile microcosms inoculated with simple bacterial communities, as well as in soil microcosms (see Griffiths, 1994; Griffiths and Bardgett, 1997). Many experiments have been carried out manipulating soil fauna using constructive or deconstructive approaches, and diversity impact upon plant growth has been shown variously to be positive, negative or neutral, depending upon the circumstances of the experiment. For example, Setälä (1995) grew birch and pine trees with mycorrhiza and functionally complex (full set of microbiota and fauna) or simple (microbes alone) detrital food webs. The biomass of both seedlings was significantly higher in the complex system, despite faunal grazing of the mycorrhiza. In a similar experiment, the mycophagous fauna overgrazed the mycorrhiza and this resulted in decreased plant growth (Setälä et al., 1997). In this experiment, greater faunal diversity reduced the growth of mycorrhizal Pinus seedlings only under N-limiting conditions; in N-rich systems there was no effect of varying faunal diversity (Setälä et al., 1997). In a third study, increasing trophic diversity from a single level (nematodes) to two levels (plus enchytreids/dipteran larvae) or three levels (plus mites) increased productivity, but increasing the intra-trophic diversity from one to three species in the second trophic level resulted in a decrease (Setälä et al., 1998). These studies show that the status of a functional group or the consequences of diversity are not pre-determined but depend strongly upon the biotic and abiotic context in which they are operating.

Mycorrhizas are clearly an important component of the soil fungal flora in relation to plant growth, affecting nutrient uptake and competitive ability of the plants with which they associate. In a particularly rigorous and effective constructive-type experimental design, van der Heijden *et al.* (1998) demonstrated how species richness within this functional group affects the diversity of plant communities and attendant productivity. They inoculated macrocosms of sterilized soil with zero, one, two, four, eight or 14 species of arbuscular mycorrhizal (AM) fungi randomly selected from a pool of 23 species. By using ten random iterations of such mixtures, they were certain

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of testing for a genuine AM species richness effect. The macrocosms were then sown with a uniform mixture of 15 characteristic old-field plant species and the resultant plant communities allowed to grow for one season. There was a clear increase in both root and shoot biomass with increasing AM species diversity, as well as an increase in the plant community diversity. Since the diversity component in this experiment was defined precisely and covered a fairly wide numeric range, it was possible to construct well-defined diversity–function response curves, which is very rare in soil diversity studies. The shapes of the curves suggest a rivet-style relationship at low diversity with a tendency to redundancy-style at higher levels. There was a suggestion that shoot and root biomass showed different degrees of sensitivity to changing diversity, with the curves being less steep for roots.

## Turnover and Losses: Diversity Effects Upon OM Decomposition and C and N Mineralization

In an early study on the effects of microbial diversity on soil respiration, Salonius (1981) established soil microcosms containing different levels of bacterial and fungal diversity by reinoculating sterilized soil with progressively diluted non-sterile soil suspensions. The concept here was that more dilute suspensions would contain lower diversity since rare forms would be diluted to extinction sooner. More dilute suspensions would also contain fewer organisms but, by allowing recolonization for 5 months, the resultant communities in the soils comprised a broadly similar number of bacterial and fungal colony-forming units. Diversity was not measured rigorously but indications were that the concept was applicable and species richness declined with dilution. Communities derived from the lower dilutions respired at similar rates and there was a distinctive drop in respiration rate at and beyond certain higher dilutions. This was more pronounced with soils taken from under feather moss than sphagnum moss. Thus there is some evidence that there may be threshold levels of diversity below which OM decomposition is impaired.

As with many of the primary productivity experiments alluded to above, the impact of trophic group diversity upon OM decomposition and C mineralization has been shown to have varying effects, and to be context dependent.

Swift et al. (1998) reiterate that there is little unequivocal evidence of a direct causal linkage between soil biodiversity and nutrient cycling efficiency. They discuss a correlative approach where land use change in Nigeria from bush to cultivation resulted in a decline in abundance and diversity of soil fauna but this resulted in little change in overall decomposition rates of surface litter; here the microbial pool appeared to compensate for the decline in faunal diversity, and is perhaps an unusual example of

redundancy at the inter-trophic level. Microbial communities in soils from field trials where plots had been amended with substrates ranging from manure, pea residues, inorganic N or no addition for 63 years were able to decompose added manure and pea residues with equal efficiency regardless of their OM input history (Fauci and Dick, 1994; Burket and Dick, 1996). Similarly, arable and horticultural soils managed conventionally or organically containing significantly different levels of OM and having different biotic properties decomposed *Medicago* residues equally well (Gunapala *et al.*, 1998). However, such field studies are rather blunt tools for looking at diversity–function relationships.

Using a model inclusive of trophic group diversity and interactions, de Ruiter *et al.* (1993) demonstrated the importance of inter-trophic diversity in relation to N mineralization. They showed that the impact of removal of a trophic group upon N mineralization considerably exceeded the direct contribution of that group. In a detailed comparison of four below-ground food webs, they further demonstrated the importance of interaction patterns and strengths to community stability (de Ruiter *et al.*, 1998).

Coûteaux et al. (1991) grew chestnut trees under ambient or elevated CO<sub>2</sub> atmospheres and obtained litter of contrasting C: N ratios (40 and 75, respectively). They mixed the litter into sterile glass bead microcosms, and inoculated the systems with organisms to provide a factorial series of ascending trophic groups, i.e. native microflora community (< 2 μm); native protozoan community; native nematode community; a collembolan species (Folsomia candida); and an isopod (Oniscus asellus). Total C mineralized from the 'ambient' litter was not affected by the community structure, but there was a trend of increasing mineralization of 'elevated' litter with increasing diversity. There was also a general increase in the amount of leached C where the number of trophic groups increased. The study shows the context dependency of trophic diversity effects, here dependent upon the quality of the substrate being decomposed. It also demonstrates that diversity effects on C losses are not restricted to mineralization to CO<sub>2</sub>. The variability between replicates was noted to be greater in the more diverse systems, which leads to the question of whether there are diversity effects on the consistency of community dynamics and processes. There is evidence for this diversity-stability relationship in terrestrial ecosystems (King and Pimm, 1983; Frank and McNaughton, 1991; Tilman and Downing, 1994) as well as in aquatic microcosms (Naeem and Li, 1997). Even if high species richness does not always play a significant role in maintaining ecosystem processes under normal environmental conditions, it may be important when conditions change (Yachi and Loreau, 1999).

Mikola and Setälä (1998) studied C mineralization from *Pinus* litter supplemented with *Betula* residues. This experiment involved biotically 'complex' and 'simple' systems comprised of three precisely defined trophic levels. The 'complex' system consisted of a mixture of ten bacterial species

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and ten fungal species, a mixture of three bacterivorous and three fungivorous nematode species, and a single predatory nematode. The 'simple' systems had the same microbial communities and predator, but the bacterivore and fungivore levels were restricted to one of the three species present in the complex systems. This design specifically allows the testing of the set of diversity-function hypotheses outlined in Table 5.1. Over 20 weeks of incubation, gross evolution of CO<sub>2</sub> from 8 weeks onward was the same in all systems. During the first 8 weeks, there were sporadic periods when the diverse system produced more CO<sub>2</sub> than the simple systems, and one of the simple systems evolved less CO<sub>2</sub> for the first 3 weeks. Microbial respiration in this particular simple system was also significantly lower than in the others throughout the experiment. The conclusions from this decisive study were that the relationships between OM decomposition and food web diversity were idiosyncratic. This was due to unknown differences in efficiency in resource utilization and vulnerability to predation, unpredictable indirect pathways controlling microbivore biomass and activity, and varying responses to competitive release. The effects of species diversity per se were certainly idiosyncratic, and this was because of subtle differences in the functional attributes of the species, rather than the basic fact that they were taxonomically distinct. This suggests that functional diversity may be a more relevant parameter in relation to soil processes such as OM dynamics than 'raw' species diversity.

Degens (1998) manipulated the diversity of a grassland soil by fumigating it with chloroform and then incubating it for 5 weeks either unamended or following reinoculation with untreated soil. Unfumigated soil provided a 'diverse' control. Species or trophic diversity was not assessed in these soils, but the functional diversity was measured using an *in situ* catabolic potential (ISCP) assay (Degens and Harris, 1997). This technique profiles the ability of the soil community to metabolize 26 defined and contrasting C substrates, and differs crucially from the conceptually similar community-level physiological profiling (CLPP or Biolog®) technique (Garland and Mills, 1991), in that the substrates are added directly to the soil. The functional diversity indices for the three treatments were significantly different and thus provided a gradient of decreasing functional diversity. However, the microbial biomass was significantly lower in fumigated soils and basal respiration significantly lower in fumigated uninoculated soil, which may have confounded diversity effects. There was no consistent relationship between functional diversity and the decomposition of added wheat straw over 100 days, and those effects that were detected were conditional on the moisture potential of the soil, again showing contextual dependency.

Using a deconstructive approach based on partial sterilization, we produced grassland soils containing different degrees of diversity (Griffiths et al., 2000). Soils were fumigated with chloroform vapour for four progressively longer times; it was hypothesized that the longer the fumigation

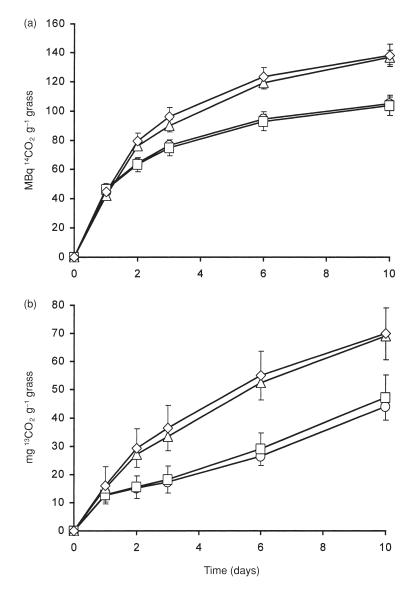
period, the greater and wider the kill of organisms. Biomass reductions were compensated for by allowing recolonization for 5 months after fumigation. Detailed analysis of nematode, protozoan, bacterial and fungal communities confirmed that diversity within and between these trophic groups had been reduced progressively by increasing fumigation time. No larger fauna survived even short fumigation. Microbial biomass and basal respiration in the soils were broadly similar. The ability of these soils to decompose added plant residues was assessed using isotopically labelled material. The two least diverse soils decomposed added grass residue significantly faster than the two most diverse (Fig. 5.1a): this effect was observed in a replication of the experiment using soils sampled from the same field 1 year later (Fig. 5.1b).

#### **Diversity Effects on Resilience**

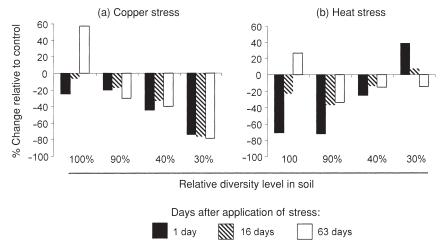
Whilst it can be hypothesized that more diverse systems have greater resilience (Giller et al., 1997), we are unaware of any studies that have studied the phenomenon explicitly. Using the soils described in the previous section, the effects of diversity on the resilience of the decomposition process were studied by subjecting them to additional stresses and measuring their subsequent ability to decompose ryegrass residues (Griffiths et al., 2000). Addition of copper, a persistent stress, to the soils resulted in an immediate decline in their ability to decompose the residue, and there was a distinct trend of greater susceptibility (i.e. greater decline in decomposition relative to the control) to copper with decreasing diversity (Fig. 5.2a). However, after a further 16 days, the decomposition rate in the most diverse soil had virtually reverted to the rate in the control soil, and after 63 days showed an enhanced decomposition ability. The less diverse soils showed no such recovery. Subjecting the soils to a heat shock (37°C for 18 h), a transient stress, produced a different response. Here, the least diverse soils were less susceptible to the stress, but the most diverse soils showed a faster recovery rate and hence greater resilience (Fig. 5.2b). The experimental approach suffers from the fact that fumigation itself represents a stress and may have selected for particular attributes in the surviving and resultant community, but nonetheless demonstrates that a general reduction in biodiversity may have implications for the susceptibility and resilience of soils to stresses.

#### Conclusions

We consider that under most non-extreme conditions, the biodiversity of soils is probably not the primary regulator of soil OM dynamics. This is not because diversity has little impact on C dynamics, but rather that the 352 K. Ritz and B.S. Griffiths



**Fig. 5.1.** Short-term decomposition of isotopically labelled ryegrass in soils containing different levels of biodiversity – see text for details. Relative diversity levels were: 90% ( $\square$ ), 40% ( $\triangle$ ) and 30% ( $\diamond$ ) of the control soil ( $\circ$ ). (a) Year 1, label was <sup>14</sup>C; (b) year 2, label was <sup>13</sup>C. Points show means (n = 4; bars show SE). Source: Griffiths *et al.* (2000).



**Fig. 5.2.** Effects of copper (a) and heat (b) stress upon ability of soils containing different levels of biodiversity to decompose grass residues. Source: Griffiths *et al.* (2000).

prevailing levels of diversity in most systems are sufficiently high so that the repertoire is complete, there is an adequate number of trophic groups with commensurate interaction pathways, and there is a significant degree of redundancy in the decomposer communities. To date, most of the studies demonstrating a significant effect of biodiversity on decomposition only do so under circumstances where diversity is artificially low. It is unclear whether there are actual diversity thresholds below which OM dynamics are impaired, and which occur outside of the laboratory or field experiment. The issue of diversity effects upon resilience also warrant further research. Above all, more high-quality research is needed with innovative experimental designs in order to understand the mechanistic basis of diversity–function relationships.

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## Microbial Catabolic Evenness: 5.1 A Potential Integrative Indicator of Organic Matter Management?

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#### Introduction

The presence of an active and diverse microbial community in soil can be considered a good indicator of a healthy and functioning soil ecosystem. In contemporary ecology, diversity of higher organisms is commonly used as an indicator of ecosystem health because this property of communities is important for the maintenance and stability of ecosystem processes. However, for soil microbial communities, it has not been possible to measure species diversity accurately (Giller et al., 1997; Trevors, 1998). The relative diversity of functions performed by microbial communities is possibly more ecologically relevant to soil processes than species diversity (Zak et al., 1994; Trevers, 1998). Furthermore, in practical terms, it is the diversity of soil functions that is of interest to farmers, agronomists and policy makers. It is unlikely that the functional diversity of soil microbial communities can be determined from species diversity (Giller et al., 1997; Degens, 1999). However, a component of the functional diversity in soil communities can be measured directly using a simple methodology (Degens and Harris, 1997; Degens et al., 2000b).

The implications of soil organic matter management for microbial diversity are poorly understood. Depletion of organic matter in soils can result primarily in loss of microbial nutrient cycling, water-holding capacity and nutrient retention capacity (Gregorich *et al.*, 1994). An understanding of whether these changes have any effect on the diversity of microbial communities or, more importantly, the functioning of these communities, is necessary to evaluate long-term effects of land uses on soils.

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This chapter is a short review of how catabolic diversity measurements can indicate changes in qualitative properties of soil microbial communities that can signal adverse impacts of land uses on soils, particularly through effects on organic C cycling. Throughout, I use examples from the literature and my own unpublished work.

## What is Soil Catabolic Diversity and How can it be Characterized?

The functional diversity of soil microbial communities is broadly considered to include the range and relative expression of activities involved in decomposition, nutrient transformations, plant growth promotion/suppression, plant nutrient acquisition and various soil physical processes influenced by microbes (Giller et al., 1997; Wardle et al., 1999). While these cannot yet be measured easily and completely in soils, it is possible to assess the catabolic diversity of heterotrophic microbial communities involved in decomposition processes. Most soil organisms are heterotrophic. Thus changes in the catabolic functioning (catabolic diversity) of these organisms present a key property for detecting adverse impacts of land uses on soil microbial communities, and possibly soil functioning. It must be emphasized, however, that catabolic diversity is only one component of the overall functional diversity of soil microbial communities.

Current methods to characterize microbial catabolic diversity are based on assessing patterns of substrate utilization (functional profiles). These are determined by adding a specified range of organic compounds to soil in separate bottles and measuring the short-term respiration responses (Degens and Harris, 1997; Degens and Vojvodic-Vukovic, 1999). This approach differs from that used by the Biolog approach (Zak et al., 1994) in that it is not culture-based and relies on more direct measurement of functional responses of the whole soil microbial community. The patterns of substrate use (termed catabolic response profiles or CRPs) can be easily compared by calculation of diversity indices, which can capture some of the characteristics of the functional profiles to enable easy comparison of this information. Catabolic diversity is comprised of catabolic richness, the range of substrates metabolized, and catabolic evenness, the variation between the use of different substrates within a soil. Catabolic evenness (E) can be described readily using the Simpson index:  $E = 1/\sum p_i^2$ , where  $p_i$  = activity of individual substrates as a proportion of total activity induced by all substrates (Degens et al., 2000b). Using this formula, the maximum value of catabolic evenness that can be achieved for a CRP determined using 25 substrates is 25, and large values indicate high levels of diversity.

#### Catabolic Evenness Under Different Land Uses

Catabolic diversity can provide a robust indicator of the condition of microbial communities that is independent of soil type effects. In a study of land uses across 14 soil types, microbial catabolic evenness was consistently greatest in soils under indigenous vegetation (range: 19.7–22.3) or long-term pasture (range: 20.1–23.3), but least in long-term cropped soils (range: 16.4–19.6; Degens *et al.*, 2000b). Soils under a mixed pasture–cropping management had catabolic evenness part way between these extremes (range: 17.7–20.5) but, under pine forest, there was no characteristic level of evenness (Degens *et al.*, 2000b). These broad ranges provide a framework for assessing the diversity in soils without knowledge of soil type or history.

Within soil types, where land use changes from pasture to cereal cropping or greater cropping intensity, there can also be a concurrent decrease in microbial catabolic evenness. A change from pasture to wheat cropping in a sandy loam resulted in a decline in catabolic evenness from 20.2 to 19.2 (P < 0.05; mean of bulked samples from two plots) over a 12-month period. In nearby plots under continuous arable cropping, however, catabolic evenness remained relatively stable (ranging between 18.8 and 19.3; not significantly different at P > 0.05). Similarly, increasing cropping pressure can result in soils with reduced catabolic evenness (Table 5.1.1), relative to levels normally found in pastures (20-22). This indicates that previous land management practices were having a deleterious effect on the biological condition of these soils. When considering the sizes of different organic C pools in the soils in isolation (Table 5.1.1), it is difficult to evaluate the significance of these without comparison with a reference soil under land use with minimal physical disturbance (e.g. long-term pasture). Comparison of these soils with a long-term pasture soil confirms that

**Table 5.1.1.** Microbial catabolic evenness, total organic C, microbial biomass C and potentially mineralizable organic C (mean of five reps) in soil under different land management histories.

Land management history	Catabolic evenness	Total organic C (mg cm <sup>-3</sup> )	Microbial biomass C (mg C cm <sup>-3</sup> )	Potentially mineralizable C (μg C cm <sup>-3</sup> h <sup>-1</sup> )
8 years crop,	19.1 <sup>ab</sup>	459	1.00h	1.040
4 years pasture	19.100	45 <sup>a</sup>	1.26 <sup>b</sup>	1.34°
10 years pasture,				
2 years crop	19.6 <sup>b</sup>	39 <sup>b</sup>	0.74ª	0.33ª
5 years crop,				
2 years pasture	18.7 <sup>a</sup>	48 <sup>a</sup>	0.74 <sup>a</sup>	0.94 <sup>b</sup>
Long-term pasture	21.4 <sup>c</sup>	56ª	1.38°	1.66 <sup>d</sup>

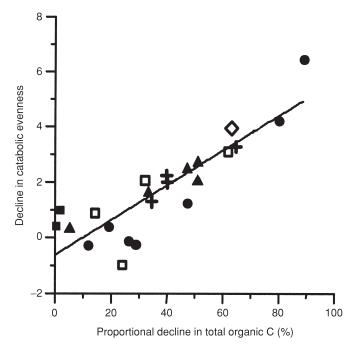
After Sparling *et al.* (2000). Values within columns followed by the same letter are not significantly different (P < 0.05).

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catabolic evenness did indeed indicate that these soils were under stress, as the organic C pools were similarly depleted (Table 5.1.1). Catabolic evenness may be an integrative indicator of biological condition that does not require parallel measurements on reference land uses in order to be interpretable. The absolute value of the catabolic evenness for a soil has the advantage in being readily judged as favourable or unfavourable, unlike other indicators of biological condition, which cannot be evaluated in isolation.

## Relationships Between Microbial Catabolic Evenness and Organic C

Loss of catabolic evenness of soils can be linked with losses in soil organic matter. Only weak generalized relationships exist between organic C pools in soils and microbial catabolic evenness that can be applied across soil types (Degens *et al.*, 2000b). However, comparisons of long-term cropping,



**Fig. 5.1.1.** Proportional decline in total organic C in relation to declines in catabolic evenness for comparisons between pasture and other land uses across a range of soil types ( $r^2 = 0.76$ ; P < 0.01). Pasture was paired with pine forest ( $\bullet$ , Pi), indigenous vegetation ( $\square$ , N), cropping ( $\bullet$ , C), chemical fallow ( $\diamond$ , CF), mixed cropping ( $\bullet$ , MC), or effluent-treated pasture ( $\blacksquare$ , PE). After Degens *et al.* (2000b).

pine forestry, indigenous vegetation and mixed pasture–cropping with long-term pasture (as a reference land use) have shown that losses in soil organic C (relative to pasture) corresponded with losses in catabolic evenness (Fig. 5.1.1), i.e. where a land use results in less organic C relative to pasture, there was also a corresponding lower catabolic evenness in this land use. The same relationships did not apply to measures of more active organic C fractions in soils (microbial biomass C or potentially mineralizable C; Degens et al., 2000b). These comparisons used long-term pasture as a reference land use because this frequently is readily obtained on most soil types. Since most soils under pasture and indigenous vegetation had similarly high levels of catabolic evenness (above), soils where catabolic evenness is less than this are most likely to contain amounts of organic C that are constrained by previous management practices.

Catabolic evenness responds positively to organic matter inputs into soils. Application of 5 t ha<sup>-1</sup> of organic matter as sewage sludge to a sandy soil increased catabolic evenness from 22.8 to 23.1 (P < 0.05, n = 3). Repeated additions of large amounts of organic C with a narrow substrate composition can result in reductions in catabolic evenness over the long term. In a study of soil irrigated with large amounts of lactose (as dairy factory effluent), catabolic evenness had declined from 21.9 in non-irrigated soil to 19.4 (P < 0.05, n = 6) in the irrigated soil. This occurred principally because of much greater response to both lactose and glucose in the irrigated soil (Degens *et al.*, 2000a). Greater catabolic evenness may be an indicator of greater substrate diversity in soils.

#### **Conclusions**

Microbial catabolic diversity presents an alternative tool for monitoring soils to detect adverse impacts of land use systems on soil biological condition. This approach is based on the premise that the presence of an active and diverse microbial community in soil can be considered a good indicator of a healthy and functioning soil ecosystem. The catabolic evenness index provides a versatile indicator to compare the impacts of a range of land uses and organic matter management strategies on different soil types. Furthermore, the index may be useful in discriminating sites where the organic C status of soils is under pressure from previous land use, without extensive knowledge of land use history or needing to compare with benchmark sites on matched soil types. This characteristic of the catabolic evenness index makes the indicator potentially useful as a generic indicator of the impact of land use on soil organic C status and biological condition.

Microbial catabolic diversity provides information on the diversity component of soils that is not encompassed within measures of the sizes of organic C pools such as total organic C, microbial biomass C or 362 B.P. Degens

mineralizable C. The implications of losses of catabolic evenness for soil function remain unresolved. Decreases in microbial catabolic evenness do not cause declines consistently in decomposition functions in soils (Degens, 1998). However, declines in microbial catabolic evenness may cause loss of soil resistance to stress or disturbance. At this stage, it is clear that changes in catabolic patterns represent a useful indicator of changes in the health of soils arising from land management.

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# 'DOK' Long-term Farming 5.2 Systems Trial: Microbial Biomass, Activity and Diversity Affect the Decomposition of Plant Residues

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#### Introduction

The role of biological diversity in natural or man-made ecosystems is poorly understood. Current knowledge, based on ecosystem theories, starts from the idea that ecosystem stability increases with increasing diversity, while the ratio of community respiration to community growth decreases (Odum, 1969). However, this assumption has rarely been proven experimentally, especially in below-ground systems (Wardle and Giller, 1996). Soil microorganisms are an important factor of soil fertility. They are not only responsible for most nutrient-releasing processes, but their presence also indicates soil quality with respect to habitat function. From a functional point of view, biomass and the activity of soil microbiota are important factors of nutrient and energy flow. The metabolic quotient  $qCO_2$  (soil respiration to soil microbial biomass ratio) often has been applied to address the question of soil quality for soil microorganisms. On the one hand, it indicates carbon mineralization, but under steady-state conditions it may also serve as an indicator of the amount of energy used by the community for maintenance of biomass (Wardle and Ghani, 1995). When discussing changes in qCO<sub>2</sub>, however, community shifts or changes in the ratio of fungi and bacteria are often assumed (Anderson and Domsch, 1990) but hardly stated.

Only recently, community structure has been combined with ecosystem data sets to address the question of whether a greater number or 364 A. Fließbach et al.

diversity of species is enhancing soil functionality. Van der Heijden *et al.* (1998) showed that plant productivity increases with increasing mycorrhiza richness, and Tilman *et al.* (1996) showed an increased total resource utilization (decreasing soil nitrate load) with increasing plant diversity. In conclusion, both papers show that species provide functions and that species diversity affects resource utilization in a system.

It was the aim of our investigations to evaluate soil quality in organic farming systems. A major below-ground process is the decomposition of organic residues which may be affected by farming systems. Combining data from holistic measurements with community-level analyses, we wanted to test the hypothesis that systems with a high diversity make better use of the available resources.

#### The 'DOK' Field Experiment

The 'DOK' field experiment (biodynamic, bioorganic, conventional) at Therwil (Switzerland) is maintained by the Swiss Federal Research Station for Agroecology and Agriculture, Zurich (FAL, Reckenholz) and the Research Institute of Organic Agriculture, Frick (FiBL) since 1978 (Mäder et al., 1999). It is an experiment with 96 plots in a Latin square. The systems are managed according to biodynamic (BIODYN), bioorganic (BIOORG) and conventional (CONFYM) agriculture (Table 5.2.1) and receive farmyard manure (FYM) at a rate corresponding to 1.4 livestock units ha<sup>-1</sup>.

A conventional system with exclusively mineral fertilization (CONMIN) and an unfertilized control (NOFERT) are also included. The systems differ mainly with respect to fertilization and plant protection strategy, whilst crop rotation (potatoes, winter wheat, red beets, winter wheat, and 3 years grass—clover ley) and soil tillage were similar.

**Table 5.2.1.** Main differences of the farming systems in the 'DOK' experiment, yield of winter wheat (average of the third crop rotation period 1991–1999), and pH and  $C_{org}$  values from 1996 (different letters indicate significant differences for manured systems, P < 0.05).

Cropping system	FYM	Mineral fertilizer	Synthetic pesticides	Winter wheat yield (t ha <sup>-1</sup> )	pH (KCI)	C <sub>org</sub> (%)
BIODYN	Composted	No	No	4.1 <sup>b</sup>	5.92 <sup>a</sup>	1.65ª
BIOORG	Rotted	P, K <sup>1</sup>	No	4.1 <sup>b</sup>	5.62 <sup>a,b</sup>	1.45 <sup>a,b</sup>
CONFYM	Stacked	Yes	Yes	4.6a	5.40 <sup>bc</sup>	1.43 <sup>ab</sup>
CONMIN	No	Yes	Yes	4.8 <sup>a</sup>	5.01 <sup>c</sup>	1.41 <sup>ab</sup>
NOFERT	No	No	No	3.0°	5.20 <sup>c</sup>	1.31 <sup>b</sup>

<sup>&</sup>lt;sup>1</sup>Rock phosphate, K<sub>2</sub>SO<sub>4</sub>, when deficient.

We used chloroform fumigation extraction to measure microbial biomass C (Vance *et al.*, 1987). CO<sub>2</sub> evolution from soils was measured according to Jäggi (1976) with pre-incubated samples. Solid-state  $^{13}$ C nuclear magnetic resonance spectra ( $^{13}$ C-NMR) were measured on a Bruker ASX-400 spectrometer (9.4 Tesla, 100.6 MHz) with crosspolarization and magic angle spinning (1.0 ms, 4000 Hz). Samples were pre-treated with 10% hydrofluoric acid for 24 h in order to remove paramagnetic substances and increase C concentration (Schmidt *et al.*, 1997). Light fraction organic matter was extracted from soils by sieving and density fractionation in Ludox ( $\rho < 1.13$ ) (Hassink, 1995). A substrate utilization test was performed according to Garland and Mills (1991) using GN microplates with 95 different C sources (BIOLOG Inc., Hayward, California). A functional diversity index was calculated from the intensities of substrate utilization and the number of substrates used (Fließbach and Mäder, 1997).

#### **Results and Discussion**

Crop yields were lower in the organic as compared with the conventional farming systems by 20% for winter wheat, and up to 40% for potatoes. Soil organic matter and pH were markedly higher in the BIODYN system compared with the conventional and the unmanured systems (Table 5.2.1). We suggest these findings to be an effect mainly of the different fertilization regimes that fertilize the plant either directly or via soil mineralization processes.

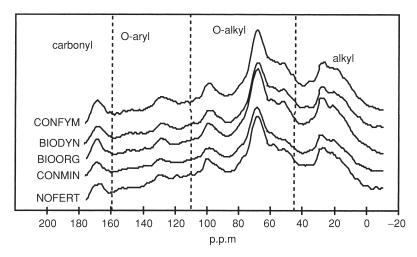
We expected to find changes in soil organic matter composition due to the long-term application of manure with different degree of maturity. <sup>13</sup>C-NMR spectra, however, did not show any significant differences between the treatments (Fig. 5.2.1) due to the variation of field replicates. Uniformity of organic matter from different farming systems has already been stated by Randall *et al.* (1995). NMR from whole samples, therefore, does not seem to be sufficiently sensitive to detect subtle differences in soil organic matter composition due to farming systems.

Nevertheless, differences in soil organic matter quality were found with respect to the amount of light fraction particulate organic matter (POM,  $\rho < 1.13$ ) (Fließbach and Mäder, 2000). The light fraction POM was found to reflect the amount of crop residues that remained in the field, but on the other hand the light fraction and microbial biomass showed an inverse relationship. With a high microbial biomass, the amount of the light fraction was low, indicating that microbial biomass is a regulator of organic residues in soil, whilst the light fraction may indicate the degree to which soil organisms are able to decompose residues. We assume, therefore, that the quotient of microbial biomass ( $C_{\rm mic}$ ) to light fraction POM

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( $C_{LF}$ , i.e. the microbial substrate) serves as an indicator of the catabolic potential of the soil microbial community (Table 5.2.2).

The catabolic potential of soil microorganisms is important with respect to the decomposition of the organic residues. In an incubation experiment with  $^{14}\text{C}$ -labelled plant material, mineralization ( $^{14}\text{CO}_2$ ) and microbial incorporation ( $^{14}\text{C}_{\text{mic}}$ ) were followed over 6 months. In BIODYN soils, the total metabolized material ( $^{14}\text{CO}_2 + ^{14}\text{C}_{\text{mic}}$ ) was > 10% higher than in the compared conventional and unmanured soils (Fließbach *et al.*, 2000) (Table 5.2.2). This suggests a more complete decomposition of the applied material and finds an equivalent in the above-mentioned quotient of microbial biomass to light fraction C. The hypothesis that the differences



**Fig. 5.2.1.** <sup>13</sup>C CP MAS NMR spectra of whole soil samples pre-treated with hydrofluoric acid (each line represents the mean of the four field replicates).

**Table 5.2.2.** Quotient of microbial biomass and light fraction organic matter ( $C_{\text{mic}}$ :  $C_{\text{LF}}$ ) and the percentage of the applied labelled plant material mineralized ( $^{14}\text{CO}_2$ ) and microbially assimilated ( $^{14}\text{C}_{\text{mic}}$ ). Figures are the mean of four field replicates (different letters indicate significant differences, P < 0.05).

	_		% <sup>14</sup> C metabolize	d
	$C_{mic}$ : $C_{LF}$ (mg g <sup>-1</sup> )	<sup>14</sup> CO <sub>2</sub>	$^{14}\mathrm{C}_{\mathrm{mic}}$	$^{14}\text{CO}_2 + ^{14}\text{C}_{\text{mic}}$
BIODYN	2.27 <sup>a</sup>	63.3ª	9.4ª	72.7 <sup>a</sup>
BIOORG	1.54 <sup>a,b</sup>	ND	ND	ND
CONFYM	1.13 <sup>bc</sup>	56.7 <sup>b</sup>	5.6 <sup>b</sup>	63.3 <sup>b</sup>
CONMIN	0.64 <sup>c</sup>	55.1 <sup>b</sup>	5.4 <sup>b</sup>	60.5 <sup>b</sup>
NOFERT	0.70 <sup>bc</sup>	54.7 <sup>b</sup>	6.6 <sup>b</sup>	61.3 <sup>b</sup>

in decomposition are due to a higher functionality in the 'DOK' field trial soils was tested by applying a community-level approach first described by Garland and Mills (1991) using BIOLOG<sup>TM</sup> microplates. Patterns of substrate utilization differed significantly between soils of conventional and organic farming systems in spring but not in autumn samples (Fließbach and Mäder, 1997). A functional diversity index obtained from the substrate utilization patterns of spring samples in winter wheat plots showed higher values in BIODYN soils than in conventional soils (Table 5.2.3), indicating a higher potential of catabolic capabilities.

With respect to a functional ecological approach, we combined the holistic soil biological parameter  $qCO_2$  and the diversity of utilized substrates. Among the soils of the 'DOK' field experiment we found a significant inverse relationship of  $qCO_2$  and functional diversity (Fig. 5.2.2). With increasing functional diversity, the energy needed to maintain the level of microbial biomass became less, indicating a resource utilization more efficient with respect to microbial growth, and a greater complexity of the microbial food web. Combining holistic soil functions with community-level analysis may provide useful tools for interpretation of soil quality and soil as a microbial habitat.

#### Conclusions

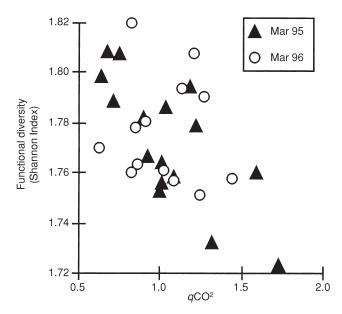
Even though total soil organic matter differs between organic and conventional agricultural systems, no substantial changes in the chemical composition as determined by solid state <sup>13</sup>C-NMR were detectable. Obviously, the carbon content of the soils was too low and quality changes in the active pool of SOM too small for a sensitive detection of differences with this technique.

Labile soil organic matter pools (soil microbial biomass, light fraction particulate organic matter) indicated distinct changes in soil organic matter

Table 5.2.3.	Soil microbial biomass ( $C_{mic}$ ), $C_{mic}$ : $C_{org}$ ratio, $qCO_2$ and functional diversity as
Shannon inde	x. Figures are the mean of four field replicates and are composed of samples from
March 1995 a	nd 1996 (Tukey's test results for manured systems only).

	$C_{mic}$ ( $\mu g g^{-1}$ )	$C_{mic}$ : $C_{org}$ (mg g <sup>-1</sup> )	$q$ CO $_2$ ( $\mu$ g CO $_2$ -C mg $^{-1}$ C $_{ m mic}$ )	Shannon (H)
BIODYN	355ª	2.20 <sup>a</sup>	0.795 <sup>a</sup>	1.792ª
BIOORG	271 <sup>b</sup>	1.91 <sup>b</sup>	1.132 <sup>b</sup>	1.778 <sup>a,b</sup>
CONFYM	231°	1.63 <sup>c</sup>	1.173 <sup>b</sup>	1.756 <sup>b</sup>
CONMIN	185°	1.35°	1.364 <sup>b</sup>	1.798 <sup>b</sup>
NOFERT	214 <sup>c</sup>	1.56 <sup>c</sup>	1.205 <sup>b</sup>	1.803 <sup>ab</sup>

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**Fig. 5.2.2.** Functional diversity as a function of the metabolic quotient  $qCO_2$  (n = 3).

dynamics. We suggest that soil microbial community structure changes as a result of the long-term organic or conventional farming and, furthermore, suggest these changes to be the main reason for differences in decomposition of organic matter. The hypothesis that community stability increases with an increase in diversity is confirmed by our results.

Organic farming systems comprise management practices that change soil organic matter cycling processes. Since the strategies of fertilization and plant protection are the main differences of the 'DOK' farming systems we suggest these factors to be the driving variables for the changes observed within the microbiota and in organic matter quality in soils from organic and conventional farming systems.

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### Comparison of Phenotypic, Functional and Genetic Diversity of Bacterial Communities in Soils

**5.3** 

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#### Introduction

Microorganisms may constitute < 0.5% of the total soil mass, but are responsible for the majority of nutrient cycling and exert a major influence on soil fertility (Tate, 1986). Microbial biodiversity has become an important issue with the realization that changes in microbial communities within the soil have implications for its overall health. There is concern over the long-term environmental effects of industrialization and urbanization during the 19th and 20th centuries, including the possibility of a decline in the general quality of soils. Soil is not only important as a means of supporting crop production but is also an essential natural resource, which should be managed for future generations. Some measure of soil quality is necessary to determine whether soil is being degraded prior to irreversible larger scale damage, and biodiversity could be an appropriate indicator. Some microbial functions are essential for important soil processes, but it is not clear whether the diversity of metabolic responses, or the genetic diversity of the population, are the most relevant indicators of soil quality.

In this chapter, methods to study soil microbial populations were compared using soils amended with either farmyard manure or sewage sludge over a 30-year period. Because urban sewage sludge contains heavy metals, the effects of sub-acute levels on soil microorganisms are also considered. Previous studies have considered losses of microbial functions resulting from metal contamination in soil with little consideration of the effects on the biodiversity of the microbial communities (Giller *et al.*, 1998). The loss of functional and/or genetic diversity in soil could have

direct economic consequences for agriculture, particularly if diversity provides a buffer against environmental changes. Biodiversity should be preserved in its present state as an insurance for future generations, until its significance is more fully understood (Bengtsson, 1998).

#### **Materials and Methods**

#### Site description and soil analysis

The Market Garden Experiment at Woburn Experimental Farm, Woburn, UK is a sandy loam (typic Udipsamment), pH 6.5 with 10% clay, 51% coarse sand. Two plots were sampled: the farmyard manure (FYM) plot, which received 10.4 t organic matter ha<sup>-1</sup> year<sup>-1</sup>; and the sewage sludge (Sewage) plot, which received 16.4 t of metal-contaminated organic matter ha<sup>-1</sup> year<sup>-1</sup> between 1942 and 1961. The plots are under grass and have similar C: N ratios. Soil was sampled with a 10 mm corer to the depth of 250 mm, and ten replicate samples were taken from each plot, bulked and sieved through a 2 mm sieve. Microbial studies were performed on fresh soil; samples for other analyses were stored at 4°C. Total carbon, nitrogen and heavy metal contents were measured as described previously (Brooks and McGrath, 1984; McGrath and Cunliffe, 1985).

#### Catabolic diversity of the culturable bacteria

Using the BIOLOG<sup>TM</sup> system (Garland and Mills, 1991), a 1 g sub-sample of the bulked soil was taken and the diluted soil extracts were exposed to 95 different carbon substances. The capacity of the soil microbes to metabolize these compounds was measured by colour development in comparison with a control containing only water. This provided C substrate utilization data which were used to assess any functional changes in culturable bacterial communities by using the Shannon Weaver equation (Magurran, 1988).

#### Population size and phenotypic diversity

Microbial counts were obtained using standard procedures (Alef and Nanninpieri, 1995). Diluted soil suspensions (obtained as above) were plated out on a range of selective media: 1/10 tryptone soya agar (TSA), general selection for fast-growing heterotrophic bacteria; potato dextrose agar (PDA), fungi; yeast mannitol (YM), slower growing heterotrophs e.g. rhizobia; *Pseudomonas* selective agar (PSA), fluorescent pseudomonads;

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McConkey, putative enterobacteria and nutrient agar (NA) heated to 80°C for 30 min, spore-forming bacteria. All plates were incubated at 28°C.

#### **Bacterial genetic diversity**

Genetic shifts within bacterial communities in both plots were examined using ERIC-PCR (enterobacterial repetitive intergenic consensus-polymerase chain reaction) fingerprinting of bacteria (de Bruijn *et al.*, 1992). ERIC-PCR fingerprints tend to be strain specific and indicate diversity at a sub-species level. The population chosen for analysis, selected on PSA medium, was an agriculturally important group implicated in healthy plant growth, i.e. the fluorescent pseudomonads. All colonies isolated were fingerprinted and analysed. The bacterial ERIC-PCR fingerprints were run on the ABI DNA Sequencer system using Genescan™ software and analysed via the Microsoft Access database program. This program converts the ERIC-PCR fingerprint data into a suitable format so that the bacterial isolates can be compared, and assigned into groups. The PHYLIP Version 3.57 software package UPGMA (Unweighted Pair Group Method using Average linkage; Felsenstein, 1996) was used to display the relationship between the bacterial isolates.

#### **Results and Discussion**

Both Sewage and FYM sites have similar C and N contents (Table 5.3.1), and the C: N ratios are 11.2 and 11.5, respectively. The levels of heavy metals are greater in the Sewage-treated plot compared with the FYM plot (Table 5.3.1) with respect to all the metals tested, although all values (except cadmium) are below the UK limits.

Analysis showed these soils to be similar in all respects except for the heavy metal content resulting from the past sewage sludge applications.

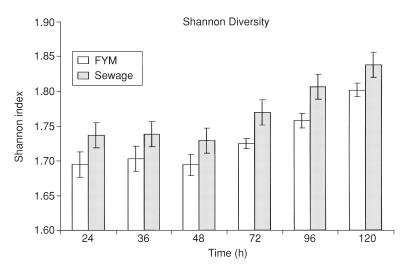
Table 5.3.1.	Percentage carbon, nitrogen and heavy metal content (p.p.m.) of soils. All errors are
± standard de	viation in parentheses after the mean.

Sewage	FYM	Element	Sewage	FYM	UK limits
0.185 (0.002)	0.163 (0.001)	Zinc	234.1 (0.79)	94.5 (1.1)	300
		Copper	69.70 (0.52)	22.7 (0.25)	135
2.141 (0.031)	1.821 (0.011)	Nickel	23.90 (0.36)	14.6 (0.32)	75
		Cadmium	6.35 (0.03)	1.44 (0.15)	3
		Chromium	99.10 (1.20)	43.5 (0.50)	400
		Lead	75.90 (0.52)	34.5 (0.59)	300
	0.185 (0.002)	0.185 (0.002)	0.185 (0.002)	0.185 (0.002)	0.185 (0.002)

The heavy metals did not appear to affect crop yields although the numbers and diversity of one group of bacteria, the rhizobia, are known to have been affected (Hirsch *et al.*, 1993). With changes in EU regulations concerning the disposal of sewage sludge at sea, its addition to agricultural soil is likely to increase, with the possibility of a resulting increase in metal contamination. Thus, this change in environmental policy and agricultural practice may increase the metal content, and consequently reduce soil quality.

The catabolic diversity of culturable bacteria as determined via BIOLOG<sup>™</sup> assays (Fig. 5.3.1) reveals a difference between the two soils. The Shannon diversity is higher in the Sewage plot, which indicates more a more diverse population using more varied carbon sources compared with the FYM plot.

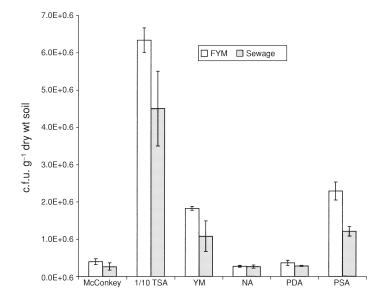
The number of culturable microbes estimated on agar media from the bacterial or fungal colony-forming units (c.f.u.) g<sup>-1</sup> soil were consistently higher in the FYM plot compared with the Sewage plot (Fig. 5.3.2). This is consistent with the finding that the total microbial biomass carbon was higher in the FYM plot than in the Sewage plot (Brookes and McGrath, 1984). None of the media showed large differences between the two plots, indicating that overall the community structure was not greatly affected. The most noticeable increase was seen in the pseudomonads from FYM on PSA medium, where the c.f.u. were tenfold higher than those from the Sewage plot. Since this group contains many root-colonizing bacteria implicated in healthy plant growth (Rovira *et al.*, 1965), is important for soil fertility, the genetic diversity of the population was investigated.



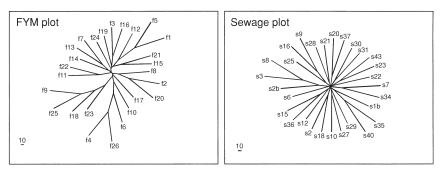
**Fig. 5.3.1.** Catabolic diversity of C utilization in the soil, as Shannon index for two soils receiving FYM or sewage sludge.

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A number of pseudomonad isolates were taken from each plot for genetic fingerprinting. This enabled comparison of the genetic structure of the group in the two plots, to look for shifts that might be due to the different organic carbon and heavy metal inputs. Fingerprint analysis, taken at one time point, showed that the individual isolates from the two plots were different. It also indicated that the population structure in the two groups was different, with some dominant groups apparent in the FYM plot whereas there were none in the Sewage plot (Fig. 5.3.3). Thus, the Sewage



**Fig. 5.3.2.** Microbial counts as colony-forming units (c.f.u.  $g^{-1}$ ) on different selective media for two soils receiving either FYM or sewage sludge. Error bars are standard deviations of the mean of five replicates on each medium.



**Fig. 5.3.3.** Comparison of genetic diversity and population structure in two soils receiving either FYM or sewage sludge. Analysis of 26 isolates (FYM plot) and 28 isolates (Sewage plot).

pseudomonad population appears to have greater diversity compared with the FYM plot. However, diversity may be masked in the FYM population by the most abundant isolates belonging to relatively few related fingerprint groups, which will dominate both the BIOLOG<sup>TM</sup> assays and the plate counts.

#### Conclusion

There was clearly an effect of the different organic inputs on the two plots. The Sewage plot had a lower overall microbial population but had an apparent increase in both the catabolic diversity of fast-growing culturable bacteria measured by BIOLOG<sup>TM</sup> and the genetic diversity of the pseudomonad population. The results indicate a real difference in the population structure of part of the bacterial communities of the two Woburn plots, probably due to the presence of heavy metals, although it is also possible that there were subtle effects arising from the different organic matter inputs. This preliminary investigation demonstrates how valuable multidisciplinary approaches are for studying soil quality and that different approaches used to study the dynamics or diversity of the soil should be compared. These initial results are promising as they indicate that there is some agreement between different approaches. However, further research is required to evaluate the significance of the results obtained and to determine what are the most relevant microbial indicators of soil quality.

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# Organic Matter in Restored Soils as Affected by Earthworms and Land Use

5.4

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#### Introduction

Early evidence (Hunter and Currie, 1956) has suggested that soil restored after opencast mining for coal lacks stable aggregation, has low organic matter levels and altered organic composition. More recently, Malik and Scullion (1998) showed that restored soils had particularly low carbohydrate contents compared with similarly managed undisturbed soils. This later study also indicated differences between restored and undisturbed soils in relationships between microbial C, respiration and total soil carbon as found in other studies on restored soils (Insam and Domsch, 1988; Gilsotres *et al.*, 1992).

Earthworm abundance is drastically reduced by soil handling practices (Rushton, 1986), and full recovery may take in excess of 20 years (Scullion, 1994). Malik and Scullion (1998) showed that earthworms increased aggregation and affected the composition of organic matter in restored soils. The present study reports on the recovery of organic matter levels and cycling in restored soils, relating this process to aggregation, earthworm populations and land use.

### Methods, Sites and Experimental Details

Data are presented from three studies investigating different aspects of soil rehabilitation after opencast mining. All sites were sampled during 1993/94. In the first study, changes in organic content and composition in

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restored soils under grassland management were related to the development of stable aggregation in these soils. A second experiment evaluated the role of earthworms in these changes, again under grassland management. The third study assessed differences in soil C cycling between some of the grassland sites used in the first study and similar soils under woodland. All sites were in the same location (UK Grid Ref. SN259214) in South Wales and were part of a long-term research programme (Scullion, 1994). The region had high rainfall, with both undisturbed and restored clay loam soils having poor to imperfect drainage. Soil conditions at replacement and management regimes were recorded carefully as part of the overall research programme, ensuring that comparisons between sites could be made with confidence.

In the first study, soils on undisturbed (UD) land and on land restored for 9 (R9) and 21 (R21) years were compared in terms of their organic composition and carbohydrate contribution to stable aggregation. It was anticipated that as restored soils aged, their organic content and composition would approach that of UD soils. Earthworm activity and resulting physical protection of organic matter within stable aggregates were expected to mediate this process (Scullion, 1994). Five fields were selected including two examples of different (9 and 21 years) stages in the recovery process; an undisturbed field was included to represent the end point of this process. The two fields from each restoration phase had different nutrient inputs, organic returns and earthworm populations. Both fields on the 9-year site received 100 kg N as urea and 8 t of poultry manure ha<sup>-1</sup> year<sup>-1</sup>, and had low earthworm populations. However, the high input field (R9H) was treated with 100 t of sewage sludge (7.1% total C, 0.6% total N, 0.8% total  $P_2O_5$  as applied) ha<sup>-1</sup> at restoration; the low input field (R9L) received only the basal fertilizer dressing at this stage. On the older site, the high (R21H) input field was treated with 100 kg N as urea and 8 t poultry manure ha<sup>-1</sup> year<sup>-1</sup>, whilst the low (R21L) input field received half of each of these annual inputs; earthworm populations were moderate on the high input and large on the low input field. The undisturbed field, with the largest earthworm population, received nutrient inputs similar to those of R21H. All fields were under grass-clover leys and, with the exception of R21H which was cut for silage, were grazed by sheep. Differences were expected between each pair of fields on restored soils in the amount and quality of organic matter, and in associated soil structural development.

On the more recently restored (R9) site, the influence of earthworms on organic content, composition and aggregation was evaluated by comparison of control and earthworm input plots (Scullion, 1994). This experiment aimed to assess the influence of earthworm activity on changes in soil organic matter and aggregation in restored soils. Four 'blocks', including one control and one earthworm input plot (each 400 m²) were designated prior to the introduction of earthworms, with treatments

allocated at random; control and input plots within a 'block' were located 40 m apart. Earthworm inoculations were carried out during the second year following soil replacement and aimed to create a population typical of undisturbed pasture over a short period of time. Inputs were equivalent to almost 70 individuals m<sup>-2</sup> (for details, see Scullion, 1994) and were representative of the full range of species in local pastures. Field drainage and general management (grazing of clover–grass swards by sheep, with an annual topdressing of 8 t fresh weight of poultry manure ha<sup>-1</sup> year<sup>-1</sup>) were considered (Scullion, 1994) to favour earthworms.

In the fourth year after soil replacement, populations on input plots were similar to those on adjacent, undisturbed pasture (Marashi, 1995) and included a high proportion of the deep burrowing species *Lumbricus terrestris* (L.) and *Aporrectodea longa* (Ude). Prior to the final earthworm survey in year 6, populations on control plots were markedly lower than those of input plots and were dominated by the surface-dwelling, early colonizing species *Lumbricus rubellus* Hoff. and *Allolobophora chlorotica* Sav.

In the final study, differences in soil microbial organic C indices under grass and woodland were assessed on the three land types described in the first study. Woodland soils were sampled in coarse grass under *Alnus glutinosa*, the most productive species in the woodland mix (Scullion, 1994). Trees were planted in the first year after restoration on former opencast land and at the same time as the R21 site on undisturbed land. During the first five or more years after planting, when tree growth was slow, organic returns would have been markedly higher under grassland. Carbon cycling was also favoured under grassland by the regular return of faeces during grazing. These effects led to markedly larger earthworm populations on grassland compared with woodland sites (Scullion, 1994). Woodland areas adjacent to the high input grassland sites used in the first study were selected for this comparison. These grassland and woodland sites were sampled concurrently, with sampling restricted to the surface 10 cm where soil changes were expected to be more pronounced.

Between six and ten replicate samples were taken from each area in the above investigations. Mean values of each measured parameter for different soil groups were compared by one-way analysis of variance, using the Statgraphics version 7 (Statistical Graphics Corporation, 1993) statistical package.

### Soil Analyses

Organic contents (LOI) were measured by loss on ignition (Ball, 1964) at 400°C. Total carbohydrates (CARB) were extracted using strong acid hydrolysis followed by refluxing at lower acid concentrations (Cheshire *et al.*, 1983) and measured by the anthrone method (Brink *et al.*, 1960).

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Non-carbohydrate (NCARB) content was calculated as the difference between percentage loss on ignition and percentage total carbohydrates. Biomass carbon was estimated by fumigation extraction (Vance *et al.*, 1987), with C in extracts determined on a Shimadzu Organic Carbon analyser (Model TOC-5050). Respired  $CO_2$  was measured (Sparling, 1981) after incubation at  $20^{\circ}C$  for 2 h using gas chromatography (PYE-Unicam series 104). For organic C ratios, loss on ignition (for  $C_{org}$ ), extractable biomass carbon ( $C_{mic}$ ) and respiration ( $C_{resp}$ ) were converted to carbon equivalents using standard factors (Vance *et al.*, 1987; Howard and Howard, 1990).

Aggregate stability was measured after shaking air-dry soil (< 2 mm) in water for 1 h (Scullion, 1994). The percentage of silt + clay and clay remaining in aggregates (> 63 and > 2  $\mu$ m, respectively) was calculated by reference to total silt + clay and clay (MAFF, 1986). This procedure was also carried out for soils pre-treated with sodium chloride or sodium periodate, with the difference between pre-treatments taken as the contribution of carbohydrates to stable aggregation (Cheshire *et al.*, 1983); periodate treatment was found to destroy between 95 and 98% of carbohydrates (Malik, 1996).

#### **Results and Discussion**

#### Organic matter and aggregation

There was a consistent increase in all organic matter fractions in restored soils between years 9 and 21 (Table 5.4.1), with the latter soils having higher total organic levels near the surface than UD soils. Since soils immediately following restoration had  $\sim$ 4% LOI (Scullion, 1994), the average annual gain in soil organic matter, allowing for changes in bulk density, can be estimated at > 3000 kg ha<sup>-1</sup>.

Table 5.4.1.	Mean values for organic matter fractions in undisturbed (UD) and restored (R) soils.
--------------	--

_	0-7.5 cm			7.5-15 cm		
Sites	%LOI	%CARB	%NCARB	%LOI	%CARB	%NCARB
UD	10.9	1.66	9.3	7.6	0.92	6.7
R21H	13.2	1.05	12.2	5.6	0.21	5.4
R21L	11.3	0.84	10.5	5.4	0.19	5.1
R9H	7.2	0.74	6.4	4.4	0.12	4.3
R9L	6.8	0.73	6.1	4.1	0.11	4.0
LSD 5%	1.58	0.25	1.33	0.85	0.13	0.72

Most of the increase in LOI following replacement of restored soils, and of increases with higher inputs on restored soils of similar age, occurred at the upper sampling depth and was accounted for by changes in the non-carbohydrate fraction. As a result, carbohydrates made up a markedly smaller proportion of the organic fraction in restored compared with UD soils. This trend was more pronounced at the lower depth where carbohydrates accounted for < 4% of organic matter in restored compared with > 12% in UD soils.

One possible explanation for this difference in organic matter composition was the low level of macroaggregation in restored soils (Table 5.4.2), even 21 years after restoration. Several studies (e.g. Eriksen et al., 1995) have identified stable aggregates as important sites of physical protection for labile organic material. In stabilizing aggregates, this organic matter may itself be protected from microbial degradation.

Without pre-treatment, UD soils had higher stability than restored soils for both aggregate sizes and depths. There was no general improvement in aggregate stability in restored soils between years 9 and 21, although silt + clay stability did increase at the upper sampling depth. Periodate-sensitive aggregate stability was markedly higher in UD soils, although the difference from restored soils was less pronounced for clay-sized aggregates. The contribution of carbohydrates to aggregation was particularly low for silt + clay aggregation on the 9-year restoration at both depths and on the 21-year restoration at the lower depth. The sludge-treated soil (R9H) had greater total and periodate-sensitive stability, particularly at depth, than the comparable (RHL) restored soil, but the older restored soils had generally similar stability characteristics.

Earthworm surveys (Scullion, 1994) indicated populations decreasing in order of UD > R21 > R9. These differences may partially explain

	0–7.5 cm				7.5–15 cm			
Sites	%CI stability	%CI periodate stability	%Z + Cl stability	%Z + CI periodate stability	%CI stability	%CI periodate stability	%Z + Cl stability	%Z + CI periodate stability
UD	91.4	30.5	42.7	25.6	88.9	38.7	36.4	23.9
R21H	84.4	19.8	30.9	11.9	55.0	33.3	13.4	5.8
R21L	74.8	22.9	28.6	12.9	52.8	33.8	13.9	5.9
R9H	78.8	27.7	18.8	1.0	69.1	39.6	12.9	7.3
R9L LSD 5%	72.5 3.03	25.2 5.66	25.2 1.47	1.1 6.2	56.0 9.94	31.1 6.21	8.1 5.61	3.1 4.97

**Table 5.4.2.** Total and periodate-sensitive clay (CI) and silt + clay (Z + CI) aggregate stability in undisturbed (UD) and restored (R) soils.

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variations in carbohydrate content as earthworm cast materials contain high levels of carbohydrates (Altemuller and Joschko, 1992). In the inoculation trial on restored (R9) land, earthworms increased soil carbohydrates and aggregate stability (Table 5.4.3) by similar proportions at both sampling depths. Clay showed a trend similar to that for silt + clay stability, so only the latter data are given. On earthworm input plots, stability was most closely related to carbohydrate contents and inputs also decreased consistently  $C_{resp}$ :  $C_{org}$  ratios (Scullion and Malik, 2000).

Carbohydrates accounted for 16.0 and 16.1% of the total organic content of the earthworm input soils at the upper and lower sampling depths, respectively, compared with 10.6 and 7.7% in the control. Input soils 9 years after their replacement showed proportions of carbohydrates similar to those of UD soils but somewhat lower levels. Input soils had higher levels of stability and of carbohydrates than corresponding soils on the older (R21) site.

#### Carbon indices

Ratios of different soil C fractions provide an indication of the status of carbon cycling in soils and have been suggested as an index of soil recovery (Insam and Domsch, 1988). Data in Table 5.4.4 show that for grassland, but not woodland, there was a significantly higher proportion of soil C as microbial C in undisturbed compared with restored soils; restored soils of different age did not differ consistently in this respect. Within similar management regimes, UD soils generally had lower  $C_{resp}$ :  $C_{mic}$  and  $C_{resp}$ :  $C_{org}$  ratios compared with restored soils, perhaps indicating respectively less stress on microorganisms and greater protection of organic matter. It is likely that the high labile organic fraction in UD soils was linked to similarly high aggregation levels. Within restored soils, there was little difference in  $C_{resp}$ :  $C_{mic}$ , but the more recently restored soils had markedly higher

**Table 5.4.3.** Organic fractions and aggregation (R9 site) as affected by earthworm inputs 8 years previously.

	0-7.5 cm			7.5-15 cm				
Treatment	%LOI	%CARB	%NCARB	%Z + CI stability	%LOI	%CARB	%NCARB	%Z + CI stability
Control Worm input	8.49 8.00	0.90 1.28	7.59 6.72	21.9 33.7	3.91 4.36	0.30 0.70	3.61 3.66	13.1 26.9
Significance level (P =)	0.2189	0.0001	0.2188	0.0004	0.0993	0.0000	0.0993	0.0000

Sites	$C_{mic}$ : $C_{org}$ (mg C g <sup>-1</sup> C)	$\label{eq:constraints} \begin{split} & C_{\text{resp}} : C_{\text{mic}} \\ & (\text{mg CO}_2\text{-C g}^{-1} \text{ C h}^{-1}) \end{split}$	$C_{resp}$ : $C_{org}$ (mg $CO_2$ -C $g^{-1}$ C $h^{-1}$ )	
UD grass	35.8	14.1	0.086	
UD wood	28.8	22.8	0.112	
R21 grass	19.0	20.3	0.096	
R21 wood	27.1	28.2	0.193	
R9 grass	27.3	19.8	0.180	
R9 wood	26.7	28.9	0.279	
LSD 5%	3.81	5.62	0.034	

**Table 5.4.4.** Mean ratios of carbon indices (0–10 cm depth) in undisturbed (UD) and restored (R) soils.

 $C_{resp}$ :  $C_{org}$  ratios, perhaps reflecting their particularly low aggregation levels. There was no consistent management difference for the  $C_{mic}$ :  $C_{org}$  ratio, but woodland soils had markedly higher  $C_{resp}$ :  $C_{mic}$  and  $C_{resp}$ :  $C_{org}$  ratios irrespective of land type. These latter differences may be linked to lower woodland soil aggregation levels and earthworm populations (Scullion, 1994).

#### Conclusions

Whilst total organic matter levels had recovered 21 years after soil restoration, differences from undisturbed soils in organic composition and dynamics remained. Variations in the degree of physical protection of labile organic fractions in stable aggregates offer one explanation for these differences. Earthworms influenced both aggregate and organic stabilization processes. A common feature of restored soils was the higher than expected respiration at any given microbial biomass or organic matter content. This feature may arise because of the limited physical protection offered to organic matter and may explain the low carbohydrate contents of restored soils. Increased inputs did not always promote soil recovery. Land use differences were confounded with tree age. Nevertheless, restored and undisturbed soils showed, for the most part, similar differences in carbon indices between grassland and woodland.

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## Rhizosphere Effects on Soil Microbial Biomass Size and Turnover in a Soil of High and Low Fertility

# 5.5

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#### Introduction

Various methods for studying the soil microbial biomass (SMB) in the rhizosphere and its influence on soil organic matter have been developed. Different measures, such as direct bacteria counting (e.g. Rovira *et al.*, 1974; Badalucco *et al.*, 1996), ATP content (Youssef *et al.*, 1989), enzyme activity (Badalucco *et al.*, 1996) or fumigation incubation (Helal and Sauerbeck, 1986) have been used for determining SMB in the rhizosphere. Likewise, several studies have investigated the decomposition of <sup>14</sup>C-labelled soil organic matter (SOM) or plant material in the presence of roots and have yielded conflicting results (e.g. Jenkinson, 1977; Sparling *et al.*, 1982; Martin, 1987; Sallih and Bottner, 1988; Cheng and Coleman, 1990). The use of a model rhizosphere system (Gahoonia and Nielsen, 1991), combined with <sup>14</sup>C labelling of an SOM pool and the SMB, enabled direct measurements of the SMB in distinct soil layers in close proximity to roots and an investigation of its influence on the turnover of recently formed microbial residues.

#### **Materials and Methods**

Two sandy loams were compared in the experiment, one with a normal fertilization record (*fertile*) (22% clay, 21% silt, 37% fine sand and 20% coarse sand; organic C content, 1.31%; N content, 0.13%; inorganic P (Olsen-P), 52  $\mu$ g g<sup>-1</sup> soil; pH (0.01 M CaCl<sub>2</sub>), 5.7; SMB nitrogen,

26  $\mu$ g N g<sup>-1</sup> soil), and one which had received inorganic N but no P and K for 30 years (*nutrient-depleted*) (20% clay, 20% silt, 37% fine sand and 23% coarse sand; organic C content, 1.25%; N content, 0.12%; inorganic P (Olsen-P), 14  $\mu$ g g<sup>-1</sup> soil; pH, 5.6; SMB nitrogen, 14  $\mu$ g N g<sup>-1</sup> soil).

 $^{14}$ C-labelled glucose (0.89 mg g<sup>-1</sup> soil, specific activity: 560 kBq g<sup>-1</sup>) was added to the sieved soil. The soil was pre-incubated at 25°C for 4 weeks. Soil samples were packed in PVC cylinders (3 cm long, 5.6 cm diameter) closed at one end with a tightly attached 53  $\mu$ m nylon mesh.

Ryegrass (*Lolium perenne*) plants were grown in PVC cylinders (length 10 cm, inner diameter 4.4 cm). When root development at the bottom was abundant, the cylinders were placed on top of the labelled soil in a model rhizosphere system (Gahoonia and Nielsen, 1991). The plant cylinders were screened from the labelled soil by the 53 µm nylon mesh. Root hairs were able to penetrate the mesh, thus creating a one-dimensional model root surface. The soil cylinders were placed on a sand bench, which maintained soil moisture at 15% (w/w) corresponding to 50% of waterholding capacity. Control tubes containing no plants were included to check for non-rhizosphere effects on the soil.

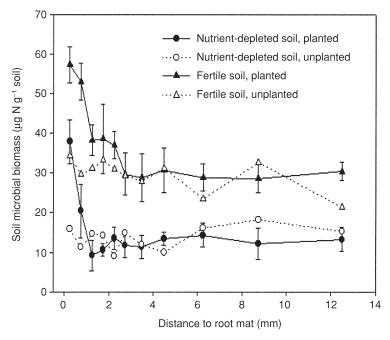
Plant (four replicates) and control (two replicates) soil cylinders were harvested after 15 days. At harvest, the labelled soil was frozen in liquid nitrogen. The frozen soil cores were sliced parallel to the root mat at 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0, 5.0, 7.5, 10.0 and 15.0 mm distance. SMB was measured using fumigation extraction (Brookes *et al.*, 1985; Vance *et al.*, 1987), SMB N was measured as ninhydrin-reactive N (Amato and Ladd, 1988; Carter, 1991) which was converted to biomass N using a  $k_{\rm ninh}$  value of 0.20 (Joergensen and Brookes, 1990). SMB <sup>14</sup>C was determined by liquid scintillation counting. Values were corrected to total <sup>14</sup>C microbial biomass using a  $k_{\rm EC}$  value of 0.45 (Wu *et al.*, 1990).

The effect of freezing on the SMB was tested by analysing frozen and unfrozen soil samples for SMB N and SMB  $^{14}$ C content.

#### Results

From the beginning of the experiment, the SMB N in the *fertile* soil was twice as large as in the *nutrient-depleted* soil. At harvest, the SMB in the soil fractions closest to the root mat had increased from 14 to > 40  $\mu$ g N g<sup>-1</sup> in the *nutrient depleted* soil, and from 26 to 57  $\mu$ g N g<sup>-1</sup> in the *fertile* soil (Fig. 5.5.1). The rhizosphere effect extended 1 mm into the *nutrient-depleted* soil and 2.5 mm into the *fertile* soil.

After freezing, SMB N was reduced by 16.5% in the *fertile* soil and by 14.3% in the *nutrient-depleted* soil. SMB <sup>14</sup>C was reduced by 5.6 and 5.3%, respectively.



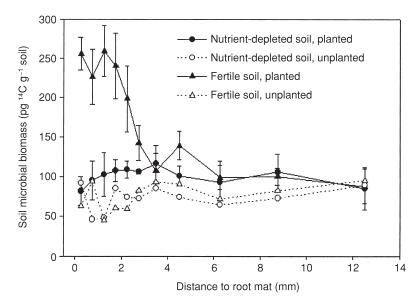
**Fig. 5.5.1.** Soil microbial biomass N at increasing distance from the root mat. Bars indicate SE(n = 4).

In the *fertile* soil, SMB  $^{14}$ C in the rhizosphere increased from 91 pg g<sup>-1</sup> soil in the bulk soil to > 250 pg g<sup>-1</sup> soil at the rhizoplane; in the *nutrient-depleted* soil, no rhizosphere effect was observed (Fig. 5.5.2).

#### Discussion

The measured extension of the rhizosphere in this experiment (1–3 mm) is in accordance with findings of previous studies, whereas the increase in SMB at the rhizoplane is in the low end of the measured range (Helal and Sauerbeck, 1986; Youssef *et al.*, 1989; Yeates and Darrah, 1991; Badalucco *et al.*, 1996). However, direct comparison between the studies is not possible due to variation in experimental set up and choice of parameters. Data from harvest at day 8 (data not shown) indicate that the SMB was no longer increasing rapidly; however, the incubation time was shorter than in most of the compared studies. The less pronounced rhizosphere effect in the *nutrient-depleted* soil was most likely caused by lack of available nutrients for microbial growth.

Studies of the effect of live plant roots on decomposition of SOM show contradicting results. Several studies found that <sup>14</sup>CO<sub>2</sub> evolution



**Fig. 5.5.2.** Soil microbial biomass  $^{14}$ C at increasing distance from the root mat. Bars indicate SE(n=4).

from labelled SOM was suppressed in the presence of living roots (e.g. Jenkinson, 1977; Sparling et al., 1982; Martin, 1987), while others (Helal and Sauerbeck, 1986; Sallih and Bottner, 1988; Cheng and Coleman, 1990) found a stimulatory effect. In this experiment, the increase in SMB <sup>14</sup>C in the rhizosphere of the *fertile* soil clearly showed that carbon was moved from a pool that was not extractable after fumigation, to one that was. Two possible explanations exist: (i) <sup>14</sup>C-labelled SOM was taken up by the SMB, and hence decomposed (a priming effect); or (ii) <sup>14</sup>C-labelled microorganisms in a dormant state were revitalized by the exudates, and became prone to fumigation. The total changes in soil <sup>14</sup>C were not significant in this short experiment and it is uncertain whether the roots in the long term would have exerted a priming effect on the SOM. Different changes in soil biological and physical factors induced by roots, depending on experimental set up, may explain the conflicting results in the literature (Cheng and Coleman, 1990; Dormaar, 1990). The complexity of the situation is accentuated by the fact that no significant priming effect was found in the nutrient-depleted soil. We propose three explanations: (i) the SMB was nutrient limited which hampered decomposition of the labelled SOM; (ii) the SMB in the nutrient-depleted soil was not capable of decomposing the <sup>14</sup>C bound in microbial residues; and (iii) unlike in the fertile soil, dormant SMB in the nutrient-depleted soil was not brought back into an active state. The first explanation appears unlikely since microbial residues would contain the necessary nutrients for microbial growth.

Moreover, unpublished results indicate that even low quality plant residues decompose readily in the *nutrient-depleted* soil.

The test of the freezing revealed a difference in susceptibility of the labelled and unlabelled SMB. Since only a minor fraction of the SMB was labelled, this does not necessarily have any implications for the results. However, it suggests that microbes are affected differently by frost depending on their guild and/or physiological status. Hence, the SMB may have responded differently to freezing in the rhizosphere than in the bulk soil.

#### Conclusion

High-resolution microtome slicing, combined with microscale fumigation extraction, enabled direct measurements of the SMB in the rhizosphere. Within 2 weeks, the total amount of SMB N was roughly doubled in the rhizosphere of the two different soils. The effect of root deposition extended 1–3 mm into the soil, with the largest effect observed in the *fertile* soil. Increased decomposition of labile <sup>14</sup>C-labelled soil organic matter in the rhizosphere, or revitalization of dormant organisms, was indicated by an increase in the SMB <sup>14</sup>C in the soil fractions nearest the root mat. This effect was not significant in the *nutrient-depleted* soil.

### **Acknowledgements**

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### Soil Teeming with Life: New Frontiers for Soil Science

6

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#### Introduction

Soil science in its sub-disciplines of physics, chemistry, biology and taxonomy/genesis is a century old. Many of the basic principles have been established and many practical questions answered. Some would argue that the significant discoveries have been made, the work done and it is time to move resources to emerging fields. In some respects, there is truth in this argument, but at others it is short-sighted and lacking in vision. If we restrict our questions and approaches to those of the past, the criticism applies, but if we consider the challenges of understanding, managing and harvesting the most complex biological community, then we have in our hands one of the greatest opportunities in science.

Many see biology at the heart of the scientific enterprise of the next century. We would agree with this projection, but we also see soil science as an integral part of the biological research enterprise. This may mean that some of our goals and the context of our research changes, but it does not mean that knowledge from any of the soil science sub-disciplines is lacking in importance. We are suggesting that soil biology can become at least one, if not the major driver, for soil science research in the next century.

Important practical issues require soil biology knowledge. These include understanding the role of soil processes in global warming and strategies to ameliorate it; enhanced and safe recycling of waste from manures, urban and industrial activity; pollutant destruction at waste disposal sites as well as on landscapes contaminated from natural processes; biological control of rhizosphere pests; enhanced groundwater

quality, including safety from the emerging water-borne pathogens; discovery of new biotechnological products, including new pharmaceuticals, pesticides and enzymes from the undiscovered microbial diversity of soil; and optimizing recycling of soil nutrients, soil texture and water content for sustainable agricultural and forestry. The terrestrial (soil) environment hosts almost all of the world's human population and provides much of its basic resources. The biology of soil and its control by the soil's chemical and physical features play a daily role in sustaining those resources. Hence, there should be no question about the importance of soil science in the 21st century.

Biology at the current level of understanding is recognized as complex, i.e. the interactions at the molecular, organismal and environmental level are multiple and often non-linear, making predictability difficult. Understanding this complexity will require expertise from most scientific disciplines including the geosciences, chemistry, physics, computational sciences and even the social sciences. The soil environment is arguably the most complex biological community because of the extremely high diversity at small scales and a chemical environment of complex and changing gradients housed in a heterogeneous physical environment. These features are influenced further by larger scale effects such as climate, geological history and human activity. Several basic facts are important in appreciating the complexity of this community, including:

- 1. Soil harbours high population density. Fertile surface soils typically contain a few billion prokaryotes (bacteria and archaea) per gram and often an equivalent amount of fungal biomass. While soil particle surfaces are not crowded with life at this density, it nonetheless means that the potential for diverse biological activity resides at virtually every microsite.
- 2. Soil harbours enormous microbial diversity. This diversity is exhibited as metabolic, genetic, kinetic, morphological and life history variation. Furthermore, and most significant, it appears that only 0.1% or so of the soil microorganisms have been cultured and hence their metabolic role understood. One of the greatest frontiers in biology remains the discovery and characterization of the particularly novel organisms that reside in soil. Understanding complexity requires knowledge about its component parts; hence novel approaches are needed to understand better the undiscovered diversity.
- 3. Soil harbours a tremendous range of physical and chemical conditions. Life in soil experiences a complexity of gradients of nutrients, oxygen, carbon and other salts which are rarely held constant. Furthermore, the types of carbon compounds are numerous, an important point in understanding a heterotroph-dominated community such as soil. Different mineral surfaces, organic coatings of different ages and composition and the extent and depth of organic surfaces add further to the microbe's complex environment. Also, the physical environment, especially as it influences moisture and the

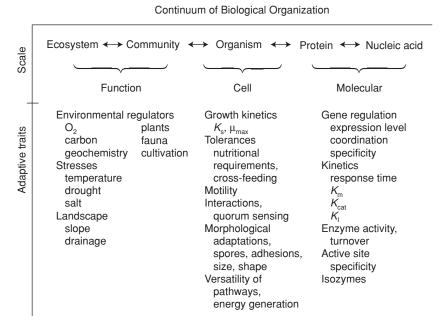
rate of supply of nutrients and electron-accepting resources, is also critical to the microbial community.

4. The soil microbial community is a product of more than 3.5 billion years of evolution. The fossil record indicates that prokaryotes have been on Earth for an extremely long period of time; 85% of their history occurred before Pangea separated. This long period of evolution and natural selection under a wide range of conditions is probably responsible for the enormous microbial diversification. It has also probably selected for organisms that survive stress conditions including starvation, desiccation and freezing. In some sense, a gram of soil may contain a reasonable historical record of the early evolutionary history of life.

The basic premise behind an attempt to understand the complex soil community is that further knowledge will pay off in improved agriculture, environmental decision making and management, and many of the major practical issues listed above. In the past, soil biological processes have been studied at the level of the 'Grand Mean', i.e. lumping all of the diversity and complexity as an average value per gram, kilogram or hectare, for example. This approach has been what was feasible and no doubt useful. The basic question now is can, or in what cases will, a more detailed level of understanding or a mechanistic level of insight be useful? Schimel (1995) has suggested that in some cases it will be and in some cases it will not be. An example of the former is when particular communities selected by one environment has kinetic features or tolerance properties somewhat different from those of communities selected under a different condition. In this case, models of nutrient flux, for example based on Grand Mean coefficients, will not be accurate for both cases. Other examples where knowledge about particular organisms matters would be a PGPR (plant growth-promoting rhizobacterium) that works in one soil type but not in another, or that atmospheric methane is consumed by soils of one ecosystem type but not by another. In other cases, the populations may not differ in ways that affect function, but instead a new level of understanding can be obtained which provides more insight into how or how fast a process is controlled, e.g. the triggering of the molecular regulation of denitrification or the response of quorum sensors that initiate root pathogenesis.

### **Operational Model for Understanding Soil Biocomplexity**

A more in-depth understanding of the soil community and its activity implies exploring biological processes at the organism and molecular levels and understanding how those levels are controlled by soil physical, chemical and climatic factors and by the overlying vegetation. Figure 6.1 shows the continuum in biological organization in the soil community and the adaptive features important at each scale of organization. The adaptive features



# Fig. 6.1. The traits that control microbiological activity and can exhibit variation at different levels of biological organization.

reflect biodiversity that can be important to function. In the past, soil science research typically has stopped at the level of function, but in the future we argue that we should take the lead in extending the continuum, not stopping artificially at the level of function. This model provides a wealth of opportunity for research in the future, a true frontier.

While a biologist may identify more easily with the model in Fig. 6.1 than a soil chemist physicist, mineralogist or taxonomist, it is extremely important that the latter provide their expertise in understanding the environmental conditions so that environmental control of these processes can be understood at the organismal and molecular levels. Understanding this complexity at a mechanistic level demands a multidisciplinary effort. Some of the basic questions to be addressed include:

- 1. Biological diversity is much greater in soil than elsewhere; why? This observation suggests that basic features of the soil matrix promote and sustain diversification. What are the soil features which are most important, does soil management alter these features, and hence diversification?
- 2. Are there microbial patterns that can be explained by soil taxonomy or by vegetative history? Are current soil taxonomic traits appropriate for mapping microbial biogeography? Microbial communities are selected by growth of the successful competitors; the outcome reflects the primary

chemistry (types of organic carbon, available), hence is it the vegetation in soil that determines biogeographic patterns?

- 3. Can microbial activity be mapped at the microaggregate scale? Are microbes in the centre of aggregates inactive relics? Are microbial processes primarily patchy? At what size scales? When? In response to what conditions? The primary regulation of cell activity is thought to be at the level of gene expression. Can mRNA synthesis be measured at aggregate scales? How fast is that expression under realistic soil conditions? For example, what is the time scale for molecular events controlling denitrification following a rainfall?
- 4. What is the degree of coupling between redox active elements and microbial processes? Are these couplings tight, in effect a symbiosis? How does such coupling influence soil geochemistry over time?
- 5. What poorly studied processes might be triggered by the microbes' in situ environment? Does the starvation state induce synthesis of a protective coat, e.g. produce hydrophobic organic matter, or a physiological state resistant to stresses such as desiccation? Such responses could change the nature of soil carbon and result in a physiology that we do not yet recognize. For example, obligate non-spore-forming anaerobes survive in well-drained aerobic sandy soils; why?
- **6.** How can we introduce or manage desired microbial populations to be more effective? How do we improve their dissemination, by earthworms or similar animal vehicles? By a combination of chemotaxis and water management, or by mechanical devices? Once the organisms are dispersed, how do we ensure gene expression?

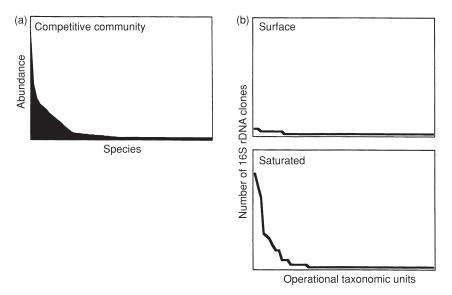
The three following sections illustrate some of the points made above and hopefully show opportunities for better understanding of the soil community in the future. The first shows how spatial isolation provided by the soil matrix apparently sustains soil diversity, the second suggests that soil populations are geographically distinct and the third provides an introduction to the use of genomic and DNA microarray technologies in microbial ecology studies. The latter is projected to have great value for understanding microbes in nature and should provide a natural synergy for collaborative research between basic biologists and environmental scientists.

# What is the Role of the Soil Matrix in Structuring Microbial Communities?

While there is likely to be general acceptance among microbiologists that soil microbial populations are highly complex, recent advances in the molecular analysis of soil communities have revealed a level of diversity previously unimagined. For example, small subunit ribosomal DNA (rDNA)-based studies have shown that clone libraries constructed from

soils can be composed of almost entirely unique members (Borneman *et al.*, 1996, 1997; B. Xia and J.M. Tiedje, unpublished). These studies agree with earlier work where DNA reassociation rates were used to estimate that 4000 non-homologous genomes were present in a forest soil sample (Torsvik *et al.*, 1990). One explanation for these high levels of microbial diversity is that some quality of the soil matrix must promote the development and maintenance of complex microbial communities. The aim of this section is to identify the soil qualities that are most active in shaping microbial community structure and to detail methods by which these qualities can be better defined.

An understanding of the mechanisms that control the structure of microbial communities would clearly benefit any strategy designed to enhance the growth and dominance of a microbial community member, whether that member is indigenous or introduced. Measures of species diversity in plant and animal communities often show that the majority of species are rare and a few species are abundant, suggesting that competitive interaction is a key determinant of community structure (Fig. 6.2a). To determine if competitive interactions also play a key role in structuring soil microbial communities, a small subunit rRNA gene-based approach (Zhou *et al.*, 1997) was carried out on surface, vadose and saturated zone soils. In surface samples, this analysis yielded a nearly uniform distribution of rDNA restriction patterns (operational taxonomic units) indicating that



**Fig. 6.2.** Community diversity profiles. (a) A common diversity pattern seen by ecologists for macroorganisms where competitive interactions define community structure, compared with (b) microbial community diversity patterns for bacteria (determined by ARDRA) in surface and saturated zone soils.

a high level of microbial diversity was maintained (Fig. 6.2b). This type of community distribution where no one member is dominant suggests that competition must be nearly absent, leading us to term this a non-competitive diversity pattern. In contrast, the saturated samples exhibited much less diversity of restriction types, and dominance of one or a few community members leading to a competitive diversity pattern (Fig. 6.2b). In these samples, it appears that a few community members were able to out-compete the rest of the community for nutrients. The vadose zone soils showed community patterns intermediate between the surface and saturated samples; not as diverse as surface communities, but lacking the strong appearance of dominance observed in the saturated zone.

# Can spatial isolation and resource heterogeneity explain these community patterns?

Two hypotheses could explain how the non-competitive and competitive diversity patterns are formed. First, spatial isolation (because of low moisture) in the surface samples could allow for the maintenance of diverse types of microbes and lead to a high level of diversity. At the surface, water films are transient, existing only after a rainfall. As gravity removes this moisture, there will be a low level of connectiveness (high spatial isolation) of soil particles, and microbial species that would normally be lost by competitive exclusion are able to persist. In saturated soils, excess water allows for a high level of connectiveness (low spatial isolation) which offers ample opportunity for the transfer of nutrients and microbes. Under these conditions, the organism best able to scavenge nutrients or migrate to a nutrient source will outgrow less fit types and become dominant.

While the spatial isolation hypothesis fits well with the varied moisture content of our soil samples, an alternative hypothesis is that greater resource heterogeneity at the surface allows for the maintenance of high microbial diversity. The merit of this proposal is that indeed total organic carbon, and probably the variety of carbon types, decreases with increasing soil depth. Thus, multiple resources at the surface could create a variety of microhabitats that support a diverse collection of species. In the saturated zone, the lack of diverse carbon sources means that fewer species will dominate the community.

# Do the non-competitive and competitive diversity patterns appear as a general theme in soils?

If spatial isolation is an important determinant of the diversity pattern, then one would predict that smaller particles, e.g. clays, would contribute more

strongly to a spatially isolated environment. Hence, we would predict that moisture and clay content would shape community structure in a manner such as that hypothesized in Fig. 6.3. We currently are testing whether this hypothesis is supported by examining the existing microbial communities in soils that vary in these two features.

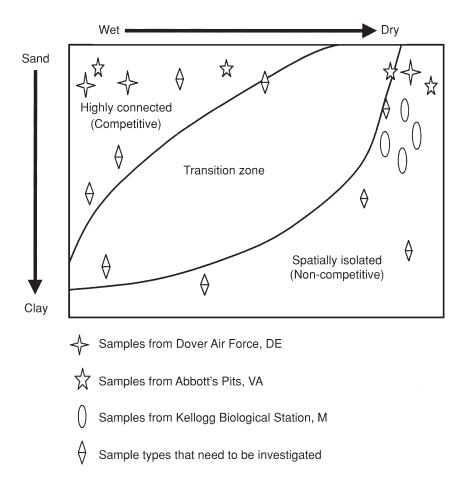
# Test of the hypotheses that spatial isolation and resource heterogeneity act to structure soil communities

In addition to examining existing soil microbial communities, we are conducting controlled laboratory studies with simple two-strain microcosms to evaluate the spatial isolation and resource heterogeneity hypotheses. While our examination of existing communities will reveal community diversity patterns, it is with these simple microcosms that we can test what forces impact on most microbial community structure. The advantage of using this simple system is that many of the abiotic soil components can be held constant while the impact of a single variable, such as moisture, undergoes evaluation. The low complexity of the two-strain community ensures that the dynamics of each population can be measured precisely.

Competition experiments performed by Gause (1934) with two species of *Paramecium* demonstrated that the more competitive species predominated in a uniform, single-nutrient environment. This pioneering work led to the concept of competitive exclusion, the idea that competitors cannot coexist on a single limiting resource (Hardin, 1960). In many ways, a species pair that exhibits strong competitive exclusion, where one species is rapidly out-competed, would be ideal for evaluating our spatial isolation and resource heterogeneity hypotheses. Thus we chose pairs of species that differed in their growth kinetics in liquid culture, when spatial isolation is low. Under these conditions, the species with superior growth kinetics was demonstrated to predominate in a predictable manner. Once these competitive interactions are defined under highly connected conditions, the impact of varied levels of isolation or resources can be tested.

With two-species competition experiments, it must be ensured that positive or negative interactions between the species do not interfere with the hypothesis being tested. For example, if strain A cross-feeds on secondary metabolites produced by strain B, then coexistence will occur even under conditions of low spatial isolation. One solution to this problem is to use two variants of the same species that differ in their growth kinetics. In this case, it may be necessary to distinguish the strains by introduction of a marker, such as B-galactosidase (LacZ) or the green fluorescent protein (Tombolini *et al.*, 1997). Ideal for this second strain pair would be a collection of strains isolated from the same environment. For example, we have evaluated a collection of closely-related 2,4-dichlorophenoxyacetic

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**Fig. 6.3.** Hypothesized relationships of microbial community structure to the texture and moisture content of different soils.

acid (2,4-D)-degrading *Sphingomonas* sp. isolated from an agroecosystem study site (Ka *et al.*, 1994) for use in our microcosm studies. Our initial experiments have focused on two microbial species in competition for a single nutrient in uniform clean sand. Each species is easily distinguished by colony morphology, and in liquid culture and saturated sand (low spatial isolation) one of the strains dominates because of a shorter lag time and superior growth rate. However, as the level of spatial isolation increases, because of decreasing moisture, we have observed that the population sizes of the two species become nearly equal. These results mimic the

non-competitive diversity pattern observed in surface soils (Fig. 6.2), and suggest that even in a highly uniform large-particle matrix, such as sand, the impact of spatial isolation appears dramatic. One would predict that in smaller particle matricies such as soil these spatial isolation effects would be experienced even more readily.

Although simple, the two-species microcosm design is remarkably flexible. They should allow us to tease apart the contributions that spatial isolation and resource heterogeneity make to the maintenance of microbial diversity. The results hopefully will shed some light on how factors such as soil particle size, total organic carbon and clay content of soils impact on microbial diversity.

# Are Soil Heterotrophic Communities Geographically Unique?

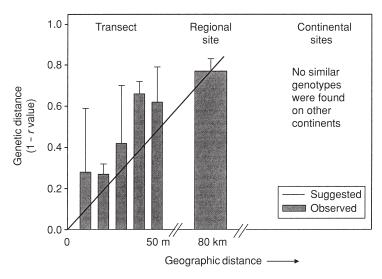
The soil matrix that maintains diversity can also promote an ongoing diversification if the rate of local genetic change exceeds the rate of microbe dispersal. The dogma in the past has been that microbes, being small, are readily transported globally by wind, birds and human activity to name the most likely. This implies that the microbes inhabiting the Edinburgh valley soils are the same as those inhabiting Michigan and New Zealand valley soils. Is this true? The question has not been seriously addressed. Until the development of molecular tools, we did not have a means for realistically addressing microbial biogeography. Most countries have quarantine systems directed against spread of plant pathogens, but these organisms are usually host-associated organisms in their growth habitat, not free-living heterotrophic soil bacteria. Hence, the experience from quarantine is not particularly helpful for resolving the question of soil microbial biogeography.

The question of whether soil microbes are basically cosmopolitan (everywhere) or endemic (geographically unique) has important implications. If endemic, the estimate of global microbial diversity expands tremendously. The answer has important implications for strategies for discovery of new biotechnology products and for national intellectual property rights. Furthermore, if endemism predominates, it means that soil microbial process information is not transported so reliably between different geographic locations. We have addressed the question of whether soil heterotrophic bacteria are endemic with two types of bacterial populations, one set selected on the member's ability to degrade 3-chlorobenzoic acid (3-CBA), a rare property in nature, and the other a coherent taxonomic group, the fluorescent *Pseudomonas*.

Our strategy was that the major ecological features influencing bacterial selection should be the same at least within ecosystem type, i.e. climate, soil

group and the same or similar vegetation, and hence population differences would be more likely to be due to distance. Soil samples were collected from two ecosystem types (Mediterranean and Boreal Forest) that exist in widely separated global regions (southwest Australia, southwest South Africa, central Chile and central California for the former, and northern Saskatchewan and northwest Russia for the latter) (Fulthorpe et al., 1996). We used a hierarchical geographic sampling strategy scaling from samples 5 m apart in 200-m transects, to multiple sites in the same continental region (100-850 km apart), to sites on different continents. The sites either were in parks or nature preserves, or otherwise unimpacted by human activity. Importantly, all soil samples were collected from below the soil surface (5-10 cm) using a soil core sterilized between each sampling. By sampling below the soil surface, we hoped to improve the probability of sampling long-term resident soil bacteria not influenced by human activity. The 3-CBA-degrading isolates obtained from this global sampling showed an endemic pattern; no genotype determined by rep-polymerase chain reaction (PCR) (a rapid measure of chromosome structure) was found on more than one of the six continental regions and most genotypes were not found at multiple sites within the region, although they were found repeatedly in samples from the same 200-m transect (Fulthorpe et al., 1998). The chlorobenzoate-degrading trait, however, is often borne on transmissible plasmids which could mean that the isolate collection contains organisms from multiple phylogenies. Hence, we also examined a readily isolated soil colonizer, the fluorescent Pseudomonas, from the same soil collection. In this case, we explored three levels of genetic difference ranging from coarse to fine resolution: (i) amplified ribosomal DNA restriction analysis (ARDRA); (ii) 16S-23S rDNA intergenic transcribed spacer fragment length polymorphism (ITS-RFLP) and rep-PCR genomic fingerprinting (Rademaker et al., 1998). As expected, no endemicity was seen at the coarse level of resolution (ARDRA method) since the rRNA operon is highly conserved. The ITS-RFLP analysis, however, showed a weak level of endemicity. This species to sub-species level of resolution also analyses a more conserved part of the genome. At the finest level of resolution (rep-PCR), however, we observed strong endemicity. No genotypes were found in more than one continental region, nor in more than one site of the same continental region; however, seven genotypes were found repeatedly along particular 200-m transects. Hence, this second biological example supports the hypothesis that soil heterotrophic populations are endemic, but only at a rather fine scale of resolution. This scale, however, is significant to many ecologically important properties such as pathogenesis, rates of reaction and biotechnological value.

We calculated the relationship between the genetic distance based on the rep-PCR fingerprinting and the corresponding geographic distance (Fig. 6.4). We used one genotype from our reference site in Australia as the



**Fig. 6.4.** The relationship between genetic distance based on genomic fingerprinting (rep-PCR) and geographic distance between isolate sources. The method does not resolve large genetic distances well but does confirm that similar genotypes were not isolated at other sites in the region or on other continents.

base and calculated similarity coefficients to every other genotype in the transect, to all transect isolates of a second Australian site and to all transect isolates in different regions. Those values plotted against geographic distance revealed a relationship of increasing diversity with distance. The methodology we have used so far provides its best resolution at transect scale genetic differences. We currently are working to obtain additional measures that will allow this relationship to be evaluated at larger geographic scales. While these findings support the endemism hypothesis, they also suggest that the resulting corollary is true, i.e. that bacterial diversification is actively ongoing.

### Applications of DNA Microarray Technology to Environmental Microbiology

### Introduction to microarray technology

Since the advent of microbial genome sequencing programmes less than 8 years ago, a massive amount of microbial genome sequence information has been collected. The complete sequences for > 20 microbial genomes have been determined, and > 100 microbial genome sequencing projects are now in progress (www.tigr.org). The next step in the era of microbial

genomics is extracting functional and evolutionary information from these large data sets and, from an ecological point of view, applying genomics technology to relevant questions in microbial ecology. This technology can have a tremendous impact on soil microbiology. Hence, in this section, we introduce DNA microarray technology, describe the basic method and detail some of the potential applications of this technology to microbial ecology as well as some of the current limitations in this field. We hope that this introduction will facilitate entry of this technology into soil science.

DNA microarrays are microscopic arrays of large sets of DNA sequences immobilized on solid substrates. Microarrays are used in hybridization experiments designed to detect gene expression under defined experimental conditions, or to detect the presence of the arrayed sequences in a given sample. There are two general types of arrays: (i) cDNA microarrays, which are constructed either with partial (expressed sequence tag; EST) or full-length complementary DNA (cDNA) sequences typically generated with PCR; and (ii) oligonucleotide microarrays, which are constructed with short (15-40 mer) or longer (i.e. 75 mer) oligonucleotide sequences, designed to be complementary to specific coding regions of interest. In cases when short oligonucleotides are used, often 10-20 probes per gene and mismatch probes are put on the array. There are numerous advantages of microarrays over other hybridization strategies: (i) the high capacity of printing the array on solid substrate (either microscope slides, or  $1 \times 1$  cm<sup>2</sup> wafers) allows tens of thousands of samples to be arrayed; (ii) the overall reduction in size of the experiment reduces amounts of probe and hybridization volume, and increases sample concentration and reaction kinetics (Eisen and Brown, 1999); (iii) global information can be accessed in studies with completely sequenced genomes, or with large numbers of ESTs, such that coverage is broad, and a collective picture of whole organism gene expression can be developed; (iv) speed and high throughput design using robotic printing of DNA samples allows the mass production of cDNA arrays, increasing quality control; (v) parallel design facilitates substantial data acquisition; and (vi) when used with two-colour fluorescence detection, direct comparison of independent experimental samples is readily obtained.

Microarray hybridization approaches promise to revolutionize biology, much in the same way that DNA sequencing and PCR have in recent years. DNA microarrays allow thousands of genes to be surveyed under copious experimental conditions in parallel. Initial studies used cDNA microarrays to determine gene function (i.e. Schena *et al.*, 1995, 1996). For organisms in which the complete genome sequence information is available, it is possible to study the expression of all genes in a single experiment (Eisen and Brown, 1999). Studies have been completed in this regard utilizing the full sequence of *Saccharomyces cerevisiae* (i.e. DeRisi *et al.*, 1997; Wodicka *et al.*, 1997). Additional applications of microarray technology have included

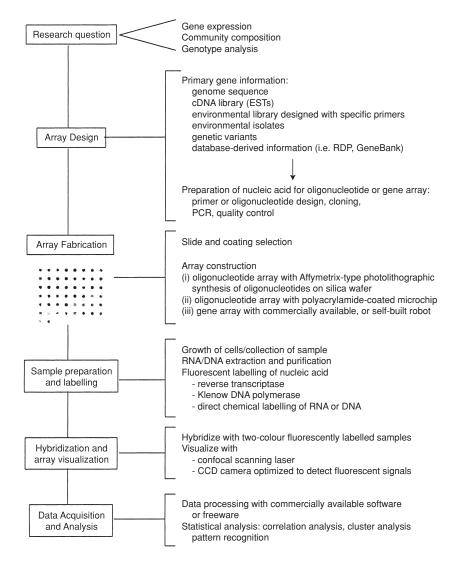
screening for mutations in specific genes (Hacia et al., 1996), identifying genes involved in genetic diseases (Heller et al., 1997), evolutionary sequence comparisons of closely related species (Hacia et al., 1998), studying mutation incurred during adaptive evolution (Ferea et al., 1999) and detecting genetic variants, or genetic expression studies in temporally expressed viral genes (Chambers et al., 1999). Oligonucleotide arrays are also used for DNA sequencing and genotyping (Gingeras et al., 1998), which is a promising application of the high-density oligonucleotide hybridization platform. Affymetrix (Santa Clara, California) is developing sequencing by hybridization technology, and currently is marketing oligonucleotide-based arrays (GeneChips) in which the probes are synthesized in situ utilizing photolithographic technology, in which all oligonucleotides are synthesized in parallel. Currently, GeneChips for rat, human and yeast open reading frames are available, with applications towards expression analysis, polymorphism analysis genotyping, and disease management.

Development of microarray technology for studies in microbial ecology is just being launched (Guschin et al., 1997; Kelly et al., 1999). The use of microarrays in prokaryote applications is also in its infancy, though the number of funded projects in the field of functional genomics, and the numbers of biotechnology and pharmaceutical companies involved in microarray research, is growing rapidly, suggesting that the future in this field is very promising. There are several microbiology/microbial ecologyoriented research areas that will benefit from microarray technology such as: determining the metabolic effects of novel antibiotics or mutations; identifying the presence of specific messages, DNA sequences or genomes in natural samples; understanding gene regulation coincident with pathogenicity; identifying pathways and regulatory networks involved in bioremediation and biogeochemical processes; and screening natural populations for evolutionary divergence. Commercial chips for soil bacteria are not likely to be available; the market is too small. Hence, we will need to make our own.

#### Microarray basics

Due to the variety of schemes for which DNA microarrays can be used, we will discuss, in general, the types of equipment and gene information that will be necessary for development of DNA microarrays that appear to be useful for microbial ecological studies. For specific methods, we refer the reader to a recent publication by Eisen and Brown (1999), which covers cDNA microarray technology as applied to gene expression studies.

The flowchart in Fig. 6.5 depicts the basic strategy for a microarray project. Only general attributes of the scheme have been listed, as this is



**Fig. 6.5.** Flowchart for DNA microarray experimental strategy. Results from a section of an actual array experiment are shown in which genes expressed under denitrifying conditions are compared with their expression under aerobic conditions. The array image has been converted from colour (red versus green), which is much more easily quantified, to black and white. Nonetheless, differences in expression can be seen. The microarray was constructed with PCR products designed from the *Shewanella oneidensis* strain MR-1 genome sequence. This organism is of interest because it can use a variety of electron acceptors for growth including Fe<sub>3</sub>+, Mn<sub>4</sub>+, NO<sub>3</sub>- and O<sub>2</sub>.

intended to give perspective to the reader for what is needed to conduct a microarray experiment. To start, careful attention needs to be paid to developing a research question, and determining the appropriate array format. Arrays which can address a variety of questions will be most valuable, since the bulk of time and cost involves array design and preparing the nucleic acid samples (oligonucleotide synthesis, PCR) for placement on the array. Array fabrication is largely automated and, other than the initial cost of the arraying device (see Bowtell, 1999 for recent listing of products available), this step is quite affordable. The technical details of sample preparation are outlined briefly, though these are important details that need to be worked out, particularly for low biomass environmental samples. The hybridization itself is straightforward; specificity, normalization and sensitivity of the hybridization reaction can be assessed with internal controls on the array. Experiments are conducted with dual fluorochromelabelled templates, with either gene expression compared under two experimental conditions, or a reference sample compared with the experimental sample. Microarrays are visualized with either a confocal scanning laser or a CCD camera specifically designed for microarrays. The image file representing the microarray is processed using commercially available software (see Bassett et al., 1999) or shareware available on the web (http://rana.stanford.edu/software/).

Arrays work in much the same way that traditional hybridization approaches have operated. In a simple case, where the relative amount of gene expression is to be assessed, the target (labelled nucleic acid in solution) samples are varied experimentally. For example, DeRisi et al. (1997) compared mRNA isolated from starved cells with mRNA isolated from cells grown under nutrient-rich conditions. The two different mRNA populations were labelled with different fluorochromes (Cy3 and Cy5), and hybridized together on the same microarray. The scanner delivers two images (one for each fluorochrome) which are overlaid using the processing software. Signal intensities of each spot are determined and a ratio of signal intensities is derived. Using the relative representation of RNA to compare different samples is the most optimal way to use these data, due to differences between sample processing, variations in labelling and other experimental conditions (Eisen and Brown, 1999). The ratio values can be analysed by a variety of statistical methods to assess relationships between coexpressed genes.

#### Microarray uses in environmental microbiology

There are a number of ways in which environmental microbiology will benefit from microbial genomics. As mentioned earlier, microarrays are being used in microbial functional genomics research to determine patterns of gene expression, and identify novel metabolic pathways and regulatory networks. These discoveries at the basic research level will provide invaluable information for environmental studies. Sequence information from completed genomes is being used to design arrays with full complements of all open reading frames for several microorganisms (*Bacillus subtilis*, *Escherichia coli*, *Haemophilus influenzae*, *Helicobacter pylori* (Matrubuthan et al., 1999), *Mycobacterium tuberculosis*, *Pseudomonas aeruginosa* and *Shewanella putrefaciens*). Even for well-characterized microorganisms such as *E. coli*, < 60% of the genome is homologous to genes of known (or hypothesized) function. For organisms less well studied, an estimated 40–60% of the genome may have no homology to characterized genes.

Microarray expression profiles offer a quick way to access functional information for these genes of unknown function. This information can aid functional diversity studies by identifying highly expressed genes and genes critical in biogeochemical pathways, bioremediation or biocatalysis. Cell regulatory function will also become better understood, which could aid in understanding environmental regulation under varying conditions of carbon supply, energy source and electron acceptor availability. Genetic expression for other important environmentally controlled phenomena such as quorum sensing, chemotaxis and antibiotic production may also be monitored, once the regulation and genetic expression for these pathways are understood.

There are a number of direct environmental microbiology applications that we have envisaged for DNA microarrays (oligonucleotide and gene arrays). These include the following:

- 1. Community genome arrays (CGAs). Arrays constructed with genomes of hundreds to thousands of bacteria (environmental isolates) would be used to study community composition and community dynamics of reactors, soil, sediment, water, gut, etc. The utility of CGAs involving DNA–DNA hybridization depends on sample complexity, hybridization kinetics, relatively high biomass samples and the requirement for cultivation of the important organisms in the environment to be studied.
- 2. SSU rDNA arrays. Oligonucleotide arrays constructed for different taxa could be used in community analysis studies. These could be designed in a phylogenetic framework to survey different levels of sequence conservation, from highly conserved sequences giving broad taxonomic groupings, to hypervariable sequences giving genus (and potentially species) level groupings. These assays would not require high biomass sample if PCR or other signal amplification techniques were applied, and would be free from cultivability bias. SSU rDNA array design would be limited by the quality of database information available for SSU gene diversity, coverage of the SSU sequences (20 probes per SSU sequence designed from hypervariable regions) and the sensitivity of hybridization. With > 12,000 prokaryotic sequences in the ribosomal database project

(RDP, http://www.cme.msu.edu/RDP/html/index.html), a large resource of sequence information is available.

- Environmental functional gene arrays. These arrays could come in a variety of styles. One concept would be to prepare oligonucleotide arrays for targeted gene expression, with genes of interest on the array. For example, oligonucleotide probes complementary to genes coding for key enzymes in all biogeochemical cycling processes could be arrayed. These would be used for specific detection of expression in the environment. Another style for an array could be designed to study functional diversity in nature. These gene arrays could include hundreds of PCR products representing the diversity found in nature (e.g. nitrate reductase, ammonia monooxygenase and dechlorinase). The limitations for these two concepts are similar. They rely on available sequence information for designing the array. Functional gene sequencing lags far behind the information available in SSU databases though, with the diversity of genome projects underway, this situation is changing rapidly. Additionally, samples of varying biomass concentration may present technical challenges, since large amounts of RNA are required for the hybridization experiments (5-10 ug total RNA per experiment). Developments in signal detection, and in signal amplification may aid in these problems.
- 4. Population biology arrays. Genetic diversity or genetic polymorphisms within specific populations can be assessed with oligonucleotide arrays. This has already been done with *M. tuberculosis* (Gingeras *et al.*, 1998), *S. cervesiae* (Ferea *et al.*, 1999), and with the human cytomegalovirus (Chambers *et al.*, 1999) in which the potential for this application was demonstrated. Oligonucleotides representing all open reading frames of a reference organism genome can be arrayed, then assayed against strain-level variants. Similarly, cDNAs for a genome of interest could be arrayed then mRNA from isolated strains could be compared with the reference organism to study speciation and functional relationships between the isolates.

There are clearly a large number of different applications of microarray technology that can be applied to relevant problems in environmental microbiology. The field of soil microbiology will benefit invaluably from the contribution to our understanding of microbial content and function in the natural environment.

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# Sustaining Soil Organic Matter 6.1

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#### Introduction

Concern about the loss of organic matter from soils and the implications of this for the sustainable functioning of soils is not new, but the increasing demands that are being placed on our environment are leading to an urgent need to reassess the role played by soils in the development of sustainable patterns of land use. This book has helped to achieve this by providing a wide-ranging selection of relevant research papers and reviews. In this chapter we review some highlights from the material presented and outline what we see as the main conclusions.

The concept of soil quality is used to define those attributes of soils that are essential to soil functions such as nutrient storage, the provision of a suitable physical environment for plant growth and the attenuation of pollutants. In the opening chapter of this book, this concept was reviewed by Carter, where he emphasized the importance of defining precisely those attributes that are pivotal in controlling organic matter quality as well as the need to develop standardized measurement and sampling procedures. Carter identified specific fractions of organic matter that describe soil quality. These include macroorganic matter, microbial biomass and carbohydrate contents. Silt- and sand-sized macroorganic matter is important in maintaining the protection effect of soil organic matter. This acts mainly in promoting and stabilizing soil aggregation. Loveland *et al.* proposed that the important component of soil organic matter is the 'active fraction', made from recent additions of crop residues and organic manures.

Cultivation and compaction can be associated with physical degradation. Cultivation generally depletes organic matter and reduces soil

structural stability. Chenu *et al.* showed that soil structural stability is largely dependent on complexes of clay and organic matter. In protecting soil from physical degradation, the quality of soil organic matter is probably more important than the overall content. Loveland *et al.* reviewed many research papers, and found that there was little consistent evidence for 'critical thresholds' of soil organic carbon above or below which soil physical properties change significantly. This does not imply that the role of soil organic matter is any less important. However, it creates more difficulties for those responsible for devising policies of soil protection, and suggests that evaluation of soil quality on a case by case basis may be required in order to ensure that soil functions are maintained adequately.

Soil organic matter modelling provides a valuable opportunity to explore ways of managing the terrestrial carbon cycle. Modelling at the regional scale is important for climate change issues. At this scale, Paustian advocated a whole ecosystem approach where the interactions between soil organic matter, crop yields, economic returns and subsequent changes in management feed back to determine organic matter and crop responses. This modelling has, amongst other things, highlighted the close linkage between C and N cycling processes. For example, Franko found that the organic matter content in some German soils has reached an optimum level. Above this level, nitrogen losses can exceed inputs resulting in a net loss to the environment. At a similar scale, organic matter modelling was applied by Gaunt et al. to predict the dynamics of soil nitrogen supply required for making more precise fertilizer recommendations. They suggested that measurable fractions of soil organic matter can be used to define the potentially available nutrient pools, making the models of greater practical value. Tillage has a large effect on carbon and nitrogen dynamics, with C and N lost after ploughing out grassland. Richter et al. used a modelling approach to show that these losses resulted in a decrease of net mineralization and a widening of the C: N ratio. They also showed the importance of microbial carbon and nitrogen associated with litter and crop debris in specifying the soil microbial biomass. Paustian identified a need for an increased collaboration between modellers. However, he stressed the continued need for long-term experimentation and for closer correspondence between theoretical and measured organic matter fractions. The availability of a richer set of field experimental data, and the use of isotopic tracers should in future allow more robust and constrained testing of conceptually based models.

## Soil Organic Matter and Land Management

The sequence and type of crop rotations have been widely shown to influence plant productivity by affecting physical, chemical and biological

aspects of soils, as well as the prevalence of weeds, pests and diseases (Sumner, 1982). Rotations comprising only annual crops may cause a decline in soil organic matter by leaving relatively few plant residues (above and below ground) compared with perennial crops. In contrast, perennial forages have been shown to build up soil organic nitrogen (Clement and Williams, 1967), improve soil physical properties (Tisdall and Oades, 1982) and reduce the risk of soil erosion. Management of crop rotations by manipulating application rates of crop residues and manures, tillage and treatment of crop residues (e.g. mulching or incorporation) can have a significant effect on soil organic matter dynamics.

Organic farming systems represent an increasingly important land use in Europe and beyond. The land area farmed organically in Europe has grown from 100,000 ha in 1985 to 2.8 Mha in 1998 (N. Lampkin, personal communication). The principles of organic agriculture as defined by the International Federation of Organic Agricultural Movements (IFOAM, 1998) specifically include the maintenance of long-term soil fertility as a prerequisite. Increases in soil organic matter in soils under organic management are widely reported (e.g. Reganold et al., 1993, Clark et al., 1998). Wander and Traina (1994) have also measured higher levels of carbon in the 'light fraction' of soils under organic management which is thought to be an indication of a more biologically active pool. As the number of organic farmers grows, there are increasing numbers of conventional, specialist, arable farmers wanting to convert to organic production. This poses particular challenges for soil organic matter management. Grass-clover leys are traditionally the 'engine' of organic systems in Western Europe, but grass-clover levs are not an economic option without livestock to utilize their productivity. Philipps has shown that with less than 25% of nitrogen-fixing crops in the rotation (i.e. a legume grown 1 year in every 4), organic matter declines over a 10-year period. However, working with a rotation with a similar proportion of legumes, but comparing the use of mineral fertilizers and farmyard manure with or without biodynamic preparations, Raupp found that soil organic matter was higher in the manured + biodynamic treatment, and lowest in the treatment which received only mineral fertilizer.

The sustainable management of crop residues in agroecosystems is a global challenge. Lal (1995) calculated that on an area of  $1 \times 10^9$  ha of agricultural land,  $\sim 3.5 \times 10^9$  Mg of crop residues are produced. Approximately 74% of this originated from cereals while the next biggest contributors were sugar crops at 11%. Crop residues have an important role not only in building soil organic matter and in conservation of soil and water, but also in supplying nutrients to subsequent crops in rotations and to simultaneous crops in, for example, agroforestry systems. The magnitude of benefits from crop residues depends on the quantity and quality of the residues, as well as the following crop, climatic, edaphic and management factors. Until

recently, it was widely accepted that equilibrium levels of carbon and nitrogen in soil were controlled largely by net input, and that qualitative aspects were relatively unimportant. Drinkwater et al. (1998), however, found that quantitative differences in inputs alone could not explain observed changes in soil carbon and nitrogen, and that plant species composition and litter quality influenced soil organic matter turnover. They suggested that managing the quality of inputs could help to increase carbon sequestration and reduce CO<sub>2</sub> emissions, in accordance with the Kyoto protocol. Cadisch and Giller move this idea forward by suggesting that soil organic matter management should begin with the decision as to whether we are managing organic matter for carbon sequestration or for crop nitrogen supply. The key residue characteristics that govern the outcome are carbon to nitrogen ratio, lignin and polyphenol content. They also highlight the importance of understanding differences in decomposition of root material compared with above-ground material. Recent research has suggested that root turnover is relatively short for many temperate species, for example ~30% of grass and clover roots survive for < 3 weeks under UK field conditions (Watson et al., 2000). Although there are now reliable estimates of root turnover for many tree and agricultural species (Black et al., 1998; Watson et al., 2000), there is still a lack of quantitative data on soil organic matter inputs from this source.

In addition to the influence of quantity and quality of residues on potential nutrient release and soil organic matter accumulation, physical management of residues is also a key issue. Baggs *et al.* and Vinten *et al.* both address the question of particle size of residues. The use of crop residues and off-farm organic wastes within cropping systems may require alterations to normal fertilizer and cultivation practices in order to maximize crop uptake and minimize nutrient losses (Shepherd *et al.*), Robertson and Thorburn, and Vinten *et al.*).

As stated in the Introduction, nutrient budgets are used increasingly as international indicators of sustainability. Nutrient budgets can be used for a number of purposes; they can identify the long-term sustainability of a system and may be able to suggest management options that can improve nutrient retention. They can also be used to identify gaps in our knowledge of nutrient fluxes by using simple models to calculate fluxes that would otherwise be difficult to measure experimentally. Finally, they can be useful as a tool for policy makers in order to allow the synthesis of data at the scale of a catchment or region. Fortune *et al.* point out some of the difficulties in interpreting budgets, due in part to the different methodologies used in their compilation, and also the need to understand the reliability and limitations of the data available. Pilbeam *et al.*, working in Nepal, use nutrient budgets to illustrate how sustainability at one level, in this case the household, may jeopardize the sustainability of the system at a higher level, when the origin of imports to the household is taken into account. This

highlights the need to define the boundaries for compiling budgets in relation to use of the resulting information.

As it is no longer acceptable to judge land management practices simply on the basis of their current productivity, we must understand the long-term implications of current practices. The combination of archaeological knowledge with modern analytical techniques and modelling is potentially a very powerful tool. McCann *et al.* provide a fascinating insight into the origins of the Terra Preter soils in Amazonia, and the paper by Glaser *et al.* explores the scientific evidence underlying farmer observations that these soils are the most productive in the region. In a contrasting setting, Adderley *et al.* draw conclusions on the sustainability of traditional manuring practices on a remote Scottish island.

Major changes in land use, such as the ploughing out of long-term pasture, are known to result in major disturbances to the carbon and nitrogen cycles (Johnston, 1991; O'Sullivan *et al.*). The question of how best to maintain soil organic matter levels following a more major land use change is difficult. Hatley *et al.* and Mazzoncini *et al.* both assess the impact of different management treatments following land use change in the contrasting environments of East Anglia and the Mediterranean.

#### **Nutrients**

One of the key roles played by soil organic matter as discussed by Goulding et al. is that involving the supply of nutrients to plants and soil organisms. Soil organic matter contains substantial pools of organic N, P, S and a number of trace elements; however, the availability and mobility of these elements in organic compounds is generally very much less that that in the inorganic state. An understanding of the process of transfer between the organic and inorganic states (mineralization) and the reverse process of immobilization is therefore critical to our approach in this area. It is often assumed that high inputs of inorganic fertilizers can substitute for the nutrient supply from organic matter pools. Work with isotopically labelled N and P, however, has shown this not to be the case. Even where high inputs are supplied, organic matter plays an important controlling role. Results from field trials in the UK and USA have shown that even when N fertilizers are added in amounts that are sufficient to satisfy the crop's demand, the crop recovery of fertilizer-derived N is no more than 60% of that which was added, with the remainder being made up from N released from organic matter pools and small amounts from atmospheric inputs. In other words, if we wish to manage fertilizers efficiently, we must be in a position to understand the interactions between added fertilizer materials and the organic matter pools. Short-term immobilization by the microbial biomass potentially can result in crop nutrient shortages. However, work by Vinten et al. has shown that crop N recovery can be optimized whilst

minimizing losses of N through leaching by careful soil management that takes account of soil microbial processes.

Measuring the process of mineralization has for many years proved difficult, but is essential if we wish to be able to quantify and/or manage nutrient transformations within a given system. Using the recently developed pool dilution techniques, we now realize that flows of nitrogen in soils that result from gross mineralization processes are very much larger than previously thought. Goulding *et al.* have shown that gross transformations of N may exceed 18 mg kg day<sup>-1</sup>, indicating that such fluxes may be many times the uptake of N by crop plants, although in this example the high mineralization rates were accompanied by high immobilization. Methodological problems remain in the use of such techniques, though they do provide a real indication of microbial activity and allow improved insight into the competition between plants and microbial populations for nitrogen.

The concept that mineralization of organic nitrogen is a necessary prerequisite for plant nitrogen acquisition has been questioned in a number of recent studies. Goulding *et al.* have found that extractable soil N in a range of arable soils in the south of England contains 55–65% of total soluble N in organic form, which clearly indicates the importance of possible N loss in this form and may indicate a possible source of N for plants. Nasholm *et al.* (1998) found rates of <sup>15</sup>N-labelled glycine uptake by coniferous trees, dwarf shrubs and grasses that were comparable with that of NH<sub>4</sub><sup>+</sup>. Uptake of organic N is known to be of importance in upland and boreal vegetation, and has often been assumed to be mediated by ectomycorrhizas (Turnbull *et al.*, 1996). It has been suggested by Jonasson and Shaver (1999) that this may explain the reason why some plants adapted to these habitats show relatively little response to additions of inorganic N.

An understanding of the factors contributing to the turnover and decomposition of dissolved organic matter (DOM) fractions is important in allowing us to predict losses and potential mineralization from this source. Several papers in this book have identified the importance of DOM in contributing to losses of organic carbon with associated nutrients from soil profiles by leaching (Kaiser et al., McTeirnan et al., and Marschner and Bredow). It is likely, however, that site properties, such as hydrology, play an important role in mediating such losses (McTiernan et al.). Chapman et al. found that concentrations of dissolved organic nitrogen (DON) remained constant despite changes in net mineralization of N and suggested that this could be explained by the equilibrium between the DON produced and a larger reserve pool. The central role of organic matter as an intermediate in the process of mineralization (Appel and Mengel, 1990) underlines the importance of developing a better understanding of dissolved organic fractions in nutrient cycling processes.

The storage of nutrients in soils is closely linked to the availability and throughput of organic matter derived from plants. Current increases

in atmospheric CO<sub>2</sub> have led to an alteration in the equilibrium between soil organic matter and the pre-industrial CO2 concentrations. This is complicated further by the enrichment of our environment with fixed N, which interacts with the added organic C in soils to produce effects on soil properties and soil organisms that are difficult to anticipate (Swift et al., 1998). Most of the carbon held in terrestrial ecosystems is in the soil  $(\sim 1500 \times 10^{15} \text{ g})$  and is derived from plant and animal material (Batjes, 1996). Changes in the soil carbon stores may result from the effects of elevated CO<sub>2</sub> on plant growth and from the climate changes resulting from the change in global atmospheric composition. Elevated levels of CO<sub>2</sub> can affect the quality of leaf and fine root litter, their decomposition rates and the relationship between litter quality and decomposition (Cotrufo et al., 1998). Elevated levels of atmospheric  $CO_2$  were shown by Torbert *et al.* to increase both soil organic carbon and total nitrogen content under soybean and sorghum. Martin-Olmdèo et al. found no direct plant-mediated effect of elevated CO<sub>2</sub> on nitrous oxide production or emission from soil. However, they considered that the positive effect on plant growth and microbial biomass by the CO<sub>2</sub> might affect potential feedback effects between soils and atmosphere.

The build-up of greenhouse gases can be limited quite considerably through improved soil management; according to Smith *et al.* agricultural soils can be particularly important. The sequestration of carbon in organic matter in agricultural soils is an important mitigation option. This can be achieved using organic amendments, improved residue management and tillage techniques, alternative cropping regimes and changes in land use cover. Afforestation and bioenergy production are the changes with the greatest mitigation potential.

Soils of the boreal and sub-arctic vegetation zones are important for carbon storage, particularly in the sub-soil. Guggenberger *et al.* stated that, in Siberia, where global warming is relatively rapid, belts of vegetation may shift northward enabling more soil organic matter to be stored, though emissions of greenhouse gases may increase.

A future problem in many areas of the world will be an increasing incidence of forest fires which leave soil exposed and vulnerable to degradation. Haslam *et al.* showed how solid-state <sup>13</sup>C-nuclear magnetic resonance spectroscopy can be used to estimate the changes in soil organic matter quality as organic material reaccumulates after fires.

## **Biodiversity**

Organic matter sustains the life of soil and this is inherently important to the concept of soil health. Microorganisms in particular play an essential role in the transformations of organic matter and nutrients that underpin

many soil processes. It has long been recognized that soil is the most complex of all environments and yet there is a great need to find effective and sensitive ways of monitoring its health. Being able to measure the response of soil microorganisms to environmental change may prove to be a valuable and rapid way of measuring the health of our soil. A number of soil microbiological parameters, notably microbial biomass carbon and basal respiration, have been suggested as possible indicators of soil quality and are employed in national and international monitoring programmes. More recently, we have moved into the 'age of communities' (Tiedje, 2000) and microbial diversity has also been recommended as a biological indicator of soil quality (Kennedy and Smith, 1995). But how do we quantify this intractable diversity? New methods to characterize, isolate and identify soil biota suggest that we have only just scratched the surface of a large and undiscovered gene bank, the reasons for which are intriguing. Organic matter also has a major effect on the soil physical environment. Indeed, the heterogeneity of the soil physical environment may in fact partly explain why soil has such a large diversity (Tiedje et al.). The pore structure of soil and its interaction with soil-water relationships can create microhabitats that lead to spatial isolation (islands), which may explain the diverse biogeography that we are only now starting to discover (Tiedje et al.).

The current momentum in soil biodiversity research is fuelled not only by the prospects of species and product discovery and the development of new molecular tools, but also by the opportunity to test new ecological theories and the pressing need to solve intractable problems associated with producing food and protecting fragile ecosystems. Linking biodiversity to ecosystem function is an exciting area scientifically and of broad interest in contemporary ecology. It is often assumed that soils with the greatest diversity of microorganisms may be the most resilient to pollutant stress. Ritz and Griffiths point out that there are many potential pitfalls when testing such hypotheses and also show that they may depend on how transient the stress is. Interestingly, they found that a physical, transient stress (heat shock) produced a different response from a persistent chemical stress (Cu contamination). The link between resilience and pollution effects is also made more difficult to understand because the same soil properties that affect diversity (e.g. organic matter, pH, texture; see Tiedje et al.) will also alter the bioavailability of many pollutants. It is important, therefore, when studying the effect of pollutants on diversity also to measure their availability to ensure that the selective pressures/or toxicity in different soils are unequivocal.

The effects of organic or biodynamic farming systems on biodiversity are areas where increased 'biodiversity' is often put forward as justification of the merits of different systems. Fliebßach *et al.* did find increased diversity in biodynamic systems compared with conventional fertilized

soils, and this was inversely related to the metabolic quotient of the soil microbial biomass. However, they did not find significant effects in the quality of the soil organic matter, measured by <sup>13</sup>C-NMR. O'Flaherty *et al.* showed that metal-rich sludges applied to land, which might otherwise have been assumed to cause stress and a reduction in diversity, actually increased diversity (measured using molecular methods). Clearly, simple generalizations may be hard to come by.

Several papers have quantified functional diversity (Fliebßach et al., O'Flaherty et al. and Degens) and attempt to relate the quality of organic matter and/or changes in land use to diversity. Fleibßach et al. and O'Flaherty et al. tested soil extracts using Biolog plates to construct community-level physiological profiles (CLPPs), while Degens has pioneered the use of whole-soil substrate-induced respiration methods to produce catabolic response profiles (CRP). There is still debate on how well such methodologies measure functional diversity. Both approaches measure the potential utilization of different carbon sources at relatively high levels of C amendment, but clearly the functional approach is seen as a useful way to gain new insight.

Biological indicators of soil health should ideally be rapid and sensitive but there also needs to be a substantive amount of background information on natural variation and what constitutes 'normal' responses before value judgements can be made. Degen's use of catabolic diversity as a generic indicator of changes in soil functioning due to land use and the application of wastes to soil is a case in point. If rapid methods could be found for measuring such parameters, then this is an approach that might be attractive to agencies that have to monitor and regulate soil protection policies. The vision of the future presented by Tiedje *et al.* suggests that functional genomics will eventually allow us to measure important functional attributes, possibly at the mRNA level, so that the limitations of the potential measures and culturability will one day be overcome.

What then, after we have fully quantified this diversity? How do we then manage or manipulate it to create a more sustainable system? The ability to manage soils to enhance key species such as earthworms (Scullion and Malik) or rhizobium is clearly an advantage. Microbial communities might in the future be managed for environmental protection as well as to enhance nutrient supply. For example, organic matter (sawdust) added in trenches adjacent to streams has be used to stimulate denitrifying organisms and create a 'denitrification wall' to protect waters from excess nitrate (Schipper and Vojvodic-Vukovic, 2000). Thus management of key functional groups or species responsible for key processes is arguably quite realistic, but how to manage the more complex generalist communities? The importance of the rhizosphere as the interface between plant–soil–microbial interactions (de Neergaard and Magid) is also now realized and research is being directed at 'rhizosphere engineering' to achieve, for

example, remediation of pollutants or the biological control of pathogens. How can we then start to think of engineering the transformation of organic matter – to release nutrients in protected organic matter in low fertility soils or to enhance protection of organic matter in soil subject to physical degradation? These are some of the challenges that might be addressed by gaining a greater understanding of how microbial diversity interacts with organic matter.

## **Concluding Remarks**

Land management must play a critical role in developing sustainable strategies of land use in the coming decades. Although cultivation too often in the past has been associated with organic matter loss and soil degradation, we are now in a position to apply our understanding of crop sequences and cultivations, many of which have been described in this volume, in a way that can actively restore organic matter storage, thereby restoring the functions that the soil supports. One of the characteristic features of organic matter, unlike many other important soil properties, is that it is significantly affected by management. Falloon et al. have shown that land management can have a significant impact on the sequestration of carbon by soils, thereby partially offsetting the imbalance between carbon release and uptake by terrestrial systems. We now have a better understanding of the relationships between organic matter quality and its function in soil (Cadisch and Giller). However, there is still much progress to be made in understanding how land management contributes to patterns of spatial and temporal heterogeneity in soils, particularly given the problems that this causes in relation to attempts to try to scale-up processes from the micro scale to the level of an ecosystem.

Modelling of soil organic matter dynamics is likely to continue to help in the understanding and management of the carbon cycle. This is important both at the global scale, enabling projections of carbon mitigation potentials in agriculture, and at the regional scale, enabling an 'optimum' organic matter level to be specified. Exceeding the optimum is likely to cause significant losses of nitrogen in addition to carbon, particularly during land use changes. Future developments are likely to include a wider range of scales of modelling. At small scales, we need further reconciliation of experimental and theoretical descriptions of soil organic matter. At larger scales, where technical development of models is well advanced, more acquisition of historical data on organic matter and land use practices is required.

The scope of the papers presented here highlights the breadth of approaches to soil organic matter research within the soil science community, and the inter-disciplinary nature of soil science *per se*. If we

are to address fully the issues associated with the sustainable management of soil organic matter, however, we need to continue to move beyond the traditional disciplines of soil biology, soil physics and soil chemistry and to work with other natural scientists and, increasingly, with social scientists. Real and perceived divisions between researchers and advisors, or between social and natural scientists, that exist within higher education and research institutes are undoubtedly barriers to the development of inter-disciplinary research. Current research funding methods may also hamper collaboration.

Interestingly, despite the importance of legumes on a global scale in contributing to N cycling and organic matter management, they received little attention at this conference. This perhaps reflects the current emphasis on the study of N fixation by plant physiologists not soil scientists, rather than a lack of research in the subject. Overcoming these barriers, together with our ability to harness the ever-increasing range of molecular and chemical techniques, not only will aid our understanding of soil organic management, but also our ability to influence policy that will protect and enhance soil organic matter across the world. In developing countries, indigenous knowledge and management systems have an important role to play in research (Pretty, 1995). Participative research approaches, which involve farmers, land managers and the extension service in research on soil fertility are being used extensively in developing countries (e.g. Corbeels et al., 2000; White et al., this volume). Such alternative approaches are beginning to gain more widespread acceptance in developed countries, and potentially could result in greater awareness of the importance of soil organic matter amongst farmers and other land managers.

Management of soil organic matter has to date involved primarily chemical and physical (mechanical) treatment to improve structure, incorporate residues and stimulate decomposition. While such field practices will continue, management in the future might also be based on greater biological understanding that seeks to manipulate or engineer the microbial population to enhance crop production and protect soils. Greater understanding of soil biodiversity may also lead to rapid biological tests that can be used to monitor and protect soil health.

The need to manage organic matter sustainably is clear. Much damage has already been caused to the world's ecosystem through neglect of the natural environment and the support systems that it maintains. This book illustrates that progress has been made in linking our understanding of soil processes with functions. It will be necessary to build upon this understanding through engagement with advisors, farmers and land managers to develop strategies that not only halt the degradation of soils but, in time, also reverse it. This will require international effort supported through national governments to value the use of natural resources and investment

in people and technologies that can achieve the ultimate goal of sustainable development.

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