

## **SESSION 4**

# MANAGING SOILS FOR CLIMATE CHANGE ADAPTATION AND MITIGATION



# Carbon Sequestration in Agricultural Soils: Separating the Muck from the Magic

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## ABSTRACT

Intense interest in soil organic matter (SOM) over its role in the storage of terrestrial carbon (C) and in the release of greenhouse gases to the atmosphere manifests itself in a renewal of research efforts to assess the impacts of different soil management systems on C dynamics. This paper assesses the future role of selected systems for managing agricultural and pastoral land; addressing the global land area, potential to sequester carbon, side effects on other sources of greenhouse gases, and other limitations. Some systems such as organic farming and the use of biochar may have positive impacts over limited areas, but agroforestry, grazing lands and conservation agriculture appear to have the greatest potential to sequester carbon by covering vast areas on which C loss is reduced or positive C sequestration occurs. An over-riding problem is the difficulty of measuring changes in soil C. Isotopic techniques applied in comparative studies of different soil management systems offer new insights that will guide land managers and policy makers.

**Key words:** *soil carbon, decomposition, agroforestry, conservation tillage, holistic rangeland management, permanent pasture.*

## INTRODUCTION

Soil organic matter (SOM) plays a key role in soil productivity: as a nutrient reserve; in the formation of stable aggregates and protecting the soil surface; in the maintenance of the vast array of biological functions, including the immobilization and release of nutrients; in the provision of ion exchange capacity; and in the storage of terrestrial carbon (C) (Craswell and Lefroy, 2001). The importance of SOM and its challenging chemical complexity has spawned a vast published literature, e.g. Waksman (1936) cited 1 311 references in his book on humus. Since Waksman's time, land transformation for agriculture through clearing of native vegetation and cultivation has led to accelerated decomposition rates over large areas of land. This led to major declines in soil organic carbon (SOC) levels estimated to have released 55 to 90 Pg ( $1 \times 10^{15}$  g = 1 billion tonnes (t)) to the atmosphere as carbon dioxide or methane (CH<sub>4</sub>; Lal, 2006). A concomitant decline in soil nutrient content led to the widespread adoption of fertilizer inputs in many countries, whereas many farmers in regions such as sub-Saharan Africa have not adopted fertilizers (Craswell and Vlek, 2013). Soil erosion by wind and water has also

displaced soil nutrients and organic C to a level estimated at 4.0–6.0 Pg/yr (Lal, 2003). The greenhouse gases released from decomposition and from the use of fertilizers also continue to contribute to global warming. Consequently the research emphasis has changed from studying the role of SOM in productivity to understanding its function in climate change mitigation by reducing or reversing greenhouse gas emissions. Given the current unprecedented need to expand food production to meet population growth, improving land management to achieve both productivity and sequestration objectives provides an opportunity for a grand win-win success.

This paper draws on several recent reviews (e.g. Trumbore, 2009; Sanderman, Farquharson and Baldock, 2010; Powlson, Whitmore and Goulding, 2011; The World Bank, 2012) to consider how selected agricultural systems may contribute to greenhouse gas mitigation. It then considers how isotope techniques can be applied to increase our understanding of the processes of C accumulation and decomposition.

## Land management effects on carbon sequestration

The management systems selected for this paper are a mixture of traditional and novel systems that may have, or have been advocated to have significant impacts on C sequestration in agricultural soils. As discussed below, some of the claims made regarding the potential for C sequestration do not meet a reality check based on valid measurements or potential rates of adoption by farmers. Hence our concern to separate the muck from the magic. The systems chosen are not mutually exclusive, e.g. combinations of biochar production with agroforestry or minimum tillage. The greenhouse gas issues associated with paddy rice in flooded rice systems have been considered in detail by various authors, so we limit our consideration of rice soils to the newly advocated system for rice intensification.

Table 1 summarizes and generalizes a huge amount of published information. Some more detailed comments on the individual systems are given below, focusing particularly on estimates of the current and potential land area in relation to C sequestration:

### Agroforestry

Different definitions of agroforestry exist, but broadly agroforestry involves the integrative growing of trees in harmony with other agricultural activities including cropping and livestock enterprises. According to Nair *et al.* (2010) forms of agroforestry have existed for centuries, reportedly going back as far as 13 000–9 000 BC in Southeast Asian fishing communities. Today, agroforestry has a diversity of forms depending on the climatic zone, environmental need and socio-economic factors. Agroforestry can include alley cropping, silvopastoral systems such as grazing or “cut and carry” practices, windbreaks and shelterbelt systems. When compared with other

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**TABLE 1. List of selected land management systems indicating some key limitations to effects on net greenhouse gas production**

| Management system               | Comments                                                                                                          | Limitations |
|---------------------------------|-------------------------------------------------------------------------------------------------------------------|-------------|
| Agroforestry                    | Adds standing biomass plus soil C; currently 1 023 million ha with potential to expand on degraded tropical lands | 1           |
| Conservation tillage            | Extensive areas globally, but crop residues decompose and C may accumulate only in the surface soil               | 1,2         |
| Holistic rangeland management   | Potentially large areas, but may be hard to implement; residues and roots incorporated by trampling               | 1,3         |
| Permanent pasture               | Well documented way to increase stable SOC                                                                        | 3           |
| Organic farming                 | External organic inputs, but low yield over limited area                                                          | 1,3         |
| Biochar application             | Inert C inputs have long half life in soil, but economics may limit the potential area                            | 1           |
| System for rice intensification | External C inputs; drying and re-flooding may increase decomposition rates                                        | 1,3         |

Limitations: 1 — few reliable soil C data; 2 — associated N<sub>2</sub>O emissions due to increased N fertilizer use; 3 — increased CH<sub>4</sub> emissions from animals or soil

agricultural activities, agroforestry has the benefit of utilizing and rehabilitating degraded soil (Albrecht and Kandji, 2003), providing windbreaks, food, fodder, habitat, wood products, reducing erosion and improving crop productivity through shelter and cooling (Nair, Kumar and Nair, 2009) and contributing to the terrestrial pool of C by adding stable forms of SOC.

Agroforestry sequesters C in both the above ground biomass and in the soil, but the actual quantity sequestered depends on a large number of variables including site location, species mix, stand age, soil type, soil depth, management and climate. Nair *et al.* (2009) has estimated that globally, 1 023 million hectares (ha) of agricultural land is used for agroforestry production with a further 630 million ha of unproductive agricultural land having the potential to be converted to agroforestry. Estimates of above and below ground C sequestration attributable to agroforestry are difficult to derive due to site variability. Nair *et al.* (2009), in summarizing the literature has found above ground C sequestration rates ranged between 0.29 and 15.21 Mg·ha<sup>-1</sup>·yr<sup>-1</sup> and in soil between 1.25 and 173 Mg·ha<sup>-1</sup>·yr<sup>-1</sup>. The variability in reported C sequestration rates indicates the difficulty in predicting a global average rate of sequestration.

Limitations/constraints include:

- risk of loss due to natural or anthropogenic causes;
- protocol and method variability of soil sampling depths and increments;
- allometric equations for estimating above ground biomass may be applied inappropriately;
- problems in estimating area under agroforestry because of variation in distance between rows;
- tree species composition influences quality and quantity of SOC;
- decomposition rates vary depending on climatic zone and soil type.

### Conservation tillage

Conventional tillage (CT) generally disturbs the soil significantly because the implements used are designed to invert the surface soil. Conventional tillage methods aim to prepare a clean seed bed for cropping activities by controlling weeds and pests (Sanderman, Farquharson and Baldock, 2010). Lal (1997) estimated that conventional mechanized farming methods typically leave less than 15 percent of crop residues on the soil surface following tillage. Conventional tillage disrupts stable soil aggregates, and its repeated use may reduce aggregation significantly, leading to increased susceptibility to wind and water erosion. Conventional tillage also exposes formerly protected C to mineralization thereby increasing emissions of CO<sub>2</sub> to the atmosphere.

Contemporary methods of CT refer to agricultural practices that include minimum tillage, direct drill and zero-till operations. These

practices are adopted to reduce the risk of soil erosion, improve soil aggregation, increase stubble retention and have the potential to enhance the sequestration of C in soil (Sanderman, Farquharson and Baldock, 2010). Derpsch *et al.* (2010) estimated CT was practised on 111 × 10<sup>6</sup> ha of agricultural land globally in 2009, with an estimated annual increased area of adoption totalling 6 × 10<sup>6</sup> ha. Lal (1997) estimated that 1 352 × 10<sup>6</sup> ha of global arable land would be under some form of CT by 2020. Further, the Intergovernmental Panel on Climate Change (IPCC, 2000) suggested that 60 percent of arable land could potentially be farmed by using CT methods. Annual rates of C sequestration associated with CT have been estimated by Lal (1997) to be 0.002 percent to a depth of 1m. This is extended to a global increase in SOC of 0.125 Pg·C·yr<sup>-1</sup>.

Carbon sequestration rates vary widely between sites and climatic zones. Reasons for this include: the natural spatial and temporal heterogeneity of SOC across the landscape; the SOC concentration at time of conversion to CT; a disproportionate increase in the labile C fraction contributing to higher decomposition rates; and responses of different soil types to C (Blanco-Canqui and Lal, 2004). Conservation tillage contributes an increase in C particularly near the surface, but is reported to make little increase and can even decrease SOC content at depth (Lal, 1997). Carbon to nitrogen (N) ratios are an additional factor contributing to the complexity of C sequestration. Since organic matter decomposition requires and immobilizes N, phosphorus (P) and sulphur (S) (Kirkby *et al.*, 2011), the sequestration of C under CT systems may require additional fertilizer inputs which may be counter-productive if increased nitrous oxide (N<sub>2</sub>O) is lost from the N fertilizer to the atmosphere.

Limitations/constraints include:

- landscape heterogeneity;
- stability and residence times of C additions;
- can decrease C at depth;
- other greenhouse gas emissions.

### Holistic rangeland management

Grazing is an important land use especially in drylands and on degraded lands that are not suitable for cultivation. It is also a major livelihood in developing countries; for example 40 percent of land in Africa is dedicated to pastoralism. However, grazing is also a cause of land degradation with an estimated 100 Tg CO<sub>2</sub>-e (27 million Tg C) emitted annually from grazing-induced desertification (FAO, 2009). Improved grazing management has significant soil C sequestration potential, with wide-reaching co-benefits from restoring productivity to degraded lands. The wide range of conventional grazing practices can be categorised broadly into continuous grazing (CG) and rotational grazing (RG). Conant, Paustian and Elliot (2001) found that improved grazing management increased soil C

by  $0.35 \text{ Mg-C}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$  on average but results were variable between studies. Pastures with a long history of grazing and low productivity showed a mean C increase of 7.7 percent whereas pastures with high productivity and a short grazing history showed a mean soil C loss of 1.8 percent, — likely a reflection of greater capacity to store C in more degraded soils. Studies comparing different grazing intensities showed high intensity grazing increased soil C by  $0.19 \text{ Mg-C}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$  relative to moderate intensity.

There is strong anecdotal evidence that holistic rangeland management (also known as planned grazing, high intensity short duration grazing, cell grazing, time controlled grazing) regenerates degraded land and enhances soil C sequestration. Although it is a type of RG, it is fundamentally different from conventional practices which are rotations determined by animal production and time period, in that timing and duration of grazing and rest periods are determined primarily by plant physiological needs. Properly implemented, stocking rate is very high in order to break up and incorporate litter into the soil, grazing periods are very short to avoid plants being re-grazed when they start to re-shoot, and rest periods are sufficiently long for plants to recover but not so long that they senesce. By stimulating plant growth, net primary productivity is increased, and by avoiding selective grazing, desirable plants are able to compete leading to a shift to a higher proportion of more productive perennials in the pasture.

Although holistic rangeland management was developed over 40 years ago (Savory and Parsons, 1980), it has been the subject of little research. Most studies of improved grazing management have focused on stocking rate or CG/RG comparisons, rather than fundamentally different methods. Nevertheless, Kahn, Earl and Nicholls, (2010) recently found that high intensity short duration grazing resulted in a productive and stable grassland with greater perennial cover, less bare ground and a 78 percent higher stocking rate. While these factors could be expected to enhance soil C sequestration, increased livestock would also lead to high  $\text{CH}_4$  emissions from enteric fermentation. However, these emissions may be reduced by improved nutrition and pasture management (Beauchemin *et al.*, 2008) and the  $\text{CH}_4$  sink provided by grasslands soil (Dalal *et al.*, 2008). Research is needed to estimate the methane balance from grazing and pasture.

Limitations/constraints include:

- low adoption rate;
- higher  $\text{CH}_4$  emissions from enteric fermentation if livestock numbers are increased — although healthy grasslands may absorb  $\text{CH}_4$ .

#### Permanent pasture

Permanent pastures (used here to include modified pastures and rangelands) account for 69 percent or 3.4 billion ha of global agricultural land and are estimated to hold 30 percent of global soil C stock (FAO, 2009). They cover a wide range of agro-ecological zones with different inherent C carrying capacities and potential sequestration rates based on edaphic and climatic factors. Of the 3.4 billion ha of permanent pasture, 73 percent are affected by soil degradation (FAO, 2009a) resulting in reduced soil C and loss of productive capacity; but also providing significant soil C sequestration potential due to the large area covered. In a review of 115 papers, Conant, Paustian and Elliot (2001) found that mean soil C increased by  $0.54 \text{ Mg-C}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$  ( $0.11$  to  $3.04 \text{ Mg-C}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$ ) with improved management and concluded that grasslands could become a significant C sink. Results were influenced by biome and climate with grassland and woodland increasing most. As most inputs of soil C are through root turnover and root exudates, management practices that increase

below ground net primary productivity and improve aggregation and other soil properties that stabilize C would be expected to raise soil C levels over time. Providing irrigation water where water is a limiting factor should increase productivity. However, excess water can lead to water logging and result in emissions of other greenhouse gases, e.g.  $\text{N}_2\text{O}$  and  $\text{CH}_4$ . Effective rainfall can be increased by reducing run-off or evaporation, or by increasing water infiltration and soil water holding capacity, e.g. by maintaining ground cover or improving soil structure. Tongway and Ludwig (2010) estimated that  $4 \text{ Mg-C}\cdot\text{ha}^{-1}$  was sequestered in the top 10 cm of soil over 15 yr at a “water ponding” site in the semi-arid zone of central-western New South Wales, Australia.

Fertilization that provides a supply of limiting nutrients can be expected to increase productivity subject to other potentially limiting factors being available, e.g. water, temperature, sunlight. However, fertilization can result in increased emissions of other greenhouse gases, e.g.  $\text{N}_2\text{O}$ ; and there are significant greenhouse gas emissions from the production and transport of synthetic fertilizers. Biological nitrogen fixation by legumes was found to have a greater influence on soil C sequestration than fertilization (Conant, Paustian and Elliot, 2001). However in pasture ley systems, increasing the time under pasture and reducing cropping time may lead to increased  $\text{CH}_4$  production that counteracts the benefits of C sequestration in the soil. Sowing more productive species of grasses showed the greatest increase in soil C sequestration of the management improvements studied (Conant, Paustian and Elliot, 2001). Legumes also improve fertility and result in higher productivity. Changes in grazing management can also shift pasture composition with a greater proportion of perennials and higher net pasture productivity expected under rotational than under continuous grazing. Wilson and Edwards (2008) have proposed that enteric  $\text{CH}_4$  from ruminants, which make up 11 percent of total greenhouse gas emissions in Australia, could be significantly reduced by the greater adoption and production of low-emission kangaroo meat. This may be a peculiarly Australian solution.

Limitations/constraints include:

- other greenhouse gas emissions such as  $\text{CH}_4$  from ruminants;
- land clearing for pasture establishment reducing above ground C stocks;
- loss of old (stable) C with input of new (labile) C — the “priming effect” (Fontaine *et al.*, 2011).

#### Organic farming

Organic farming has been advocated for more than a century as a movement opposing the widespread use of artificial fertilizers and other agrochemicals in food production. In 2009 the total area (certified and uncertified) was estimated by FAOSTAT to be 29.8 million ha or 0.6 percent of the total agricultural area. The areas in 2007 and 2008 were respectively 28.9 and 29.5 million ha indicating a levelling out of the rate of increase in land utilized for organic farming, which serves what is essentially a niche market. This is in line with calculations by Connor (2008) indicating that organic farming cannot feed the world because of the high cost in terms of land area planted to legumes to meet the demand for N rather than using fertilizers for N inputs.

The rate of C sequestration in organically farmed soil would be expected to be high and FAO (2009b) estimated a potential of  $0.9$ – $2.4 \text{ Pg CO}_2\text{e per annum globally}$ . A key unanswered question is the extent to which organic matter turns over in such soils, the C dynamics of which have not been studied in depth.

Limitations/constraints include:

- few controlled carefully conducted studies (Scialabba and Mueller-Lindenlauf, 2010);

- reduced use of N fertilizers may reduce N<sub>2</sub>O emissions;
- increased tillage to control weeds may increase fossil fuel use and hasten organic matter decomposition.

### Biochar application

The production of biochar from crop residues and other feedstocks has been advocated as a major innovative approach to C sequestration for climate change mitigation (Lehmann, Gaunt and Rondon, 2006; Sohi *et al.*, 2010). Biochar is produced by pyrolysis of biomass at low oxygen levels and temperatures less than 700°C. When applied to soils, biochar is inert and has a long residence time, hence Roberts *et al.* (2010) calculated that the annual rate of C sequestration by utilising all of the “unused” crop residues produced globally would be 0.65 Pg CO<sub>2</sub>e. The idea that so much of the residues of crops are unused may not reflect the real alternative needs of farmers for these residues for animal feed and bedding, for fuel and for protection of the soil surface against erosive forces of water and wind. The value of biochar as a soil amendment derives from the historical practice of using biochar amendments to create the productive Terra Preta soils from acid, infertile soils in the Amazon region. Biochar can increase the nutrient- and water-holding capacity of soils and provides a better substrate for microbes including beneficial mycorrhizae. More research is needed to gain a better understanding of which soil types benefit most from the biochar applications. For example, the C sequestration achieved in studies reviewed by The World Bank (2012) ranged from 2.3 Mg to 3.8 Mg·ha<sup>-1</sup>·yr<sup>-1</sup>, yet actual levels achievable will depend on the availability of feedstock, and the economics of the capital and operating costs of a pyrolysis unit. Transport will also add to the costs. The importance of these variables underlines the need for a life cycle analysis that takes account of the biochar production issues as well as the direct and indirect impacts on greenhouse gases (Roberts *et al.*, 2010).

Limitations/constraints include:

- inconsistency in product quality which may also contain contaminants;
- availability of feedstock and transport may be major constraints;
- markets for trading C do not exist and institutional basis for certification lacking;
- threshold price of C of US\$37 per tonne to make biochar economic;
- needs precautionary research in relation to large scale adoption effects on biodiversity.

### System for rice intensification (SRI)

The system of rice intensification was developed in Madagascar and is based on the use of young seedlings planted at wide spac-

ings. Another key element is the management of water to allow wetting and drying cycles during crop growth. Soil management is based on the use of compost rather than chemical fertilizers, and major increases in yield due to the adoption of the SRI have been claimed. A concerted effort, largely by non-government organizations in 40 countries, has led to two million rice farmers adopting the scheme (Kassam, Stoop and Uphoff, 2011). The use of compost and the reduction in fertilizer use would be expected to increase C sequestration and reduce greenhouse gases such as N<sub>2</sub>O, and the aeration of the soil during wetting and drying cycles would reduce CH<sub>4</sub> production. However, Dumas-Johansen (2009) found little evidence of C sequestration under SRI in Cambodia, and found out that Cambodian farmers do not have sufficient water control to implement SRI. Thus SRI remains a controversial system which requires more research to pinpoint where it can be effective.

Limitations/constraints include:

- wetting and drying soil may increase organic matter decomposition rates (Patrick and Wyatt 1964);
- use of compost may increase sequestration but the restricted use of NPK fertilizers will limit C sequestration.

### Overall assessment of different systems

Several overarching features are noteworthy. Firstly, increases in soil C sequestration may be offset by increased emissions of other more potent greenhouse gases (Table 2). Secondly, the productivity and C sequestration potentials of different systems depends on adequate supplies of stabilizing nutrients such as N, P and S. Ultimately the total impact of land management depends on the actual and potential areas of land under the particular system. On this basis, the most important systems are agroforestry, conservation tillage, holistic rangeland management and permanent pasture.

### Measurements of soil carbon

Many published reports are not based on rigorous measurements of changes in SOC. The main problem with monitoring changes in SOC using conventional methods centres on the need to measure, over a limited number of experimental years, small changes in the relatively large pool of organic C, which has high spatial variability. The long term experiments that help resolve this problem are expensive and relatively rare. Furthermore, since effective sequestration depends on the long-term sustainability of observed changes, so the dynamics as well as the quantity of SOM need to be better understood. In particular, differentiating the labile fraction of SOC from the more passive long-term C can provide such information to be fed into simulation models for use in predictions and economic policy analysis.

**TABLE 2. Current and potential areas, and greenhouse gas impacts of selected land management systems; areas based on FAOSTAT (\* information not available, \*\* numbers are approximate)**

| Management system             | Current area 10 <sup>6</sup> ha (% $\Sigma$ Ag area) | Potential area Increase (10 <sup>6</sup> ha) | Issues                                                    |
|-------------------------------|------------------------------------------------------|----------------------------------------------|-----------------------------------------------------------|
| Agroforestry                  | 1023 (21%)                                           | 630                                          | Above ground C $\uparrow$                                 |
| Conservation tillage          | 100 (2%)                                             | 1252                                         | N <sub>2</sub> O $\uparrow$                               |
| Holistic rangeland management | *                                                    | *                                            | CH <sub>4</sub> $\uparrow$                                |
| Permanent pasture             | 3356 (69%)                                           | *                                            | CH <sub>4</sub> $\uparrow$                                |
| Organic farming               | 30 (0.6%)                                            | *                                            | N <sub>2</sub> O $\downarrow$                             |
| Biochar                       | *                                                    | *                                            | *                                                         |
| System rice intensification** | 2 (0.02%)                                            | 4                                            | N <sub>2</sub> O $\uparrow$<br>CH <sub>4</sub> $\uparrow$ |

### Conventional soil carbon studies

When monitoring changes in SOC following changes to land use and management there are a number of uncertainties associated with methodological strategies and measurement techniques that can be used. Differences in SOC concentration can vary from micro-site scale, to field, farm, landscape, catchment and national scales. A number of heterogeneous characteristics including topography, relief, bulk density, gravel content, nutrient availability, mineralogy, soil texture and structure, previous land use, vegetation composition, spatial variability and seasonal variation can influence the SOC concentration. Therefore, within economic constraints, it is valuable to replicate sampling and analysis as much as possible.

The World Bank (2012) has classified soil C assessment into two broad methods: direct and indirect methods. Direct methods are referred to as methods which measure SOC empirically based on field sampling and laboratory analysis. However, this is costly and time consuming. Indirect methods are those that are based on C simulation models. The advantage of using indirect methods is that they can provide a cost effective means of estimating spatial and temporal changes to SOC at a range of scales, and can also be used for C accounting. In summary the following issues need attention:

- high variability requires longer time frames between measurements;
- dearth of long-term studies;
- bulk density a key parameter;
- landscape dimension not considered sufficiently; lack of awareness and rigour in many studies.

### Role for isotope studies

Isotope techniques provide a unique means for studying decomposition processes and retention of soil C, as well as its interactions with other soil processes and especially nutrients such as N (Celano *et al.*, 2012). Some examples are the following:

- C turnover studies using  $^{13}\text{C}$  natural abundance to track C4 versus C3 plant residues and organic matter. This technique can be used to trace corn crop residues (C4) in soils previously dominated by C3 crops or native vegetation;
- associated studies of real time  $^{13}\text{C}$  dynamics in gases respired in soils with different crop residues. This new methodology provides data on the dynamics of  $^{13}\text{C}$  in systems in which soil moisture and temperature can be varied; useful for studies of the effects of global warming on decomposition and isotope fractionation processes;
- profile studies of  $^{13}\text{C}$  and  $^{15}\text{N}$  abundances to analyse decomposition processes;
- radiocarbon dating of soil C and respired C to distinguish soil organic fractions with short and long mean residence times;
- $^{15}\text{N}$  abundance and C:N ratios in SOM turnover to differentiate biologically fixed N and its contribution to C dynamics and storage;  $^{14}\text{C}$  pulse and continuous labelling for tracing organic C dynamics and particularly the residence time of different C pools.

The objectives of these studies should be clearly defined, particularly the search for the “holy grail” — the labile C fraction that is differentiated from the passive C that has a long residence time. The time is ripe for a coordinated global effort to apply such techniques (especially 1. and 4.) in studies of soil C dynamics at diverse sites where different high priority management systems are compared.

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# A Review of Carbon Sequestration through Conservation Agriculture

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## ABSTRACT

This review aims at developing a clear understanding of the impacts and benefits of the prevalent types of agriculture with respect to soil organic carbon (SOC) sequestration and carbon (C) pools, and examining if there are any misleading findings at present in the scientific literature. Most of the world's agricultural soils have been depleted of organic matter and soil health over the years under tillage-based agriculture (TA), compared with their state under natural vegetation. This degradation process can be reversed and the review identifies conditions that can increase soil organic matter (SOM) content and improve soil health under conservation agriculture (CA) practices which involve minimum soil disturbance, maintenance of soil cover and crop diversity. The review also discusses the need to refer to specific C pools when addressing C sequestration, as each C category has a different turnover rate. With respect to greenhouse gas emissions (GHG), sustainable agricultural systems based on CA principles are described which result in lower emissions from farm operations as well as from machinery manufacturing processes, and that also help to reduce fertilizer use. The review concludes that terrestrial sequestration of carbon can be achieved efficiently by changing the management of agricultural lands from high soil disturbance TA practices to low disturbance CA practices and by adopting effective nitrogen (N) management practices to provide a positive nitrogen balance for C sequestration. However, full advantages of CA in terms of C sequestration can usually be observed only in the medium- to longer-terms when CA practices and associated C sequestration processes in the soil are well established.

**Key words:** *conservation agriculture, soil organic carbon sequestration, environmental conditions; crop rotation, organic matter returned, soil disturbance.*

## INTRODUCTION

Concerns about rising atmospheric carbon dioxide (CO<sub>2</sub>) levels and climate change mitigation efforts have stimulated an interest in using the world's soils for carbon (C) sequestration due to their large sink potential. Soils are important for C management due to their large C content, and also because soil organic carbon (SOC) is particularly responsive to modification through agricultural land use.

Usually, conventional agriculture is tillage-based (TA) in industrialized as well as developing countries and relies as a key operation on mechanical soil tillage with no organic mulch cover for seed bed preparation. This kind of agriculture is generally considered to speed up the loss of soil organic matter (SOM), by increasing its mineralization and through soil loss by erosion. In addition, tillage is a high energy-consuming operation that uses large amounts of fossil fuel per ha in mechanized systems.

In contrast to tillage-based systems, conservation agriculture (CA) is an agro-ecological approach to resource-conserving agricultural production that requires compliance with three linked practical principles, namely: (i) minimum mechanical soil disturbance (with no-till and direct seeding); (ii) maintenance of permanent organic soil cover (with crops, cover crops and/or crop residues); and (iii) species diversification through crop rotations and associations (involving annual and/or perennial crops including tree and pasture crops) (FAO, 2012).

This review aims at developing a clear understanding of the impacts and benefits of the two aforementioned types of agriculture, TA and CA, with respect to the SOC sequestration and C pools, and examining if there are any misleading findings at present in the scientific literature and highlighting the evidence that exposes their flaws.

## METHODOLOGY

The review is based on a desk study of the research literature on soil C sequestration and soil organic matter (SOM) stabilization published in leading peer-reviewed journals.

The relevant literature was reviewed to identify the variables that intervene in CA and TA management systems and produce verifiable estimates of their effect on C accumulation in agricultural soils. The variables analysed were: (i) environmental conditions, (ii) the pattern of the crop rotation, (iii) the quantity and the type of the organic matter returned to the soil, (iv) the management of crop residues, and (v) soil disturbance. This review aims at developing a clear understanding of the impacts and benefits of CA and TA on SOC sequestration, soil C pools, and the potential GHG emissions.

The C footprint of other variables in addition to the above that constitute the CA and TA production cycles was also analyzed because many agronomic practices and methods often recommended to increase C accumulation in soils contain hidden C costs in terms of ancillary greenhouse gas (GHG) emissions. The following variables were included in the review: (i) the C costs attributable to the use of machinery, (ii) GHG effluxes from the soil induced by different treatments, (iii) the use of fertilizers, and (iv) strategies to reduce nutrient leaching, methane and nitrous oxide emissions.

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## RESULTS

There are different C pools in the soil as a result of transformations from the undecomposed form to the decomposed stable form. The C sequestration potential of any soil, for the C pool considered, depends on the vegetation it supports (which influences the amount and chemical composition of organic matter being added), soil moisture availability, soil mineralogical composition and texture, depth, porosity and temperature. Therefore, when addressing C sequestration, rates should always be referred to specific C pools, as each C category has a different turnover rate.

In most soils no measurable C is sequestered when rotations do not return enough biomass to the soil, when crop residues are removed for other concurrent uses and under repeated monocropping in no-till systems (Angers *et al.*, 1997; Wanniarachchi *et al.*, 1999; VandenBygaart, Gregorich and Angers, 2003). Several studies showed that changing from mono-crop to multi-crop rotation results in higher SOC concentration due to a more balanced “diet” the latter provides to soil organisms, although different rotations have different potential to promote and support C sequestration. In general terms, C accumulates in the soil when the nitrogen (N) balance of the crop rotation is positive, i.e. when the input from N fixation or fertilizer is higher than the N exported with harvested produce plus the amount lost by leaching or in gaseous forms (Sidiras and Pavan, 1985; Boddey *et al.*, 1997; Alves *et al.*, 2006).

With respect to SOC accumulation in deeper soil layers, it is quite often reported that when soil sampling is extended deeper than 30 cm, the SOC concentration in deep soil layers is usually higher under TA vis-à-vis undisturbed soil (Centurion and Demattê, 1985; Corazza *et al.*, 1999; Baker *et al.*, 2007). The reason for this is that the top layer, which is C-enriched through fertilization, is turned upside down. However, in this way the recalcitrant C from deeper layers becomes exposed to rapid oxidation and mineralization at the soil surface. Further, SOC accumulation achieved with deeper fertilization regresses as soon as the external C input is interrupted.

Soil disturbance is another very important determinant for soil C accumulation, as many of the factors determining the soil C budget are influenced by land management practices. Most of the world's agricultural soils have been depleted of organic matter and soil health over the years under TA systems compared with their state under natural vegetation. This degradation process has been shown to be reversible and the main ways to increase SOM content and improve soil health seem to be: (i) keeping the contact and interactions between mechanical implements and soil to an absolute minimum, (ii) using effective crop rotations and associations, and (iii) returning crop residues as C source to the soil.

The implementation of these practices can help restore a degraded soil to a sustainable and productive state. However, SOC sequestration is generally non-linear over time (Freibauer *et al.*, 2004), and the effectiveness of conversion of TA to CA depends on many variables: for example, soil C sink strength increases most rapidly soon after a C-enhancing change in land management has been implemented, and reduces with time and the stable SOC stock approaches a new equilibrium (Smith, 2004). Although some authors report significant increases in microbial activity soon after transition to CA, full advantages of CA in terms of soil health can usually be seen only in the medium- to longer-terms when CA practices and soil biological processes become well established within a farming system. To provide an idea of the time scale, Smith (2004) reported that the period for European agricultural soils to reach a new steady state level after a C-enhancing land-use change was introduced is approximately 100 years.

Other aspects relevant for C budgets and GHG emissions are the power and energy requirements. Not tilling the soil results in less fuel consumption, lower working time and slower depreciation rates of equipment per unit area per unit of output, all leading to emission reductions in the farm operations as well as in the machinery manufacturing processes (FAO, 2001). In addition, the crop residues left in CA fields return the C fixed in the crops by photosynthesis to the soil C, resulting in improvement in soil health and fertility leading to reduced fertilizer use and CO<sub>2</sub> emissions. Other GHG emissions from agriculture, namely methane and nitrous oxide, can also be reduced within a CA environment with some complementary practices.

## DISCUSSION

Conservation agriculture allows agro-ecosystems to store more and emit less CO<sub>2</sub> and overall, improves ecosystem functioning and services, such as the control of run-off and soil erosion, C sequestration also below the plough layer and, when a mulch cover is adopted, increase infiltration (Pisante *et al.*, 2010). Despite the beneficial environmental impacts of CA, the main incentives for farmers to shift to it are related to productivity and economics rather than environmental sustainability, i.e. improving farm competitiveness and cutting some of the most relevant production costs, thereby increasing profit margins (Hengxin *et al.*, 2008).

Nevertheless, where TA is deeply rooted in the cultural background, lack of knowledge about CA systems and their management make it particularly difficult for farmers to produce crops without ploughing. Additionally, tillage is efficient in the short term (the season) and it is relatively easy to control since it relies on external inputs and actions that the farmer can control and that give the feeling of delivering fast results (such as fertilizer applications). Most farmers would be able to incorporate chemical nutrients mechanically into the soil, bury weed seeds, and recreate a temporary soil structure on a seasonal basis as a precarious environment favourable for crop growth. Conservation agriculture, on the other hand, is a different approach to farming that requires many technical skills and knowledge to be implemented correctly.

Fewer farmers would know how to set up a crop rotation aimed at producing adequate biomass, providing soil nutrients, reducing weed growth in time, diminishing pest incidence and producing competitive yields. For it to work, CA needs to start a virtuous process in the soil, so that in time soil life and a more balanced ecosystem can reduce labour, the need for external inputs and increase the resilience of the ecosystem. Lack of knowledge about CA systems and their management is why technical extension is crucial in the transition phase. The shift to CA has indeed been achieved where: (i) farmers have been informed of the system and convinced of its benefits by experience, (ii) training and technical support to pioneers have been provided, and (iii) adequate support policies (e.g. funding through C sequestration contracts with farmers) have been implemented.

The important lessons learned from around the world regarding the high potential for C sequestration with CA systems (Lal *et al.*, 1998) and the associated opportunity for C trading and reduction in greenhouse gas emissions should be taken into consideration in any future climate change mitigation and sustainable crop production strategies.

## CONCLUSIONS

The relevant literature was reviewed to identify the most frequent situations in which no C is sequestered under non-traditional agricultural systems. This is most frequently associated with any one or a combination of the following reasons: (i) soil disturbance, (ii)

monocropping, (iii) specific crop rotations, and (iv) poor management of crop residues.

Terrestrial sequestration of C can be achieved efficiently by changing the management of agricultural lands from high soil disturbance practices to low disturbance and by adopting effective N management practices as described by CA.

With CA fewer or smaller tractors can be used and fewer passes over the field done, which also result in lower fuel and repair costs. However, full advantages of CA can usually be obtained only in the medium- to longer-terms when CA practices and soil biological processes are well established.

The combined environmental benefits of CA at the farm and landscape levels can contribute to global environmental conservation and also provide a low-cost option to help offset emissions of the main greenhouse gases.

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# The Use of $^{13}\text{C}$ and $^{15}\text{N}$ Based Isotopic Techniques for Assessing Plant C and N Changes under Conservation Agriculture System

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## SUMMARY

A long-term field experiment was conducted to investigate the effect of tillage and the addition of residues in a wheat–faba bean rotation. The soil was fertilized with a total of 150 kg nitrogen (N)/ha enriched with 9.96 percent nitrogen-15 ( $^{15}\text{N}$ ) atom excess, in four applications. The first crop was corn, a  $\text{C}_4$  plant cropped under till (T) and no-till (NT) conditions. Wheat delta carbon-13 ( $\delta^{13}\text{C}$ ) values changed significantly with the addition of corn residues ( $R^2 = 0.53\text{--}0.64$ ) and with time. The wheat residues had the lowest  $\delta^{13}\text{C}$  ( $-30.5\%$ ) and the seeds the highest ( $-28.5\%$ ). Residue and till treatment (RT) had the highest wheat percent  $^{15}\text{N}$  values (1.5 and 1.9 percent, respectively). Residues and no-till (-) had the lower percent  $^{15}\text{N}$  values (1.1 and 1.1 percent, respectively). Residues increased mineralization by 50 percent as the quantity of  $^{15}\text{N}$  taken up by wheat increased with tillage. Tillage and residues treatments did not affect biological nitrogen fixation (BNF). Following wheat and faba bean, crops derived less N from fertilizer (10 to 2 percent). Residues and tillage increased significantly the percent N derived from fertilizer (Ndff). The wheat crop recovered  $6.45 \text{ kg-N-ha}^{-1}$  (4.3 percent),  $13 \text{ kg-N-ha}^{-1}$  (8.7 percent) and  $11.1 \text{ kg-N-ha}^{-1}$  (7.4 percent) for no residues no-till (NRNT), no-residues and till (NRT), residue and no till (RNT) and residue and till (RT) treatments, respectively. Faba bean recovered less N in all treatments ( $0.66\text{--}1.74 \text{ kg-N-ha}^{-1}$  or 0.4 to 1.2 percent). The fourth wheat crop recovered between  $1.5$  (1 percent) and  $5.63 \text{ kg-N-ha}^{-1}$  (3.7 percent). Cumulative N recovery during the four growing seasons was  $122 \text{ kg-N-ha}^{-1}$  (81.5 percent) for RT and  $116 \text{ kg-N-ha}^{-1}$  (77.1 percent) for RNT. Less N was recovered in the NRNT treatments in the four cropping seasons:  $104 \text{ kg-N-ha}^{-1}$  (69.3 percent) and  $106 \text{ kg-N-ha}^{-1}$  (71 percent) respectively. Nitrogen not recovered by the crops amounted to 18.5 percent for RT, 22.9 percent for RNT, 31 percent for NRNT and 29 percent for NRT. Most of the N not recovered was in the soil organic matter (SOM).

**Key words:** conservation agriculture,  $^{13}\text{C}$ ,  $^{15}\text{N}$ , soil organic matter, tillage.

## INTRODUCTION

The effect of drought and changing climate continue to reduce cereal production in Morocco. Conservation agriculture (CA) based on management of crop residues reduces soil degradation, increases water

and nitrogen (N) fertilizer use efficiency, sustains yields and improves soil fertility and structure. Long-term sustainability of crop production is the agricultural strategy adopted worldwide and this can only be reached through good management of water resources and soil fertility and giving particular attention to soil N (mineralization and immobilization) (Galloway *et al.*, 2008). Zeller and Dambrine (2011) traced the fate of litter-derived N in beech forest soils and showed that after 4–5 years on litter decomposition, most of the nitrogen-15 ( $^{15}\text{N}$ ) input was present as particulate organic matter and that litter N became rapidly re-available for trees. Natural abundance stable isotope investigations in plant ecology have focused largely on variations in bulk delta carbon-13 ( $\delta^{13}\text{C}$ ) (Bowling, Pataki and Randsen, 2008). Recently-formed soil organic matter (SOM) may follow different pathways of mineralization and/or stabilization depending on soil properties and climate conditions (von Lutzow *et al.*, 2006). Smith *et al.* (2000) showed that  $\delta^{13}\text{C}$  values of plants can be used to predict irrigation needs and optimize production. Crop residues reduce the effect of drought and improve water and N storage in soil and protect soil from the impacts of high temperatures (Cassman *et al.*, 2003). The wide diversity of  $\delta^{13}\text{C}$  values of grassland plants was found to be due to environmental conditions (Dungate *et al.*, 2010). Previous studies have reported differences in leaf bulk  $\delta^{13}\text{C}$  values between life forms (Zheng and Shangguan, 2007). Only between 30 percent and 50 percent of fertilizer N is taken up by crops (Ladha *et al.*, 2005), the remainder becoming part of the SOM or being lost (Tillman, 1999). Nitrogen that remains immobilized will lead to reduced N losses via leaching and/or denitrification (Scow, 1997). Soil temperature and humidity affect the activity of soil micro-organisms and increasing soil temperature induces rapid SOM degradation and increased emission of carbon dioxide ( $\text{CO}_2$ ) (Grall *et al.*, 2006).

The  $^{15}\text{N}$  isotope dilution technique is used to investigate N recovery by crops, available soil N and SOM-N pools (IAEA, 2003). Soil organic matter has the ability to store nutrients and improve soil structure, and has long been used as a key indicator of the sustainability of cropping systems (El Alami and Ismaili, 2007a). The form of N application, i.e. organic versus inorganic or in combination, may affect the amount of C that becomes sequestered (Moran *et al.*, 2005). Carbon isotope discrimination ( $\delta^{13}\text{C}$ ) was correlated with grain yield under water stress conditions (Monneveux *et al.*, 2005). The  $\delta^{13}\text{C}$  values decreased with increased water stress and increased with increasing N stress (Stewart *et al.*, 1995). These authors also reported a negative correlation between  $\delta^{13}\text{C}$  values of the plant and water use efficiency (WUE), with water stress and increasing N availability improving WUE. The use of minimum tillage and mulching limits the potential for water erosion (Pansak *et al.*, 2007), the relationship and slope between available  $\text{NO}_3\text{-N}$  and  $\delta^{13}\text{C}$  values suggesting that

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differences in  $^{13}\text{C}$  isotopic discrimination were more related to availability of N than to differences in water availability. In a thorough literature review, Giller *et al.* (2009) showed positive effects of CA on water availability (reduced soil evaporation, reduced water runoff, increased water infiltration and reduced soil temperature).

Cerri *et al.* (1985) used differences in  $\delta^{13}\text{C}$  values of  $\text{C}_3$  plants (varying from  $-24$  to  $-34\text{‰}$ ) and  $\text{C}_4$  plants (range  $-6$  to  $-19\text{‰}$ ) to evaluate WUE in arid environments. The  $^{13}\text{C}$  abundance of incoming SOM is preserved during mineralization and humus formation and the  $\delta^{13}\text{C}$  content of SOM reflects the  $^{13}\text{C}$  content of the vegetation. A significant water and N effect on maize  $\delta^{13}\text{C}$  values was shown by Dercon *et al.* (2006). If a change occurs from  $\text{C}_3$  to  $\text{C}_4$  plants at a known time, it is possible to infer the turnover time of the SOM. The  $^{13}\text{C}$  natural abundance technique provides useful data to test SOM models in complex cropping systems in which both  $\text{C}_3$  and  $\text{C}_4$  plants are intercropped or rotated (Diels *et al.*, 2004). The  $^{13}\text{C}$  natural labelling in combination with modelling was used by Diels *et al.* (2000) to study long-term SOM changes in agro-forestry systems through investigation of yearly changes of  $\delta^{13}\text{C}$  in plant materials. Important and consistent within-year variations were observed for legume tree pruning (up to  $2.4\text{‰}$   $\delta^{13}\text{C}$ ) and weeds (up to  $7\text{‰}$   $\delta^{13}\text{C}$ ) pointing to the need for repeated sampling within a single year.

The objectives of this study were to: (i) evaluate the beneficial effects of a CA system (no-till and residues left on the soil) on a wheat-faba bean rotation, (ii) estimate the extent of biodegradation of corn crop residues through time and quantify the contribution of residues to wheat and faba bean N nutrition and BNF, and (iii) determine the effect of corn residues on the  $\delta^{13}\text{C}$  values of follow-on crops (wheat and faba bean).

## MATERIALS AND METHODS

A field experiment was conducted to investigate a CA system based on wheat (*Triticum durum* Oum Rbia G3 2004) — faba bean (*Vicia faba* L.) — wheat rotation. Average rainfall was  $500\text{ mm/yr}$  and the experiment lasted four years (four cropping seasons). The soil was a well-drained sandy loam of poor fertility and low water holding capacity. The first crop was corn (*Zea mays* var. *rugosa*), a  $\text{C}_4$  plant seeded on 16 plots ( $16\text{ m}^2$ ) fertilized with  $150\text{ kg N ha}^{-1}$  in four applications:  $25\text{ kg N ha}^{-1}$  at seeding,  $25\text{ kg N ha}^{-1}$  two weeks after seeding,  $50\text{ kg N ha}^{-1}$  one month after seeding and  $50\text{ kg N ha}^{-1}$  two months after seeding. Nitrogen was added as ammonium sulphate enriched with 9.96 percent atom  $^{15}\text{N}$  excess at all applications. The corn crop (a  $\text{C}_4$  plant) was used to enrich the soil with  $\text{C}_4$  plant residues ( $23\text{ 825 kg/ha}$  of residues or  $9616\text{ kg/ha}$  total C, with a  $\delta^{13}\text{C} = -12.4\text{‰}$ , and  $340\text{ kg/ha}$  total N labelled with 4.28 percent atom excess  $^{15}\text{N}$  (Vanlauwe *et al.*, 2001).

Soil was amended with phosphate ( $100\text{ kg P}_2\text{O}_5/\text{ha}$ ) and potassium ( $40\text{ kg K}_2\text{O/ha}$ ) before the corn crop was established. The corn crop was irrigated as needed, but the following wheat (second crop) and faba bean (third crop) crops were only rain-fed (El Alami and Ismaili, 2007b). The second crop was wheat (a  $\text{C}_3$  plant) planted after the corn under no-till and till and with or without corn residues left on the soil. The first wheat crop of the wheat-faba bean system was sampled three times during the first growing season: at one and two months after seeding, and at the end of the season. After the first wheat crop, subsequent crops (faba bean as a third crop and wheat as fourth crop in the wheat-alfalfa system after the first crop of corn to provide  $\text{C}_4$  plant residues) were sampled only at harvest time. The  $\delta^{13}\text{C}$  and percent  $^{15}\text{N}$  values were determined at each sampling time and at harvest.

After the first corn crop, the treatments were NRNT — no residues, no-tillage, NRT — no residues with tillage, RT — with residues

and tillage, RNT — with residues and no tillage. The percent C and  $^{13}\text{C}$  natural abundance, percent N and percent  $^{15}\text{N}$  excess were determined simultaneously with a Europa Scientific ANCA-SL stable isotope mass spectrometer. Carbon-13 natural abundance was expressed in  $\delta^{13}\text{C}$  units using the international PDB standards. Nitrogen-15 and  $\delta^{13}\text{C}$  analyses were performed at the IAEA Analytical Laboratory at Seibersdorf, Austria. Crop residue inputs were quantified by weighing fresh residues on each plot and calculating the dry weight using the dry matter contents of sub-samples. The results were analysed using an ANOVA with  $p \geq 0.05$  and linear regression.

## RESULTS AND DISCUSSION

After a corn crop and addition of corn residues, the following wheat plant  $\delta^{13}\text{C}$  values changed significantly with time (Table 1), being higher at two months than at one month after seeding. The greatest difference was found in the NRNT treatment ( $-1.05\text{‰}$ ) while for the treatments NRT, RT and RNT the difference was lower ( $-0.54\text{‰}$ ) (Table 1).

At the end of the season, the  $\delta^{13}\text{C}$  values were higher than at one or two months after seeding. In all treatments, the seeds had higher  $\delta^{13}\text{C}$  values than the leaves and plant residues. The RNT treatment produced the largest difference in  $\delta^{13}\text{C}$  values between residues and seeds ( $-2.3\text{‰}$ ) while NRNT and NRT showed the smallest difference ( $-1.5\text{‰}$ ). Residue treatment resulted in a difference of  $-1.8\text{‰}$  as well as an increase of  $\delta^{13}\text{C}$  from the source (leaves) to the sink (seeds). The addition of  $\text{C}_4$  plant residues (i.e., corn residues) increased the  $\delta^{13}\text{C}$  values of wheat residues and grain, but tillage had no significant effect on these values (Table 1), while the difference between the till and no-till treatments reached a maximum of  $0.5\text{‰}$ . Regression of  $\delta^{13}\text{C}$  values with wheat total N showed a relation between both factors for the first and second harvests and for grain at the end of the growing season. One month after seeding, wheat  $\delta^{13}\text{C}$  values in one and two months after seeding were correlated with plant total N ( $R^2 = 0.53\text{--}0.64$ ) and at the end of the growing season  $\delta^{13}\text{C}$  values of the grain were still correlated with plant total N ( $R^2 = 0.53$ ). The  $\delta^{13}\text{C}$  values of wheat were significantly affected by the previous  $\text{C}_4$  corn crop and by the residues left on the surface of the soil. These values increased from  $0.77$  to  $2.63\text{‰}$  for the grain and from  $0.5$  to  $1.54\text{‰}$  for residues, the grain of NRT treatment showing the highest increase (from  $-29.03$  to  $-26.4\text{‰}$ ) followed by RT treatment (from  $-28.67$  to  $-27.07\text{‰}$ ). Dungait *et al.* (2010) found that total mean leaf  $\delta^{13}\text{C}$  values for 26 plants from Manor Farm were  $-28.8\text{‰}$  with a range spanning  $7.5\text{‰}$ . Therefore tillage induced a much greater effect by the corn residues on wheat  $\delta^{13}\text{C}$  values. The residues affected the  $\delta^{13}\text{C}$  values of the following wheat and the tillage induced an additional increase due to organic matter mineralization.

The legume faba bean  $\delta^{13}\text{C}$  values were less affected by addition of residues to the soil and changed only with the tillage treatment ( $1.25\text{‰}$ ) (Table 1). Dungait *et al.* (2010) demonstrated that the wide range of  $\delta^{13}\text{C}$  values of grassland plants could be ascribed to grazing management, interspecific and spatiotemporal influences. Total N of crops correlated well with  $\delta^{13}\text{C}$  values, the  $R^2$  being equal to  $0.26$  to  $0.83$ . Correlation was best with the fourth (wheat) crop and increased from the first (corn) to the fourth crop.

The percent  $^{15}\text{N}$  of wheat was affected significantly by the addition of residues and tillage treatments. At one month after seeding, the difference was not significant, but at one and two months after seeding, the treatment RT had the highest percent  $^{15}\text{N}$  values ( $1.5$  and  $1.9$  percent, respectively) showing the effect of tillage on residue mineralization and the higher contribution of residues to the N nutrition of wheat when the soil was tilled. When the residues were

**TABLE 1.** Effect of time, residues and tillage on dry weight, total N, % $^{15}\text{N}$  and plant  $\delta^{13}\text{C}$  of wheat planted after corn in a rotation system of wheat — faba bean — wheat cropped under CA treatments. The first wheat crop was sampled at one month after seeding (M1), two months after seeding (M2) and at the end of season (grain and residues). In each column, means with different a, b, c, d letters are significantly different at  $p \geq 0.05$

| Dry weight (kg/ha) |                    |                    |                    |                    |                    |                    |
|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|
|                    | Wheat              |                    | Faba bean          |                    | wheat              |                    |
|                    | Grain              | Residues           | Grain              | Residues           | Grain              | Residues           |
| NRNT               | 4 357 <sup>c</sup> | 6 130 <sup>b</sup> | 1 987 <sup>a</sup> | 3 012 <sup>a</sup> | 4 997 <sup>a</sup> | 6 031 <sup>a</sup> |
| NRT                | 4 336 <sup>c</sup> | 7 298 <sup>b</sup> | 2 025 <sup>a</sup> | 3 685 <sup>a</sup> | 4 716 <sup>a</sup> | 6 495 <sup>a</sup> |
| RT                 | 5 726 <sup>c</sup> | 6 735 <sup>b</sup> | 2 906 <sup>b</sup> | 3 267 <sup>a</sup> | 5 665 <sup>a</sup> | 6 562 <sup>a</sup> |
| RNT                | 6 500 <sup>b</sup> | 7 226 <sup>b</sup> | 2 956 <sup>b</sup> | 3 672 <sup>a</sup> | 6 170 <sup>b</sup> | 7 575 <sup>b</sup> |

| Total N (kg·N·ha <sup>-1</sup> ) |                    |                   |                    |                    |                   |
|----------------------------------|--------------------|-------------------|--------------------|--------------------|-------------------|
|                                  | Wheat              |                   | Faba               | wheat              |                   |
|                                  | Grain              | Residues          | Plant              | Grain              | Residues          |
| NRNT                             | 66.52 <sup>a</sup> | 34 <sup>a</sup>   | 68.53 <sup>a</sup> | 71.62 <sup>a</sup> | 23.1 <sup>a</sup> |
| NRT                              | 82.31 <sup>b</sup> | 31.8 <sup>a</sup> | 71.53 <sup>a</sup> | 116.9 <sup>b</sup> | 35.6 <sup>b</sup> |
| RT                               | 91.3 <sup>b</sup>  | 33.6 <sup>a</sup> | 126.7 <sup>b</sup> | 91.06 <sup>b</sup> | 31.1 <sup>b</sup> |
| RNT                              | 118.5 <sup>c</sup> | 32 <sup>a</sup>   | 105.4 <sup>b</sup> | 87.63 <sup>b</sup> | 24.8 <sup>a</sup> |

| Plant $\delta^{13}\text{C}$ (‰) |                     |                     |                     |                     |                     |                     |                     |
|---------------------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
|                                 | Wheat               |                     | Faba bean           |                     | Wheat               |                     |                     |
|                                 | M1                  | M2                  | Grain               | Residues            | Plant               | Grain               | Residues            |
| NRNT                            | -30.8 <sup>a</sup>  | -31.85 <sup>a</sup> | -28.71 <sup>a</sup> | -30.20 <sup>a</sup> | -28.85 <sup>a</sup> | -27.77 <sup>a</sup> | -29.7 <sup>a</sup>  |
| NRT                             | -30.78 <sup>a</sup> | -31.32 <sup>a</sup> | -29.02 <sup>b</sup> | -30.51 <sup>b</sup> | -28.58 <sup>a</sup> | -26.4 <sup>b</sup>  | -29.17 <sup>b</sup> |
| RT                              | -31.08 <sup>b</sup> | -31.62 <sup>a</sup> | -28.66 <sup>a</sup> | -30.47 <sup>b</sup> | -29.86 <sup>b</sup> | -27.07 <sup>c</sup> | -29.24 <sup>b</sup> |
| RNT                             | -30.57 <sup>b</sup> | -31.13 <sup>a</sup> | -28.56 <sup>a</sup> | -30.85 <sup>c</sup> | -28.61 <sup>a</sup> | -27.79 <sup>a</sup> | -29.31 <sup>b</sup> |

| % $^{15}\text{N}$ (atom % $^{15}\text{N}$ excess) |                    |                    |                    |                    |                    |                    |                    |                    |                    |
|---------------------------------------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|
|                                                   | Wheat              |                    | Faba               |                    | Wheat              | Ray                |                    | Corn               |                    |
|                                                   | M1                 | M2                 | Grain              | Residues           | Plant              | Grain              | Residues           | Plant              | Plant              |
| NRNT                                              | 0.928 <sup>a</sup> | 1.505 <sup>a</sup> | 0.939 <sup>c</sup> | 0.988 <sup>b</sup> | 0.144 <sup>a</sup> | 0.246 <sup>a</sup> | 0.209 <sup>a</sup> | 0.416 <sup>a</sup> | 1.261 <sup>a</sup> |
| NRT                                               | 1.170 <sup>a</sup> | 1.193 <sup>a</sup> | 0.831 <sup>c</sup> | 0.882 <sup>b</sup> | 0.364 <sup>a</sup> | 0.28 <sup>a</sup>  | 0.275 <sup>a</sup> | 0.548 <sup>a</sup> | 1.251 <sup>a</sup> |
| RT                                                | 1.464 <sup>a</sup> | 1.895 <sup>b</sup> | 1.542 <sup>b</sup> | 1.610 <sup>c</sup> | 0.19 <sup>a</sup>  | 0.705 <sup>b</sup> | 0.638 <sup>b</sup> | 0.538 <sup>a</sup> | 0.412 <sup>b</sup> |
| RNT                                               | 1.174 <sup>a</sup> | 1.287 <sup>a</sup> | 1.103 <sup>c</sup> | 1.118 <sup>b</sup> | 0.17 <sup>a</sup>  | 0.451 <sup>b</sup> | 0.394 <sup>a</sup> | 0.558 <sup>a</sup> | 0.763 <sup>b</sup> |

added to soil, the effect of tillage on percent  $^{15}\text{N}$  was significant (Table 1). At the end of the season, a significant difference was found between the residues and tillage treatments. The RT treatment maintained the highest percent  $^{15}\text{N}$  values in the grain (1.5 percent) and in the residues (1.6 percent), while RNT had the second lowest values in the grain (1.1 percent) and residues (1.1%), showing the effect of residues and tillage on the N nutrition of wheat. Tillage increased N mineralization by more than 50 percent. When no residues were left on the soil, tillage did not have a significant effect on the percent  $^{15}\text{N}$  of wheat.

Total  $^{15}\text{N}$  accumulated by the wheat was affected significantly by residue and tillage treatments. Total  $^{15}\text{N}$  of the plant did not correlate with  $\delta^{13}\text{C}$  at the first crops and showed a positive relation at the fourth wheat crop ( $R^2 = 0.63$  for grain and 0.53 for residues). When residues were left on the soil, the wheat plants accumulated more N and the quantity of  $^{15}\text{N}$  taken up increased with tillage (Table

1). Wheat accumulated  $^{15}\text{N}$  from residues and from soil in NRNT, NRT, RT and RNT treatments. During the four cropping seasons, percent  $^{15}\text{N}$  decreased in all treatments demonstrating high rates of OM degradation in the soil whether tilled or not tilled. The percent  $^{15}\text{N}$  of faba bean did not show significant treatment differences. Two control plants were used to determine  $\text{N}_2$  fixation and differences were not found in the percent  $^{15}\text{N}$  with any treatment. Residues and tillage increased significantly the total  $^{15}\text{N}$  in all crops, showing the positive effect of residues and tillage on N nutrition of the crops. Tillage and residues treatments did not affect BNF by faba bean (Table 2).

After corn at the wheat crop, less N was derived from fertilizer (10 percent). At the end of the cropping season, percent Ndff values showed significant differences between residues and tillage treatments. First, residues increased these values significantly (15.4 percent in grain and 16.1 percent in residues) in RT and (11 percent and 11.2 percent) in RNT compared with no residues (9.4 percent

**TABLE 2. Effect of tillage and residues addition on %Nd<sub>fa</sub> of faba bean in a rotation system of wheat and faba bean planted under CA when rye grass (%Nd<sub>fa1</sub>) and corn (%Nd<sub>fa2</sub>) were used as control plants**

| Controls | %Nd <sub>fa1</sub> | %Nd <sub>fa2</sub> |
|----------|--------------------|--------------------|
| NRNT     | 65.38              | 88.58              |
| NRT      | 33.58              | 70.9               |
| RT       | 64.68              | 53.88              |
| RNT      | 69.53              | 77.72              |

in grain and 9.9 percent in residues) in NRNT and (8.3 percent and 8.9 percent) in NRT. Second, tillage also increased the percent Nd<sub>ff</sub>, but not significantly. In the no-residue treatment, the effect of tillage on fertilizer use was lower than when residues were left on the soil. For the faba bean, the third crop, percent Nd<sub>ff</sub> was low because of BNF. At the fourth crop, wheat derived less N from fertilizer and the recovery was low (2–7 percent). Dourado *et al.* (2009) found that the average recovery of fertilizer and residue in the soil after the first growing season was 38 percent (residues added) and 71.2 percent (residues removed). On average, an additional 4.8 percent of the <sup>15</sup>N fertilizer and 6.6 percent of <sup>15</sup>N residue was recovered by the crop during the 2<sup>nd</sup> and 3<sup>rd</sup> growing season (Table 3).

Small quantities of fertilizer N were recovered in the second (wheat) crop. When no residues were added to the soil, N recovery was 6.5 kg-N-ha<sup>-1</sup> (4.3 percent) under NRNT and NRT treatments. When residues were added 13 kg-N-ha<sup>-1</sup> (8.7 percent) were recovered under RT and 11.1 kg-N-ha<sup>-1</sup> (7.4 percent) under RNT (Table 4). Recovery was higher in RT and RNT because of the additional N added to soil through mineralization of corn residues. Less N was recovered from faba bean under all treatments (between 0.66 kg-N-ha<sup>-1</sup> (0.4 percent) and 1.7 kg-N-ha<sup>-1</sup> (1.2 percent)). Recovery from the fourth wheat crop was between 1.5 and 5.6 kg-N-ha<sup>-1</sup> (1–3.7 percent). With the residue and till treatments, N recovery from fertilizer was greater than from all the other treatments. Cumulative N recovery during the four growing seasons was 122 kg-N-ha<sup>-1</sup> (81.5 percent) for RT and 116 kg-N-ha<sup>-1</sup> (77.1 percent) for RNT. In the four

cropping seasons recoveries under NRNT and NRT were respectively 104 (69.3 percent) and 106 kg-N-ha<sup>-1</sup> (71 percent). The percentages of N not recovered by the crops were respectively 18.5 for RT, 22.9 for NRT, 31 for NRNT and 29 for NRT (Table 4). Most of the N not recovered was in the SOM.

## CONCLUSIONS

The use of <sup>13</sup>C isotopic discrimination together with <sup>15</sup>N labelled fertilizer in CA experiments showed that leaving crop residues on the soil and no tillage improved soil organic C as reflected in <sup>13</sup>C signature of the crop residues in the subsequent crops) and the N nutrition of the crops as shown by <sup>15</sup>N dilution. The use of C<sub>4</sub> plant crop residues as a tracer of C in the subsequent crops is a good indicator of N nutrition of crops and soil organic carbon stocks. Addition of residues to soil affected <sup>13</sup>C discrimination of grasses but had no effect on legumes. Total <sup>15</sup>N of crops did not correlate with plant δ<sup>13</sup>C values.

The residue and tillage treatments had the highest percent <sup>15</sup>N values and increased mineralized N by 50 percent. When the corn residues were left on the soil the subsequent wheat plants accumulated more N. Biological N<sub>2</sub> fixation was reduced significantly by water stress, increased by residue additions and was not affected by tillage. Most of the fertilizer N was recovered by the first crop; less N was recovered from the following crops. Nitrogen recovery increased with the addition of residues and with tillage. Most N not recovered by the crops was held in the SOM. Maintaining organic matter levels in the soil therefore remains a crucial component of sustainable agricultural practices.

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**TABLE 4. Recovery of fertilizer N in crops as affected by season, residues and tillage in a rotation system of wheat and faba bean planted under CA treatments. In each column, means with different a, b, c, d letters are significantly different at p ≥ 0.05.**

| Recovery of fertilizer N (kg-N-ha <sup>-1</sup> ) |                   |                   |                   |                   |                   |                   |                   |                    |
|---------------------------------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|--------------------|
|                                                   | Wheat             |                   |                   | Faba              | Wheat             |                   |                   | Total              |
|                                                   | Grain             | Residues          | Plant             | Plant             | Grain             | Residues          | Plant             | recovery           |
| NRNT                                              | 4.18 <sup>a</sup> | 2.25 <sup>a</sup> | 6.43 <sup>a</sup> | 0.66 <sup>a</sup> | 1.18 <sup>a</sup> | 0.32 <sup>a</sup> | 1.5 <sup>a</sup>  | 104.0 <sup>a</sup> |
| NRT                                               | 4.58 <sup>a</sup> | 1.88 <sup>a</sup> | 6.46 <sup>a</sup> | 1.74 <sup>b</sup> | 2.19 <sup>b</sup> | 0.66 <sup>b</sup> | 2.85 <sup>b</sup> | 106.5 <sup>a</sup> |
| RT                                                | 9.42 <sup>b</sup> | 3.62 <sup>b</sup> | 13 <sup>b</sup>   | 1.61 <sup>b</sup> | 4.3 <sup>c</sup>  | 1.33 <sup>c</sup> | 5.63 <sup>c</sup> | 122.2 <sup>b</sup> |
| RNT                                               | 8.75 <sup>b</sup> | 2.39 <sup>c</sup> | 11.1 <sup>c</sup> | 1.2 <sup>c</sup>  | 2.65 <sup>d</sup> | 0.65 <sup>b</sup> | 3.3 <sup>d</sup>  | 115.7 <sup>c</sup> |

| Percent N recovery of fertilizer N |       |         |       |       |       |         |       |       |           |
|------------------------------------|-------|---------|-------|-------|-------|---------|-------|-------|-----------|
|                                    | Wheat |         |       | Faba  | Wheat |         |       | Total | Not       |
|                                    | Grain | Residue | Plant | Plant | Grain | Residue | Plant |       | recovered |
| NRNT                               | 2.79  | 1.5     | 4.29  | 0.44  | 0.79  | 0.22    | 1     | 69.35 | 30.6      |
| NRT                                | 3.05  | 1.25    | 4.3   | 1.16  | 1.46  | 0.44    | 1.9   | 71.01 | 29.0      |
| RT                                 | 6.28  | 2.41    | 8.7   | 1.07  | 2.86  | 0.89    | 3.75  | 81.49 | 18.5      |
| RNT                                | 5.83  | 1.6     | 7.43  | 0.8   | 1.76  | 0.44    | 2.2   | 77.13 | 22.9      |

**TABLE 3.** Effect of season, residues and tillage on %Ndff of wheat and faba bean cropped in a rotation system of wheat and faba bean planted under CA treatments. The first wheat crop was sampled at one month after seeding (M1), two months after seeding (M2) and at the end of season (grain and residues). In each column, means with different a, b, c, d letters are significantly different at  $p \geq 0.05$ .

|      | Wheat             |                   |                   |                   | Faba bean         | Wheat             |                   |
|------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
|      | M1                | M2                | Grain             | Residues          | Plant             | Grain             | Residues          |
| NRNT | 9.28 <sup>a</sup> | 15.1 <sup>a</sup> | 9.4 <sup>b</sup>  | 9.88 <sup>b</sup> | 1.44 <sup>a</sup> | 2.46 <sup>a</sup> | 2.09 <sup>a</sup> |
| NRT  | 11.7 <sup>a</sup> | 11.9 <sup>a</sup> | 8.31 <sup>b</sup> | 8.82 <sup>b</sup> | 3.65 <sup>b</sup> | 7.05 <sup>b</sup> | 6.38 <sup>b</sup> |
| RT   | 14.6 <sup>a</sup> | 19 <sup>c</sup>   | 15.4 <sup>c</sup> | 16.1 <sup>c</sup> | 1.9 <sup>a</sup>  | 2.8 <sup>a</sup>  | 2.75 <sup>a</sup> |
| RNT  | 11.7 <sup>a</sup> | 12.9 <sup>a</sup> | 11 <sup>b</sup>   | 11.2 <sup>b</sup> | 1.71 <sup>a</sup> | 4.52 <sup>a</sup> | 3.94 <sup>a</sup> |

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# Biological Nitrogen Fixation by Soybean and Fate of Applied $^{15}\text{N}$ -Fertilizer in Wheat under Conservation Agriculture

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## ABSTRACT

Nitrogen (N) is one of the key drivers of global agricultural production. Four field experiments with irrigated summer-grown soybean (*Glycine max* L.) and winter-grown wheat (*Triticum aestivum* L.) were conducted during 2006–08 to investigate whether biological  $\text{N}_2$  fixation (BNF) by soybean can be improved under conservation agriculture (CA) where crop residues (CR) retained on the soil surface, as compared with conventional tillage (CT) and to assess the fate of N fertilizer in subsequent wheat on Fatehpur loamy sand soil (*Typic Haplustept*) using  $^{15}\text{N}$  labelled fertilizer. The BNF estimated using  $^{15}\text{N}$  isotope dilution and  $^{15}\text{N}$  natural abundance methods, were comparable suggesting that the latter method which does not require costly  $^{15}\text{N}$ -enriched fertilizer, could be employed to estimate BNF by legumes. Use of sorghum as a reference plant, grown in the same plot as soybean, led to up to 36 percent lower estimation of BNF than with the use of *in situ* spontaneous weeds (*Eleusine aegyptiacum* L., *Euphorbia hirta* L. and *Cynodon dactylon* L.), which have a similar size and rooting depth as the soybean. The irrigated soybean in the semi-arid subtropical soils could biologically fix 81–125 kg-N-ha<sup>-1</sup> (68–85 percent of total N uptake), depending upon tillage and CR management. Significant increases in BNF by soybean were recorded when CR was retained on the soil surface of CA presumably due to better rhizobia activity of caused by cooler rhizosphere environment. Recovery of applied  $^{15}\text{N}$  in the soil–plant system at harvest of the wheat crop showed that 36–47 percent of it was utilized by the crop, 37–49 percent was left in the soil profile and 5–27 percent was lost, which was estimated as unrecovered  $^{15}\text{N}$ . Utilization of  $^{15}\text{N}$  was significantly lower when 25 percent more fertilizer N was applied than recommended in both CT and CA without CR. It was also lower when CR was incorporated in CT, or retained on the soil surface in CA. The recovery of  $^{15}\text{N}$  in the soil profile at harvest of the wheat crop revealed that the majority of the residual fertilizer N was present in the first 15 cm (54–61 percent), although downward movement of  $^{15}\text{N}$  below this level of the soil surface layer was also evident in the soil profile up to a depth of 120 cm. These results illustrate the benefits of CA with CR retained on soil surface on BNF by soybean, and similar patterns in N uptake and translocation from

vegetative parts to grain and utilization of applied N by wheat in both tillage systems.

**Keywords:** *Conventional tillage, reduced tillage,  $^{15}\text{N}$  isotope dilution technique,  $^{15}\text{N}$  natural abundance technique,  $^{15}\text{N}$  balance, denitrification.*

## INTRODUCTION

Nitrogen (N) is an important nutrient for global agricultural production as 150–200 million tonnes (t) mineral N are required each year by crops (Unkovich *et al.*, 2008). These large N requirements are met mainly through application of N fertilizers, soil organic matter (SOM), and biological  $\text{N}_2$  fixation (BNF) by legumes. Improvements in the build-up of SOM, BNF and the plant use efficiency of fertilizer N, and reduction in leaching and gaseous losses of N are vital to the long-term sustainability of agricultural systems, conservation of natural resources and environmental quality. Conservation agriculture (CA) including continuous soil cover through retention of crop residues on the soil surface (CR), with no or reduced tillage, the use of cover crops and the inclusion of grain legumes or green manure crops in rotations, have shown many positive benefits for increasing agricultural productivity and systems' sustainability in several parts of the world (Hobbs, 2007). However, limited information is available to support such claims in India.

Efforts are being made to develop efficient production technology for soybean (*Glycine max* L.) to replace some of the area under the predominant summer-grown rice crop (Aulakh, Pasricha and Bahl, 2003; Aulakh *et al.*, 2010, 2012). Information about the potential of irrigated soybean for BNF in soils of the semi-arid subtropical regions of South Asia is limited.  $^{15}\text{N}$  isotopic dilution and  $^{15}\text{N}$  isotope abundance techniques have been widely used for estimating BNF by legumes (Boddey *et al.*, 1995; Chalk and Ladha, 1999; Unkovich *et al.*, 2008). While the  $^{15}\text{N}$  isotopic dilution method requires costly  $^{15}\text{N}$ -enriched fertilizer, the  $^{15}\text{N}$  natural abundance method is based on natural abundance of  $^{15}\text{N}$ . Therefore, if both techniques provide comparable BNF estimates, the  $^{15}\text{N}$  natural abundance method is much cheaper to use. In addition to the potential of soybean for BNF under conventional tillage (CT) and CA without or with CR, the fate of fertilizer N applied to wheat (*Triticum aestivum* L.) under such systems is not known. Nitrate ( $\text{NO}_3^-$ ) enrichment of groundwater beneath irrigated soils of semi-arid subtropical regions is evident from the  $\text{NO}_3^-$  analysis of tubewell waters (Bajwa, Bijay-Singh and Parminder-Singh, 1993). However no data have been reported on the movement of  $^{15}\text{N}$  fertilizer in the profile of coarse-textured porous irrigated soils under CA and CR management systems. Such studies would become

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more important as porous soils are used increasingly for irrigated cropping systems (Aulakh and Bijay-Singh, 1997).

Among the various concerted efforts made to expand CA in Asia, a four-year field study was conducted with soybean-wheat rotation under CT and CA with or without CR on irrigated soil in the semi-arid subtropical region. The results on crop yields, nutrient uptake, water storage in the soil profile, and temperature dynamics were presented earlier (Aulakh et al., 2012). The present study investigated if (i) BNF by soybean is improved under CA as compared with CT when CR is retained on the soil surface, (ii) both nitrogen-15 ( $^{15}\text{N}$ ) dilution and  $^{15}\text{N}$  natural abundance techniques provide comparable BNF estimates because employing the  $^{15}\text{N}$  natural abundance method is much cheaper, and (iii) the fate of fertilizer  $^{15}\text{N}$  applied to succeeding winter-grown wheat including N uptake and utilization by crop, its recovery and distribution in the soil profile and the losses from the soil-plant system under CA and CT is similar.

Information on these aspects will contribute greatly to the development of optimal CA management practices according to local needs and conditions.

## MATERIALS AND METHODS

### Experimental site, climate and soil characteristics

A field study was conducted with an annual soybean-wheat rotation at Punjab Agricultural University Research Farm, Ludhiana, India (30° 54' N and 75° 48' E, 247 m a.s.l.). The long-term mean monthly minimum air temperature ranges from 4–5°C in January to 27–28°C in July, while the maximum temperature ranges from 17–20°C in January to 37–38°C in June. The long-term annual rainfall ranges from 600 to 1 200 mm with between 74 percent and 85 percent falling during the monsoon period from July to September. During the study period (2005 to 2008), the mean monthly minimum air temperature ranged from 4°C in January to 28°C in July, the maximum temperature ranged from 17°C in January to 40°C in June; the annual rainfall ranged from 563 to 995 mm of which 71–88 percent was received during June–September.

Subtropical regions have summer and winter crop growing seasons. Summers are characterized by high temperatures and rainfall (monsoons) whereas winters are often dry with low temperatures. In the present study, soybean was grown during summer (June to October) and wheat during winter (November to April). The experiments were established on a Fatehpur loamy sand soil, which was loamy sand up to 60 cm (sand, 833–882 g/kg; silt 36–69 g/kg and clay 82–102 g/kg soil), thereafter sandy loam up to 90 cm (sand, 730–888 g/kg; silt 36–114 g/kg and clay 76–100 g/kg soil), clay loam up to 120 cm (sand, 234 g/kg; silt 400 g/kg and clay 366 g/kg soil) and silty clay loam up to 150 cm (sand, 157 g/kg; silt 443 g/kg and clay 400 g/kg soil), and classified as *Typic Haplustept* (USDA, 1999). The important characteristics of the surface soil (0–15 cm) at the beginning of the study were: pH 8.1; electrical conductivity (1:2 soil:water ratio) 0.09 dS/m; organic C 3.0 g·C·kg<sup>-1</sup> soil; and Olsen-phosphorus (P) 12 kg·P·ha<sup>-1</sup> and total nitrogen (N) 665 mg/kg.

### Treatments

The study involving  $^{15}\text{N}$  labelled techniques formed part of a larger four-year field study with a soybean-wheat rotation, which had 16 treatments consisting of combinations with respect to CT and CA system, CR and fertilizer N and P in individual macro-plots of 3.15 × 8.30 m size as described in Aulakh et al. (2012). All treatments were replicated three times. Within the large macro-plots, micro-plots of 1 m<sup>2</sup> (0.8 m wide × 1.25 m long) were confined by 30-cm high rectan-

gular metal retainers pressed to a 15-cm depth in the soil, separately for soybean and wheat crops for conducting four experiments as described below.

## Experiments for measuring BNF by soybean and fate of $^{15}\text{N}$ -fertilizer in wheat

### Experiment 1

Biological N<sub>2</sub> fixation by soybean was measured with the  $^{15}\text{N}$  isotopic dilution method (Unkovich et al., 2008; ). To apply the isotope dilution technique, it is necessary to grow the “N<sub>2</sub>-fixing” crop and a suitable “non-N<sub>2</sub>-fixing” reference crop in the same  $^{15}\text{N}$ -labelled soil and the  $^{15}\text{N}$  enrichment of the reference plants is assumed to be equal to the  $^{15}\text{N}$  enrichment of the N derived from the soil in the “N<sub>2</sub>-fixing” crop. Therefore, the proportion of the unlabelled N being derived from the air via BNF in the “N-fixing” crop is proportional to the dilution of the enrichment of the N derived from the labelled soil. Boddey et al. (1990) suggested that as compared with a single reference plant, the use of several reference plants better accounts for temporal changes in  $^{15}\text{N}$  enrichment of soil mineral N and produces a range of different estimates of the BNF contribution to the “N<sub>2</sub>-fixing” crops, and that the extent of this range gives a measure of the accuracy of the estimates. Therefore, in Experiment 1, sorghum *Sorghum (Sorghum vulgare L.)* and spontaneous weeds (weed plant species that grew *in situ* in the experimental plots) were used as the multiple non-legume reference plants (Chalk, 1985; Boddey et al., 1995; Chalk and Ladha, 1999). Two weed plants “*Madhana*” (*Eleusine indica L.* Gaerth) and weed “*Badi dodhak*” (*Euphorbia hirta L.*) were used in the study.

The four treatments selected for measuring BNF consisted of tillage systems with and without CR and were replicated thrice [2 (CT and CA) × 2 (0 and 6 t-wheat residue·ha<sup>-1</sup>) × 3 (replications) = 12 plots]. In CT treatments, soil was tilled to a layer of 10–12 cm. In CR treatments, wheat residue (6 t·ha<sup>-1</sup>) was incorporated in CT and spread on the soil surface in the CA system before seeding soybean (cv SL 295) in June 2006 in rows 45 cm apart in individual micro-plots. Furrows were opened by hand and levelled after seeding soybean with each row having 12 plants. Just prior to seeding, soybean seed was inoculated with *Rhizobium* culture (*Bradyrhizobium japonicum*) obtained from the Department of Microbiology of the Punjab Agricultural University. Besides the  $^{15}\text{N}$  soybean micro-plots, additional micro-plots were established where sorghum (cv SGL 87) was seeded at the same time in three rows 30 cm apart each having 12 plants. The advantage of having the soybean plot with an adjacent non-legume reference micro-plot is that variation in soil fertility and physical conditions between them is minimized. Soybean and sorghum crops were fertilized with 10 kg·N·ha<sup>-1</sup> of  $^{15}\text{N}$ -labelled ( $^{15}\text{NH}_4$ )<sub>2</sub>SO<sub>4</sub> (10 percent atom  $^{15}\text{N}$  excess) and 26 kg·P·ha<sup>-1</sup> (equivalent to 60 kg·P<sub>2</sub>O<sub>5</sub>·ha<sup>-1</sup>) as KH<sub>2</sub>PO<sub>4</sub> in solution form. Additional water was applied immediately to wash the fertilizer N and P down in the plough layer. As and when required, irrigation of 7.5 cm water was given by taking into consideration the water received through rainfall.

Plants of soybean, sorghum and spontaneous weeds were harvested during the last week of September 2006 at the mid-pod (R2–R3) stage of soybean because at this stage the plants usually cease fixing as well as absorbing N from the soil. An area of 80 cm × 80 cm of each  $^{15}\text{N}$ -labelled micro-plot was harvested after leaving 20 cm on either side of the plot to avoid the roots picking up N from areas which had not been labelled. Weeds were separated species by species, providing thereby replicates of weed “*Madhana*” (*Eleusine*

*indica* L. Gaerth) and weed "Badi dodhak" (*Euphorbia hirta* L.) in all plots. In this way there were three independent reference plants (sorghum and two weeds).

### Experiment 2

BNF by soybean was again measured in 2008 in the same four treatments with the  $^{15}\text{N}$  isotope dilution method and following the procedure described for Experiment 1. However, the location of micro-plots within the large macro-plots was changed in order to avoid carryover effect of  $^{15}\text{N}$  applied earlier in Experiment 1. At the time of harvesting soybean, plants of sorghum and spontaneous weed "Khabal" (*Cynodon dactylon* L. Pers) were collected; the plants of two weeds collected as non- $\text{N}_2$ -fixing reference plants in Experiment 1 were not found in the micro-plots of Experiment 2.

### Experiment 3

BNF by soybean was also measured in 2008 with the  $^{15}\text{N}$  natural abundance method (Unkovich *et al.*, 2008). The samples of soybean and the reference weed "Khabal" were collected from the replicated macro-plots of four treatments as described for Experiment 1. However, the spots selected for sampling were 4 m away from the  $^{15}\text{N}$  micro-plots used in Experiments 1 and 2.

### Experiment 4

To investigate the fate of fertilizer  $^{15}\text{N}$  applied to a wheat crop seeded after harvesting soybean and to construct a  $^{15}\text{N}$  balance sheet, eight treatments were selected. These treatments consist of two tillage systems (CT and CA), two N and P rates [ $\text{N}_{120} \text{P}_{26}$  (120 kg N + 26 kg-P-ha $^{-1}$ ) and  $\text{N}_{150} \text{P}_{33}$  (150 kg-N + 33 kg-P-ha $^{-1}$ )] without or with CR (0 and 3 t-soybean residue-ha $^{-1}$ ). The recommended rates of fertilizer nutrients for optimum yields of wheat are 120 kg N and 26 kg-P-ha $^{-1}$  (Aulakh *et al.*, 2000). A rate 25 percent higher than the recommended NP rate ( $\text{N}_{150} \text{P}_{33}$ ) was included to verify whether modified tillage and residue management practices require more fertilizer nutrients for producing optimum yields. Twenty four micro-plots (eight treatments  $\times$  three replications) were established before seeding the wheat crop in November 2006. Phosphorus as  $\text{KH}_2\text{PO}_4$  corresponding to 26 and 33 kg-P-ha $^{-1}$  was applied in micro-plots according to the respective treatments before seeding wheat in three rows, 22.5 cm apart, on 6 November 2006. Ammonium sulphate (5 percent  $^{15}\text{N}$  atom excess) was applied in two equal splits, the first dose before seeding wheat and the second as a top dressing one d after first irrigation (30 d after seeding). In CR treatments, soybean residue (3 t/ha) was incorporated under CT and spread on the soil surface under CA before seeding the wheat crop.

At maturity, the wheat plants in the central rows were harvested on 17 April 2007 while leaving one row on both sides of the micro-plot and 10 cm along the front and rear side. Representative soil samples were collected from each micro-plot by combining five soil cores taken with an auger from six soil layers (0–15, 15–30, 30–60, 60–90, 90–120 and 120–150 cm), processed and ground to pass through a 2 mm sieve.

All plant samples (soybean, weeds, wheat grain and straw) collected in the four experiments were oven-dried at  $60 \pm 3^\circ\text{C}$  and ground. Soil and plant samples from all four experiments were analysed for total N and  $^{15}\text{N}$  enrichment using an elemental analyzer (NA1500, Carlo Erba Instruments) coupled to an isotope ratio mass spectrometer (Isoprime from GV Instruments, UK) at the IAEA Seibersdorf Laboratories, Austria.

## Computation and statistical analysis

The percentage of N derived from the atmosphere (%  $\text{N}_{\text{dfa}}$ ) and the amount of BNF by soybean as measured using the  $^{15}\text{N}$  isotope dilution method were calculated as follows:

$$\% \text{N}_{\text{dfa}} = 100 [1 - (\% \text{ atom } ^{15}\text{N} \text{ excess in soybean} / \% \text{ atom } ^{15}\text{N} \text{ excess in reference plant})]$$

$$\text{BNF (kg-N-ha}^{-1}\text{)} = [\% \text{N}_{\text{dfa}} \times \text{total N uptake (kg/ha)}] / 100$$

The %  $\text{N}_{\text{dfa}}$  by soybean as measured using the  $^{15}\text{N}$  natural abundance method was calculated as follows:

$$\delta^{15}\text{N (‰)} = 1000 \times (\% \text{ atom } ^{15}\text{N} \text{ of plants} - 0.3663) / (0.3663)$$

$$\% \text{N}_{\text{dfa}} = 100 [(\delta^{15}\text{N of reference weed plant} - \delta^{15}\text{N of soybean}) / (\delta^{15}\text{N of reference weed plant} - B)]$$

where B is delta  $^{15}\text{N}$  ( $\delta^{15}\text{N}$ ) of N fixed by soybean nodules and transported to shoots and assumed to be  $-1.3\text{‰}$  (Unkovich *et al.*, 2008).

Nitrogen-15 fertilizer derived by the wheat crop from fertilizer (%  $\text{N}_{\text{dff}}$ ), N derived from soil (%  $\text{N}_{\text{dfs}}$ ),  $^{15}\text{N}$  fertilizer left in the soil profile and  $^{15}\text{N}$  balance sheets were calculated as follows:

$$\% \text{N}_{\text{dff}} = 100 (\% \text{ atom } ^{15}\text{N} \text{ excess in plant sample} / \% \text{ atom } ^{15}\text{N} \text{ excess in fertilizer})$$

$$\% \text{N}_{\text{dfs}} = 100 - \% \text{N}_{\text{dff}}$$

$$\text{Total uptake of fertilizer } ^{15}\text{N (kg N ha}^{-1}\text{) in wheat straw} = \% \text{N}_{\text{dff}} \times \text{total N uptake by straw (kg N ha}^{-1}\text{)}$$

$$\text{Total uptake of fertilizer } ^{15}\text{N (kg N ha}^{-1}\text{) in wheat grain} = \% \text{N}_{\text{dff}} \times \text{total N uptake by grain (kg N ha}^{-1}\text{)}$$

$$\% ^{15}\text{N fertilizer utilization} = 100 [\text{total } ^{15}\text{N-fertilizer uptake by wheat grain and straw (kg N ha}^{-1}\text{)} / \text{rate of } ^{15}\text{N fertilizer applied (kg-N-ha}^{-1}\text{)}]$$

$$\% \text{N}_{\text{dff}} \text{ by soil} = 100 (\% \text{ atom } ^{15}\text{N} \text{ excess in a soil layer} / \% \text{ atom } ^{15}\text{N} \text{ excess in fertilizer})$$

$$\text{Total N in a soil layer (kg-N-ha}^{-1}\text{)} = \% \text{N in a soil layer} \times \text{total mass of a soil layer (kg-soil-ha}^{-1}\text{)}$$

$$\text{Total amount of } ^{15}\text{N fertilizer in a soil layer (kg-N-ha}^{-1}\text{)} = [\% \text{N}_{\text{dff}} \text{ by soil} \times \text{total N in soil layer (kg-N-ha}^{-1}\text{)}] / 100$$

where total mass of a soil layer (kg/ha) = soil depth (m)  $\times$  bulk density ( $\text{Mg/m}^3$ )  $\times 10^3$  kg/Mg  $\times$  area ( $10^4$  m $^2$ /ha); and % recovery of  $^{15}\text{N}$  fertilizer =  $100 \{[\text{total } ^{15}\text{N-fertilizer uptake by wheat grain and straw (kg/ha)} + \text{total amount of } ^{15}\text{N-fertilizer in all soil layers (kg/ha)}] / [\text{rate of } ^{15}\text{N-fertilizer applied (kg/ha)}]\}$

Statistical analysis of values for BNF by soybean,  $^{15}\text{N}$  uptake and utilization by wheat and recovery  $^{15}\text{N}$  in different layers of soil profile was carried out by ANOVA (Cochran and Cox, 1950) using a randomized block design (RBD). The effects of different treatments viz. (four for soybean experiments and eight in the wheat experiment) were evaluated using the least significant difference (LSD) test at the 0.05 level of probability. The data presented in figures are means  $\pm$  standard deviation (SD) of three replications.

## RESULTS

### Biological nitrogen fixation by soybean

In Experiment 1, %N<sub>dfa</sub> values for soybean with sorghum and two spontaneous weeds as non-legume reference plants were 51, 72 and 79 in CT without CR, 54, 68 and 77 in CA without CR, and 66, 79 and 85 in CA with CR retained on soil surface (Figure 1). In Experiment 2, with the soybean crop of 2008, % N<sub>dfa</sub> by soybean using sorghum and the spontaneous weed “Khabal” varied among different treatments from 77 to 79 in CT and from 76 to 84 in CA, respectively (Figure 2).

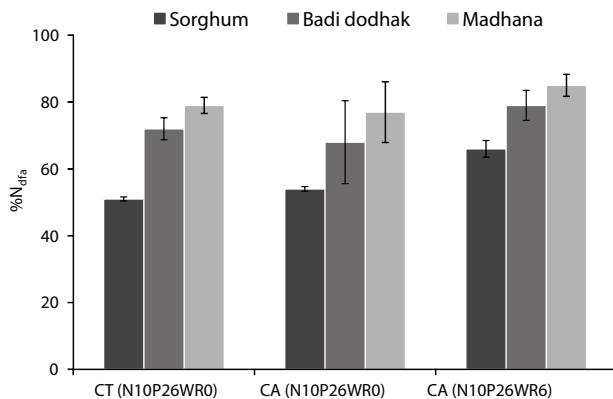
On an average of three reference plants in Experiment 1, N<sub>dfa</sub> was 67 percent in CT and 66 percent in CA without CR but increased to 77 percent in CA with CR (Figure 1). When sorghum was excluded, on an average of two reference plants the corresponding N<sub>dfa</sub> was 75 percent in CT and 73 percent in CA without CR but increased to 82 percent in CA with CR. In Experiment 2, where sorghum and only one weed “Khabal” were used as reference plants, the trends of %N<sub>dfa</sub> among the treatments were similar to those observed in Experiment 1 (Figure 2), again suggesting that as compared with CT, %N<sub>dfa</sub> was enhanced significantly in CA when CR was retained on the surface. The results of Experiment 3, where the <sup>15</sup>N natural abundance method was used, further illustrated that N<sub>dfa</sub> by soybean did

not differ in CT with or without CR (72 and 73 percent, respectively but was significantly higher (81 percent) in CA when CR was retained on the soil surface (Figure 3).

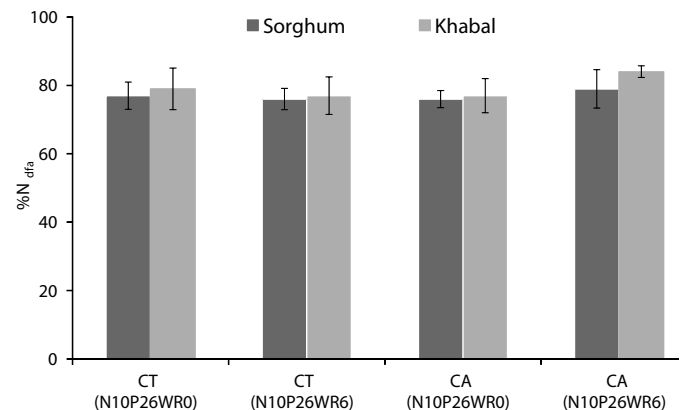
Estimates of the total amount of BNF by soybean in all three experiments showed significant differences among tillage and CR treatments (Table 1). In Experiments 1 and 2 conducted using the <sup>15</sup>N isotope dilution method, BNF ranged from 61 to 115 kg/ha in CT without CR, 64 to 115 kg/ha in CA without CR and 81 to 125 kg/ha in CA with CR. In both experiments, sorghum as reference plant showed the lowest amount of BNF, and the effects of reference plants on the amount of BNF were significant in Experiment 1. Overall results from the three experiments showed that irrigated soybean grown on soils of semi-arid subtropical region fixed N<sub>2</sub> between 81 and 125 kg/ha, which is equivalent to between 68 percent and 85 percent of total N uptake by soybean, depending upon tillage and CR management.

### Uptake and utilization of applied <sup>15</sup>N fertilizer by wheat

The amounts of <sup>15</sup>N and fertilizer N removed by wheat grain and straw based on plant <sup>15</sup>N, the proportion of <sup>15</sup>N fertilizer utilized



**FIGURE 1.** Estimates of %N<sub>dfa</sub> by soybean under conventional tillage (CT) and conservation agriculture (CA) systems using <sup>15</sup>N isotope dilution method with three reference plants in Experiment 1. Treatment details are given in Table 1. Error bars denote standard deviations (three replicates). LSD (0.05) = 9.7.



**FIGURE 2.** Estimates of %N<sub>dfa</sub> by soybean under conventional tillage (CT) and conservation agriculture (CA) systems using <sup>15</sup>N isotope dilution method with two reference plants in Experiment 2. Treatment details are given in Table 1. Error bars denote standard deviations (three replicates).

**TABLE 1.** Biological N fixation (kg/ha) by soybean in Experiments 1, 2 and 3 under conventional tillage (CT) and conservation agriculture (CA)

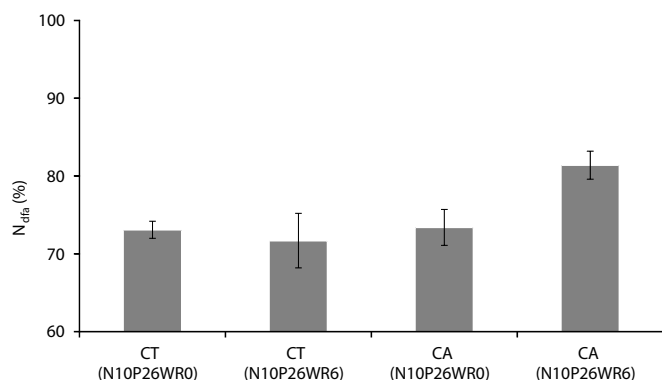
| Treatments                                                      | Reference plants in Experiment 1 |                 |         |      | Reference plants in Experiment 2 |                 |      | Experiment 3 <sup>2</sup> |
|-----------------------------------------------------------------|----------------------------------|-----------------|---------|------|----------------------------------|-----------------|------|---------------------------|
|                                                                 | Sorghum                          | Badi Dodhak     | Madhana | Mean | Sorghum                          | Khabal          | Mean |                           |
| CT N <sub>10</sub> P <sub>26</sub> WR <sub>0</sub> <sup>1</sup> | 61                               | 86              | 95      | 80   | 112                              | 115             | 114  | 107                       |
| CT N <sub>10</sub> P <sub>26</sub> WR <sub>6</sub>              | na                               | na              | na      | na   | 115                              | 115             | 115  | 109                       |
| CA N <sub>10</sub> P <sub>26</sub> WR <sub>0</sub>              | 64                               | 81              | 91      | 79   | 112                              | 115             | 114  | 109                       |
| CA N <sub>10</sub> P <sub>26</sub> WR <sub>6</sub>              | 81                               | 97              | 104     | 94   | 118                              | 125             | 122  | 120                       |
| LSD (0.05)                                                      | Treatment                        | Reference plant |         |      | Treatment                        | Reference plant |      | Treatment                 |
|                                                                 | 8                                | 8               |         |      | 5                                | ns              |      | 7                         |

<sup>1</sup> N — fertilizer N (kg N/ha); P — fertilizer P (kg P/ha); WR — Wheat crop residue (t/ha)

<sup>2</sup> Using <sup>15</sup>N natural abundance method

na — Not available

ns — non-significant

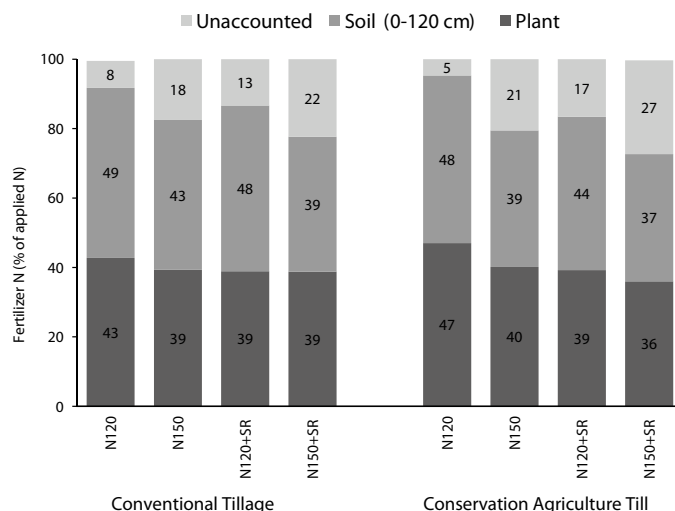


**FIGURE 3.** Estimates of %Ndfa by soybean under conventional tillage (CT) and conservation agriculture (CA) systems using <sup>15</sup>N natural abundance method with one reference plant in Experiment 3. Treatment details are given in Table 1. Error bars denote standard deviations (three replicates).

by wheat and the amounts of <sup>15</sup>N retained in different layers of soil profile up to 120 cm are presented in Table 2.

The uptake of <sup>15</sup>N fertilizer in wheat grain and straw ranged respectively from 36–44 kg·N·ha<sup>-1</sup> and 11–17 kg·N·ha<sup>-1</sup> in CT and between 33–43 kg·N·ha<sup>-1</sup> and 14–17 kg·N·ha<sup>-1</sup> in CA. Similarly, utilization of applied fertilizer <sup>15</sup>N by wheat ranged from 39 to 43 percent and from 39 to 47 percent in CT and CA, respectively.

Utilization of <sup>15</sup>N was highest with the application of 120 kg/ha without CR in CA (47 percent) followed by 120 kg/ha without CR in CT (43 percent), 120 kg/ha with CR in both CT and CA (39 percent), and was lowest with 150 kg/ha applied with CR in CA (36 percent).



**FIGURE 4.** Balance sheet of <sup>15</sup>N-labelled fertilizer N in wheat in Experiment 4. Treatment details are given in Table 2. Unaccounted <sup>15</sup>N represents losses of fertilizer N from soil–plant system.

Conversely, the utilization of <sup>15</sup>N by wheat was significantly lower when a rate 25 percent higher than recommended N rate (N<sub>150</sub>) was applied in CA with CR retained on the soil surface.

### Recovery of applied <sup>15</sup>N fertilizer in the soil profile after wheat

The recovery of <sup>15</sup>N in different layers of soil profile (Table 2) at the harvest of wheat (five and half months after fertilizer application)

**TABLE 2.** Effect of tillage, fertilizer rate and crop residue treatments on uptake and utilization of applied <sup>15</sup>N fertilizer by wheat, and recovery in different layers of soil profiles

| Treatments                                                    | <sup>15</sup> N fertilizer uptake by wheat (kg/ha) |       |       | <sup>15</sup> N utilization by wheat (%) | <sup>15</sup> N fertilizer recovered in soil profile (kg/ha) (depths in cm) . |       |       |       |        |       |
|---------------------------------------------------------------|----------------------------------------------------|-------|-------|------------------------------------------|-------------------------------------------------------------------------------|-------|-------|-------|--------|-------|
|                                                               | Grain                                              | Straw | Total |                                          | 0–15                                                                          | 15–30 | 30–60 | 60–90 | 90–120 | Total |
| Conventional tillage                                          |                                                    |       |       |                                          |                                                                               |       |       |       |        |       |
| N <sub>120</sub> P <sub>26</sub> SR <sub>0</sub> <sup>1</sup> | 39.3                                               | 12.0  | 51.3  | 42.8                                     | 35.7                                                                          | 17.2  | 2.3   | 1.8   | 2.4    | 59.4  |
| N <sub>150</sub> P <sub>33</sub> SR <sub>0</sub>              | 43.8                                               | 15.2  | 59.0  | 39.3                                     | 35.6                                                                          | 18.8  | 6.0   | 1.3   | 3.0    | 64.7  |
| N <sub>120</sub> P <sub>26</sub> SR <sub>3</sub>              | 35.9                                               | 10.8  | 46.7  | 38.9                                     | 33.3                                                                          | 16.8  | 4.4   | 1.4   | 1.4    | 57.3  |
| N <sub>150</sub> P <sub>33</sub> SR <sub>3</sub>              | 41.4                                               | 16.8  | 58.2  | 38.8                                     | 33.1                                                                          | 16.4  | 5.1   | 1.5   | 2.2    | 58.3  |
| CT Mean                                                       | 40.1                                               | 13.7  | 53.8  | 40.0                                     | 34.4                                                                          | 17.3  | 4.5   | 1.5   | 2.3    | 60.0  |
| Conservation agriculture                                      |                                                    |       |       |                                          |                                                                               |       |       |       |        |       |
| N <sub>120</sub> P <sub>26</sub> SR <sub>0</sub>              | 39.4                                               | 17.0  | 56.4  | 47.0                                     | 32.3                                                                          | 11.7  | 6.9   | 3.9   | 3.2    | 58.0  |
| N <sub>150</sub> P <sub>33</sub> SR <sub>0</sub>              | 43.4                                               | 16.8  | 60.2  | 40.1                                     | 34.0                                                                          | 14.5  | 3.6   | 3.7   | 3.2    | 59.0  |
| N <sub>120</sub> P <sub>26</sub> SR <sub>3</sub>              | 33.0                                               | 14.1  | 47.1  | 39.3                                     | 32.3                                                                          | 12.7  | 3.5   | 2.6   | 1.9    | 53.0  |
| N <sub>150</sub> P <sub>33</sub> SR <sub>3</sub>              | 38.1                                               | 15.3  | 53.4  | 35.6                                     | 29.6                                                                          | 17.8  | 4.5   | 1.2   | 1.9    | 55.0  |
| CA Mean                                                       | 38.5                                               | 15.8  | 54.3  | 40.5                                     | 32.1                                                                          | 14.2  | 4.6   | 2.9   | 2.6    | 56.3  |
| LSD (0.05)                                                    |                                                    |       |       |                                          |                                                                               |       |       |       |        |       |
| Treatment                                                     | 3.02                                               | 2.70  | 4.38  | 3.27                                     | ns                                                                            | ns    | ns    | ns    | ns     | ns    |
| Tillage                                                       | ns                                                 | ns    | ns    | ns                                       | ns                                                                            | 0.45  | ns    | ns    | ns     | ns    |

<sup>1</sup> N — fertilizer N (kg N/ha); P — fertilizer P (kg P/ha); SR — soybean crop residue (t/ha)  
ns — non-significant

revealed that between 54 and 61 percent of the residual fertilizer N was present in the 0–15 cm layer. However, movement of  $^{15}\text{N}$  below 0–15 cm soil surface layer was evident in the soil profile up to 120 cm soil depth. Of the total  $^{15}\text{N}$  left in soil profile, between 20 and 32 percent moved down to the 15–30 cm layer, and even 1–3 percent was found in the 90–120 cm soil layer.

### $^{15}\text{N}$ balance and losses in wheat

Recovery of applied  $^{15}\text{N}$  in the soil–plant system at the harvest of wheat showed that 36–47 percent was utilized by the crop, 37–49 percent was left in the soil profile and 5–27 percent was lost, which was estimated as unrecovered  $^{15}\text{N}$  (Figure 4). Losses of N were between five and eight percent when  $120 \text{ kg-N-ha}^{-1}$  was applied without CR in both CT and CR but increased to 18 and 21 percent with the application of  $150 \text{ kg-N-ha}^{-1}$ . As a result of enhanced losses of applied fertilizer N, the uptake and utilization of  $^{15}\text{N}$  were significantly reduced (Table 2).

## DISCUSSION

### Biological nitrogen fixation by soybean

While the  $\%N_{\text{dfa}}$  values were quite comparable among all three experiments (Figures 1, 2 and 3), the amounts of BNF were substantially less in Experiment 1 than in Experiments 2 and 3. This was essentially due to the lower yields and total N uptake by soybean obtained in Experiment 1 (data not shown). The results of both Experiments 1 and 2 revealed lowest  $N_{\text{dfa}}$  by soybean when sorghum was used as the reference plant.

Earlier studies, where multiple reference plants were used, showed that the lowest  $\%N_{\text{dfa}}$  by soybean was with sorghum as the reference plant. Boddey and Urquiaga (1990) reported that non-nodulating soybean, sorghum and sunflower had  $^{15}\text{N}$  enrichments of 0.4314, 0.4272 and 0.6217 percent atom respectively, while nodulated soybean had 0.3616 percent atom. Boddey *et al.* (1995) showed that estimates of  $N_{\text{dfa}}$  by soybean cv 29W using four non- $\text{N}_2$  fixing reference plants varied significantly between 14 percent (okra), 16 percent (sorghum), 47 percent (non-nodulated soybean) and 50 percent (rice). The results of this study confirmed earlier observations that sorghum, having deeper roots with different root architecture than soybean, absorbs mineral N from much deeper soil depths and therefore, most likely utilizes mineral N with a different  $^{15}\text{N}$  enrichment than the soybean roots (Boddey *et al.*, 1995). Thus, the spontaneous weeds chosen as reference plants in the present study which were in closer proximity to soybean than sorghum, had a similar root depth and grew to a similar size as soybean, thereby facilitating similar exploration of soil mineral N and presumably provided a reliable index of  $\%N_{\text{dfa}}$  by soybean.

These results support the hypothesis that, on average from different reference plants, BNF was significantly higher in CA with CR retained on the soil surface. Using automated temperature monitoring probes, Aulakh *et al.* (2012) measured soil temperature at 4 and 10 cm soil depth in macro-plots of the same experiment and demonstrated that during daytime, the maximum soil temperature at 4 cm depth remained 4–8°C lower in plots with retention of CR on the soil surface in CA as compared with bare soil surface. Thus, the beneficial effect on BNF by soybean of CR retention on the soil surface was presumably due to better activity of rhizobia created by the relatively cooler environment in the rhizosphere.

Comparable results were obtained in Experiment 2 where the  $^{15}\text{N}$  isotope dilution method was used and in Experiment 3 where

the  $^{15}\text{N}$  natural abundance method was used, with spontaneous weeds as reference plants. While the former method requires costly  $^{15}\text{N}$ -enriched fertilizer, the latter is based on natural abundance of  $^{15}\text{N}$  and hence is much cheaper to use.

### FATE OF $^{15}\text{N}$ FERTILIZER APPLIED TO WHEAT

The uptake of  $^{15}\text{N}$  fertilizer in wheat grain and straw indicated quite similar patterns in both CT and CA tillage systems for N removal from soil as well as translocation from vegetative parts to grain. Similarly, the range of  $^{15}\text{N}$  utilization by wheat in this study (39–47 percent) was similar to that reported from earlier studies with CT at the same site (Katyal *et al.*, 1987; Bijay-Singh *et al.*, 2001). The reduction in the utilization of  $^{15}\text{N}$  where crop residue was incorporated in CT or retained on soil surface in CA could possibly be due to higher losses of N via ammonia ( $\text{NH}_3$ ) volatilization and/or denitrification as discussed later. The reduced utilization of  $^{15}\text{N}$  at  $150 \text{ kg/ha}$  suggests that fertilizer was applied in excess of crop needs.

The significantly lower uptake of fertilizer N by wheat (straw + grain) in CA with CR retained is in line with the results obtained by Aulakh *et al.* (2012) from the macro-plots of the same study. At these macro-plots, reduced wheat grain yield and total N uptake were observed due to the cooler soil surface and related delayed germination with CR retained on soil surface in CA as discussed above.

The recovery of applied  $^{15}\text{N}$  fertilizer in different layers of the soil profile (Table 2) at the harvest of wheat revealed that the majority of the residual fertilizer N was present in the top soil layer with little evidence for effects of fertilizer N rate, tillage and crop residue management. Movement of  $^{15}\text{N}$  downwards was not affected significantly by fertilizer rate, tillage and CR treatments. In an earlier study with a rice–wheat rotation under CT system on the nearby site, Aulakh *et al.* (2000) reported rapid distribution of fertilizer N to a soil depth of 60 cm within 1–5 d after its application. Also, they observed that significant amounts of nitrate ( $\text{NO}_3^-$ )-N from soil layers up to 60-cm depth were used by wheat following rice because of its deeper and extensive rooting system.

Losses of N were low when  $120 \text{ kg-N-ha}^{-1}$  was applied without CR in both CT and CA but increased with the application of  $150 \text{ kg-N-ha}^{-1}$ , again suggesting that excessive N application led to higher N losses as a consequence of reduced utilization of  $^{15}\text{N}$  by the wheat crop (Table 2). The losses of N could be via  $\text{NH}_3$  volatilization and/or denitrification as the possibility of fertilizer N losses due to leaching to deeper soil layers appears to be low (only 2–6 percent of  $^{15}\text{N}$  in fertilizer was recovered in the 60–120 cm soil layer). Since soil pH was 8.1, there is a possibility of  $\text{NH}_3$  volatilization when  $\text{NH}_4\text{-N}$  fertilizer is applied on the soil surface (Aulakh and Bijay-Singh, 1997). The opportunity for this N loss process to occur was greater from soil under CA than that under CT. The increased  $^{15}\text{N}$  losses in CA with the retention of CR on the soil surface in conjunction with  $120 \text{ kg-N-ha}^{-1}$  and  $150 \text{ kg-N-ha}^{-1}$  were also presumably in part through denitrification supported by soluble carbon (C) that moves downward from surface residue. On the other hand, the increased  $^{15}\text{N}$  losses in CT with the incorporation of CR in conjunction with  $120 \text{ kg-N-ha}^{-1}$  and  $150 \text{ kg-N-ha}^{-1}$  were mainly through denitrification as decomposition of CR creates congenial conditions by consuming oxygen from the soil and supplying organic C substrate to denitrifying organisms (Aulakh *et al.*, 1991, 2001).

## CONCLUSIONS

Results of this study support several conclusions that may have important agronomic and environmental implications. Soybean fixed N ranging from 81 to  $125 \text{ kg-N-ha}^{-1}$ , equivalent to 68–85 percent

of total crop N uptake, depending upon tillage and crop management. Furthermore, significant increases in BNF by soybean (between 5 and 20 percent) were recorded when crop residue was retained on the soil surface of CA and indicate the benefits that could be accrued by adopting such systems. Thus, partial substitution of rice (which requires 120–150 kg/ha of fertilizer N as well as 3–4 times the amount of irrigation water) by soybean, especially in rotation with wheat under irrigated conditions, could lead to an environmentally and economically sustainable cropping system. Although soybean grains remove a major part of N derived from BNF, earlier studies have well documented that soybean residues have substantial N content and enrich the soil upon mineralization leading to sustainable systems (Aulakh *et al.*, 1991, 2012).

The recommendation for the adoption of CA in soybean–wheat rotation in the Indo-Gangetic Plains of northwestern India is further strengthened by quite similar patterns of N uptake, translocation from vegetative parts to grain, and utilization of applied <sup>15</sup>N by wheat in both tillage systems. The amount of unaccounted <sup>15</sup>N, which may have arisen from losses of fertilizer N from the soil–plant system, was higher with 150 kg-N-ha<sup>-1</sup> in both CT and CA systems, suggesting the need for future research to improve N use efficiency in winter-grown wheat. The use of isotopic techniques in the present investigations facilitated the identification of factors and practices that can form the basis of future field studies for the development of effective strategies for the management of tillage and CR for fostering sustainable and environmentally sound agricultural systems. The good performance of the cheap natural abundance-based <sup>15</sup>N techniques also offers great opportunities for further multiple site and on-farm research.

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# Soil Organic Carbon Preservation and Sequestration in European Agricultural Soils: An Overview

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## ABSTRACT

The decrease of fertility in European soils is fully recognized both from the scientific arena and from institutional bodies. Soil organic matter (SOM) decline is on the agenda of the Thematic Strategy for Soil Protection of the European Commission (EC) and the implementation of pilot long-term experiments (LTE) to detect soil organic carbon (SOC) changes is considerably increasing. Several bodies have started national inventories of forested areas, the Reducing Emissions from Deforestation in Developing Countries (REDD) mechanism is non-stop aiming to reduce emissions from forests, and the role of peatlands in climate change has been widely debated. Also, the integration of agricultural soils, climate change influence and food security in Europe is a topic under investigation. Fighting hunger in a sustainable way is on the agenda of the European Union food security policy and the Joint Research Centre's (JRC) role is to build a cross-disciplinary approach between the science and policy to take actions. Studies on SOC changes and the best management for C sequestration are continuously appearing, but how to deal with a growing population and the related food demand in the context of a climate change scenario is unknown. This paper gives an overview of the activities carried out in the JRC-SOIL Action with a specific focus on agricultural soils and their SOC content. A short paragraph covers the potential application of nuclear and isotopic techniques to support the knowledge of SOM and C sequestration dynamics.

**Key words:** *organic carbon, agriculture, soil, Europe, isotopic and nuclear techniques.*

## SOIL PROTECTION IN EUROPE (COM (2006) 231 FINAL)

### The thematic strategy for soil protection

The decline of soil organic matter (SOM) is recognized as one of the eight soil threats (together with erosion, contamination, sealing, compaction, loss of biodiversity, salinization, impermeabilization, and

landslides) expressed in the European Commission's Thematic Strategy for Soil Protection with the objective of ensuring that the soils of Europe remain healthy and capable of supporting human activities and ecosystems (EC, 2006). One of the key goals of the strategy is to maintain and improve SOM levels to assure fertility. Climate change is recognized as a common element in many of the soil threats and for this reason the European Commission (EC) intends to assess the actual contribution of soil protection to climate change mitigation and the effects of climate change on SOM content. The overall objective of the strategy is to establish a Soil Framework Directive for the protection and sustainable use of soil, by preventing further soil degradation, preserving biophysical functions and by restoring degraded soils taking into account land use. In 2012, a report (COM (2012) 46 final) was published on the implementation of the Soil Thematic Strategy and ongoing activities by the EC's Directorate General for the Environment (DG ENV). In the document the use of appropriate management practices is mentioned as a way to maintain and increase SOM content (EC, 2012a).

### Land use, land use change and forestry (LULUCF)

LULUCF refers to the forestry and agricultural sector in the context of the international climate negotiations under the United Nations Framework Convention on Climate Change (UNFCCC). Land use, land use change and forestry covers greenhouse gas (GHG) emissions and removals related to soils, trees, plants, biomass and timber. Forests and agricultural lands currently cover more than three-quarters of the European Union (EU) territory and hold large stocks of carbon (C). The accounting of C stored in the forests and soils is only partly recognized in the total GHG accounting due to the lack of robust C databases and common rules on how to account for emissions and removals. The EU intended to close the gap in the GHG accounting in its climate policy, submitting a proposal to harmonize rules to account for forests and agricultural soil emissions across the EU (the first step to incorporate these major sectors without common EU-wide rules into the EU's reduction efforts). The proposal (COM (2012) 93 final) aims to make accounting for grassland management and cropland management mandatory for Member States (MS) (EC, 2012b). Appropriate land use and management practices in forestry and agriculture could limit emissions of C and enhance removals from the atmosphere. The accounting framework could lead to positive mitigation efforts in the sectors becoming more visible and at the same time provide a solid basis for further cost-effective mitigation options and sustainable growth.

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## Agriculture, soils and climate change: the CAP towards 2020

A substantial proportion of land in Europe is occupied by agriculture, and consequently this sector plays a crucial role in natural resources protection, a precondition for other human activities in rural areas. Adopting inefficient and non-sustainable land uses, management and farming practices, including poorly managed intensification as well as land abandonment, have adverse impacts on natural resources (EC, 2009a). According to the available literature (monitoring programmes, long-term experiments (LTE) and modelling studies), C preservation and sequestration in the EU's agricultural soils could have some potential to mitigate the effects of climate change. Of this potential, the highest mitigation share is related to C preservation linked to preventing certain land use changes (conversion of grassland or native ecosystems to cropland) and maintaining C stocks in organic soils. The most effective strategy to prevent global SOC losses would be to halt land conversion to cropland, but this may conflict with growing global food demand unless per-area productivity of the cropland continues to grow. The adoption of best management practices within all different land use categories could solve this conflict. (Kimble, Lal, and Follett, 2002) Implementing these technical mitigation strategies at the EU level would require changes to current agricultural practices. For example, changes in the cropping systems (already to some extent part of cross compliance minimum requirements), conversion of land use or the adoption of practices that leads to yield decreases. Other measures may be the production of neutral or imply long-term gains in terms of soil fertility and additional benefits (Schils *et al.*, 2008). Since 2009, with the White Paper, the EC and specifically the Directorate General for Agriculture and Rural Development started to take into account the role of European agricultural soils in climate change at the policy level. One of the two working documents of the White Paper, (SEC (2009) 417), explored the effect of projected climatic changes on crop yields, livestock management and the location of production, with a focus also on depletion in SOM and soil C preservation capacity (EC, 2009b). The other document, (SEC (2009) 1093 final), matched the reduction of GHG emissions coping with the changing climate to ensure synergies between adaptation and mitigation for co-benefits (soil and tillage practices, protection and management of pastures, organic farming) (EC, 2009c). The EC is actually preparing the groundwork for the new Common Agricultural Policy (CAP) post-2013. The new policy aims to assure (1) a viable food production, (2) a sustainable management of natural resources, and (3) climate action and a balanced territorial development. The communication on the new CAP, (COM (2010) 672 final), describes how farming practices could limit soil depletion, water shortages, pollution, C sequestration and loss of biodiversity (EC, 2010).

### Soil action at the Joint Research Centre

The Joint Research Centre (JRC) is the EC's in-house science service aiming to serve society, stimulate innovation and support legislation. The JRC has the mission to provide scientific advice and technical know-how to support the conception, development, implementation and monitoring of EU policies. One of the seven institutes of the JRC is the Institute of Environment and Sustainability (IES) which deals with EU policies for the protection of the environment and the sustainable management of natural resources. The SOIL Action, which belongs to the Land Resource Management Unit of IES, aims to develop policies relevant to soil data and information systems. This activity has been running for more than 20 years and has developed

the main reference data on the European soils at various levels of detail including:

- Establishment of the European Soil Data Centre (ESDAC) as a single point for all soil data;
- Development of procedures and methods for data collection, quality assessment and control, management and storage, distribution to the EC and external users, fully complying with INSPIRE (Infrastructure for Spatial Information in Europe) Directive principles;
- Research and development of advanced modelling techniques, indicators and scenario analyses in relation to the main threats to soil;
- Scientific support and technical assistance to other Commission services, European bodies and MS regarding soil information and negotiations;
- Extension of the coverage of the European Soil Information System (EUSIS) towards a fully operational Global Soil Information System (GLOSIS).

Focusing on SOC, the SOIL Action is involved in several issues and more specifically the assessment of SOC content and stocks; the collection and harmonization of the EU MS data; monitoring of the SOC decline and sequestration potential; the establishment of a proposal for a common sampling methodology aiming at C estimates; the refinement of pedotransfer rules (PTRs) and functions (PTFs); the mapping land cover/land use changes; the assessment of the effect of agricultural management practices on SOC; and a research study on the potentiality of biochar application, etc. The final aim of the SOIL Action will be to develop tools that will allow for the rapid, cost-effective and precise measurement of SOC content on a regular basis in Europe allowing for easy verification and reporting of SOC changes. Of particular importance is also the development of new advanced digital soil mapping techniques based on geo-statistical analysis and new measurement techniques.

### European Soil Data Centre (ESDAC)

ESDAC is the thematic platform for soil related data in Europe. It was established through a decision taken at the end of 2005 by the EC's DG ENV, DG JRC, DG Eurostat (ESTAT) and the European Environment Agency (EEA) to establish ten environmental data centres in Europe. As a new development within the Seventh Framework Programme (FP7) for research of the EU (2007–2013), the SOIL Action has established ESDAC as an INSPIRE system on soil data and information operated by a network of national and regional data centres. The primary responsibility of ESDAC, as the single focal point, is to organize the availability and quality of the soil data required for policy making. Data come from projects run inside the EU institutions but also from collaborative projects between the JRC and several research partners (FAO, ISRIC world soil information data centre and the European Soil Bureau Network, etc.). The European Soil Data Centre can support EU policies related to soil as many key data sources are held by it and readily accessible to policy makers, the scientific community and to the public at large (Panagos *et al.*, 2012).

### European Environment Information and Observation Network (EIONET)

At the European level, there is a serious lack of reliable geo-referenced, measured and harmonized data on soil properties and specifically on SOC from systematic sampling programmes. At present, the most homogeneous and complete data on the SOM and SOC contents of European soils remain those that can be extracted and/or

derived from the European Soil Database (ESDB), in combination with associated databases on land cover, climate and topography. The latter database is the only comprehensive source of data on the soils of Europe harmonized according to the FAO standard international classification. In the context of the development of ESDAC, the European Environment Information and Observation NETwork (EIONET) has been established with the aim of providing timely and quality-assured data, information and expertise to assess the state of the environment in Europe, and the pressures acting upon it. The European Environment Information and Observation NETwork consists of representative organizations from 38 European countries (including the 27 MS of the EU plus other European countries). The network is built on national focal points that co-ordinate primary contact points and national reference centres for specific areas such as soil, water, waste, etc. The Directorate General for the Environment and EEA have identified the decline in SOM and soil losses by erosion as priorities in relation to the collection of the EU policy relevant soil data. The need for data is urgent and several efforts to establish agreed datasets for the EU countries have been made in the past. To support modelling activities and to display variations for these soil degradation indicators, EEA and the JRC jointly decided that all soil data management activities carried out by EEA in collaboration with EIONET would be transferred to the JRC. The latter, through EIONET, performed a data collection exercise in 2010 among MS with the final objective of creating a European wide dataset for SOC and soil erosion according to a grid based approach. There was no legal obligation for the EIONET MS to participate and PCPs and NRCs for soil contributed on a voluntary basis. The technical specifications of the data requested followed the standard 1 km grid defined by the INSPIRE Directive and the data submitted by participating countries could be based on measurements or be the best "estimate". Coming from different data sources, the data collected were harmonized to account for different methods of soil analyses, scales and time periods. As shown in

Figure 1, for the SOC content (in percent), data were received from 12 EIONET Members (32 percent of the total EIONET network), but only five of them provided data for more than 50 percent of their geographical coverage (Panagos *et al.*, 2013).

### Topsoil organic carbon content map (OCTOP)

The geographical distribution of the SOC content has been mapped for the EU and the JRC is undertaking SOC sequestration potential investigations (Jones *et al.*, 2005). The European soil portal provides the map of topsoil organic carbon content (OCTOP) of soils in Europe, shown in Figure 2. The estimation of SOC and the management of SOC sequestration from soils and more specifically from agricultural soils have become a priority issue and needs to be addressed not just at the scientific level, but also within the policy arena. Already several projects supported by the EC took into consideration the use of agricultural conservation practices in relation to the main soil protection objectives, and provide a stocktaking of the current situation with regards to the policy measures that address (or contribute to) soil conservation within a EU-wide perspective (EC 2009a).

The JRC has a research activity on the implementation of the CAP policy options with the land-use modelling platform. The study estimated changes in SOC stocks from changes in land use as an indicator to evaluate the impact of various options of agricultural policies in support to DG AGRI and DG ENV. The changes in land use were defined based on scenarios of land use changes between the base year (2010) until a projected year (2020). The changes in SOC stocks are estimated following the Tier 1 approach of the IPCC (Lavalle *et al.*, 2011).

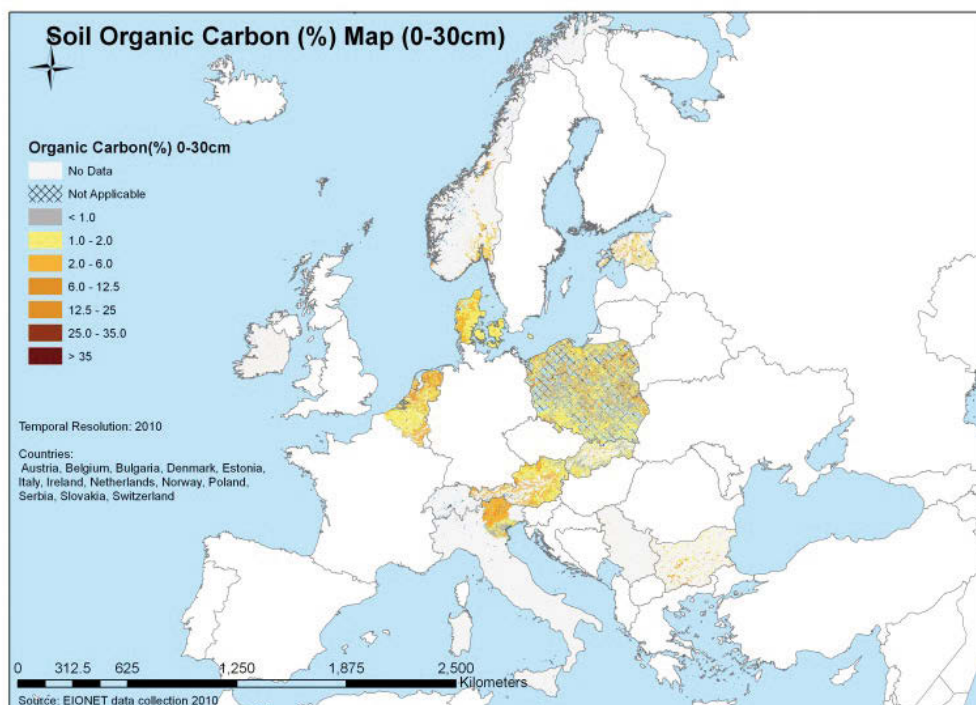
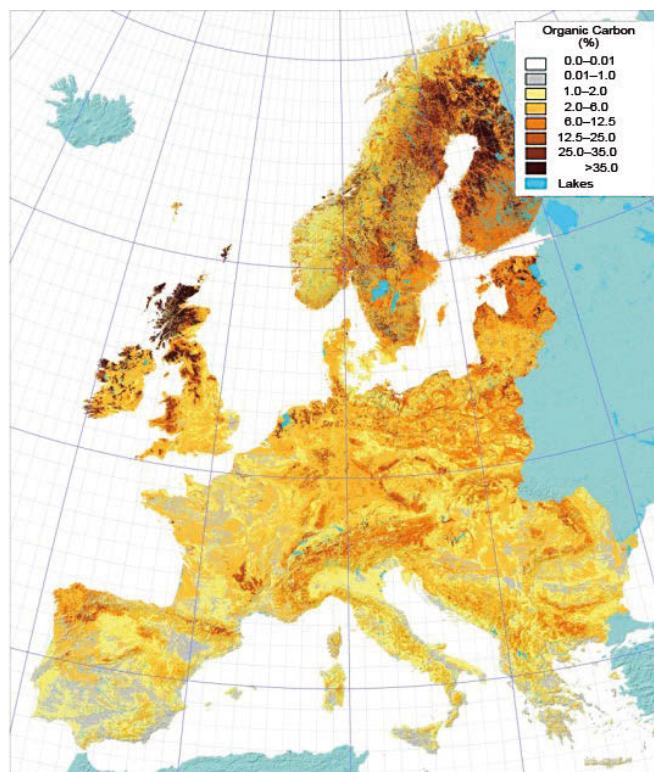


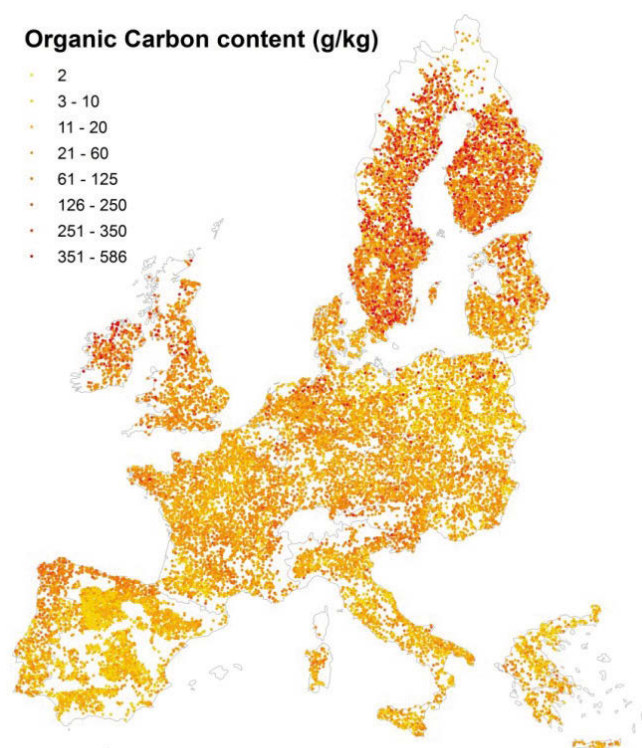
FIGURE 1. Results obtained from the 2010 SOC data collection exercise (Panagos *et al.*, 2013).



**FIGURE 2.** OC content (in percent) in the surface horizon (0-30 cm) of soils (Jones et al., 2005).

### Land use/cover statistical area frame survey (LUCAS) project

Land represents a key actor for policy makers and European ecosystem planning, and for this reason there is a need for data: on changes in land cover, in biophysical attributes of the earth's surface and in land use affected by human actions. The capability to monitor those changes is linked to the availability of information on the coverage and use of the land. The LUCAS project is a field survey carried out on a sample of about 235 000 geo-referenced points spread over Europe. As such it is essentially a monitoring tool to follow the status of landscape diversity, to provide harmonized information on land cover and land use and to monitor changes in management and coverage in the European territory. The selection of the points was based on a regular  $2 \times 2$  km grid covering the EU territory and defined as the intersection of around 1 million geo-referenced points. Each point was classified into seven land-cover classes using orthophotos or satellite images. From the stratified master sample, a sub-sample of 235 000 points was extracted to be classified by field observations in 2006 according to the full land nomenclature. The pilot phase (2000–2007) involved 13 to 15 MS, with the first survey being held in 2001 to test the methodology at the EU level with a restricted budget. The focus of the survey was agricultural land with a sampling rate of 50 percent for arable land and permanent crops, 40 percent for grassland and 10 percent for non-agricultural land. The latest survey took place in 2009 in 23 EU MS. During the campaign, the field surveyors collected data on land cover/land use plus landscape photographs. In addition, linear elements and land-cover changes along a 250 m walk eastwards from each point (transect) were recorded.



**FIGURE 3.** Distribution of 19 515 points and their level of topsoil OC concentration (g/kg) (JRC, 2011).

### LUCAS soil 2009

In 2009, within the LUCAS general project, an exercise coordinated directly by the JRC–SOIL Action called LUCAS Soil 2009 was performed to support the thematic strategy for soil protection. Its main aim was to produce the first coherent soil sampling exercise and harmonized set of analysis in the EU. This exercise is organized jointly by Eurostat, the DG ENV and the JRC and includes a topsoil module to the survey involving a subset of around 20 000 sites that were sampled in 25 MS (EU-27 except Romania and Bulgaria). The soil samples were collected by the LUCAS surveyors at points of their visit and dispatched to a central laboratory for the analysis of physical, chemical and multispectral properties (particle size distribution, pH, OC, carbonates, nitrogen, phosphorus and potassium [NPK], cation exchange capacity (CEC) and visible and near infrared diffuse reflectance). The same soil sampling exercise was carried out in Bulgaria and Romania in the early summer of 2012. The samples, weighing around 11 t in total, are now stored in the European Soil Archive Facility, at the JRC -Ispra. The points were extracted from the general LUCAS for topsoil sampling as being representative for soils of Europe and its 27 MS, stratified according to the topography and land use, and giving priority to points located on agricultural land. The LUCAS Soil 2009 dataset represents the latest and most comprehensive and harmonized information on physico-chemical properties of topsoil from the EU-27. The data on land use/land cover from the general LUCAS and the parameters collected from the soil sampling exercise are under evaluation, showing links between land use/cover classes, land management and the physico-chemical properties. Figure 3 shows the distribution of LUCAS points for SOC content. The LUCAS project provides new data for the estimation of topsoil C content at European level with a special focus on agricultural land.

A quality assessment of data was performed on the dataset, taking the main climatic zones, regions, land cover classes and management practices into consideration. Results highlight important linkages among these factors and help to understand and quantify the potential of European croplands with regards to C content and other major soil functions and indicators of soil health. Woodland and shrubland show the highest level of SOC in all climatic regions; this trend is in line with the common understanding of high values of SOC in forest compared to other land-cover classes. The lowest levels of SOC are observed in the Mediterranean climatic region; this general trend confirms that SOC content in northern regions is higher than in southern parts of the continent. Levels of SOC in arable land and the boreal climatic region are at least three times higher than in the other climatic regions. The map of OC content in topsoils in Europe from the LUCAS soil 2009 survey is the first step towards the interpolation of the LUCAS SOC points for creating a map using geo-statistical methods with a specific focus on agricultural topsoils (Jones *et al.*, 2005). Another evaluation in the LUCAS dataset is the C and nitrogen (N) limitation in soils. Preliminary results show that forested soils tend to be N limited and plant available C is rather a more limiting factor in the other soils than N. No specific trend is observed between climatic regions. The SOIL Action is performing other soil survey campaigns aimed at the collection of updated SOC data for Europe. For forest soils these data are already completed and are part of the data collected within the BioSoil demonstration project carried out in the course of the Forest Focus1 Regulation.

### LUCAS spectral library for SOC prediction

In the context of global environmental change, the estimation of C fluxes between soils and the atmosphere has been the object of a growing number of studies. This has been motivated notably by the possibility to sequester carbon dioxide (CO<sub>2</sub>) into soils by increasing SOC stocks and by the role of SOC in maintaining soil quality (Lal, 2004). The spatial variability of SOC masks its slow accumulation or depletion, and the sampling density required to detect a change in SOC content is often very high and thus labour intensive and costly (Shepherd and Walsh, 2002). Visible near infrared diffuse reflectance spectroscopy (Vis-NIR DRS) is a fast, cheap and efficient tool for the prediction of SOC at fine scales (Stevens *et al.*, 2008; Morgan *et al.*, 2009). However, when applied to regional or country scales, Vis-NIR DRS does not provide sufficient accuracy as an alternative to standard laboratory soil analyses for SOC monitoring (Brown *et al.*, 2006). Within the framework of the LUCAS project, soil samples were scanned with a Vis-NIR spectrometer in the same laboratory. The scope of this research was to predict SOC content at the European scale using the LUCAS spectral library. A modified local partial least square regression (I-PLS) was implemented including, in addition to spectral distance, other potentially useful covariates (geography, texture, etc.) to select a group of predicting neighbours for each unknown sample. The dataset was split into mineral soils under cropland, under grassland, under woodland and organic soils due to the extremely diverse spectral response of the four classes. Four of every class training (70 percent) and test (30 percent) sets were created to calibrate and validate the SOC prediction models. The results showed very good prediction ability for mineral soils under cropland and under grassland, with a root mean square errors (RMSEs) of 3.6 and 7.2 g-C/kg respectively, while mineral soils under woodland and organic soil predictions were less accurate (RMSEs of respectively 11.9 and 51.1 g-C/kg). The RMSE was lower (except for organic soils) when sand content was used as a covariate in the selection of the I-PLS predicting neighbours. Despite the enormous spatial variability of European soils, the modified I-PLS algorithm developed

was able to produce stable calibrations and accurate predictions. It is essential to invest in spectral libraries built according to sampling strategies based on soil types, and a standardized laboratory protocol. Vis-NIR DRS spectroscopy is a powerful and cost effective tool to predict SOC content at regional/continental scales, and should be converted from a pure research tool into a reference operational method to decrease the uncertainties of SOC monitoring and terrestrial ecosystem C fluxes at all scales.

### LUCAS in the future

The land use/cover statistical area frame survey design has been initiated in Iceland (an EU candidate country), with soil samples planned to be collected in 2012 and 2013. In subsequent years, a large contribution is expected from other candidate countries. Preliminary studies on the database show the applicability of the LUCAS exercise for monitoring and accounting of the soil properties at the EU level. In addition, the data covering chemical and physical properties can be correlated with the general LUCAS data on land use/land cover and the impact of land-use/cover changes over time monitored. For this reason the LUCAS topsoil survey exercise represents a potential EU soil monitoring reporting verification system. If this exercise could be carried on over time, it would be possible to detect changes, for example, in SOC stocks and subsequently to evaluate the potential of EU agricultural soils for C preservation and sequestration.

### CAPRESE project

There is a lack of knowledge as to how SOC-based mitigation options in agriculture are distributed across different agricultural production systems, and how the implementation of such mitigation options affects present land use practices (Jones *et al.*, 2005). SOIL Action is underway to focus on a new assessment of the current state of SOC levels in the agricultural soils of the EU. A review of current literature on the science of SOC fluxes in relation to current land covers/uses and policies is on-going, with particular attention being given to land changes and land conversion status. The literature review is also addressing and evaluating a range of agricultural management mitigation options and their theoretical impacts in stabilizing or enhancing SOC stocks (e.g. land-use change, tillage methods, cropping systems, irrigation methods, nutrient management, cross-compliance, rural development measures, etc.) in interaction with soil characteristics and climatic conditions. The final results will be presented in a report that will also evaluate the possible constraints underlying the full application of mitigation options. In the context of this review, a new project for DG AGRI started in 2012. The CARbon PREservation and SEquestration in agricultural soils — options and implications for agricultural production (CAPRESE) project aims to provide the necessary background that will contribute to the mid-to long-term development of policies addressing climate change soil-related aspects in European agriculture. Particular objectives of the study are: review of the potential climate change mitigation actions for agricultural soils across the EU; potential impact of selected mitigation measures in relation to OC levels; the effect of selected soil management measures on production patterns for different agricultural products; associated costs and support mechanisms to monitor and quantify the effects of the measures on the greenhouse gas balance; support for future evaluation of CAP measures; mitigation target scenarios for agricultural soils; and development of recommendations.

## Soil transformation into European catchments (SoilTrec) projects

SoilTrec is one of the FP7 projects in SOIL Action. The “Spatial prediction of soil organic carbon of Crete & Koiliaris CZO by using geostatistics” study is one of the SoilTrec projects, the purposes of which are: predicting SOC distribution of Koiliaris CZO and Crete by using geostatistics; developing up-scaling rules for Koiliaris CZO and Crete; developing a current OC map; and detecting C changes in space (land-use types) and in time. Soil organic carbon distribution on Koiliaris CZO and Crete were predicted using soil samples and environmental predictors (slope, aspect, elevation, CORINE land-cover classification, geological formations, World Reference Base for Soil Resources (WRB) soil classification, texture, average temperature and precipitation) with the Regression-Kriging (RK) method. These studies showed that the RK method is useful and reliable for predicting SOC. A significant correlation was found between the covariates and the OC dependent variable. The combination of a local dataset and LUCAS samples was advantageous for calibrating the land-use based soil data. The incorporation of associated local soil data improved the SOC estimates of the Crete map. The current SOC map of Crete was developed using the RK method. Currently, the project is focusing on a new CZO for the Damma Glacier in Switzerland for detecting SOC with the same method (Aksoy *et al.*, 2011; Aksoy, Panagos and Montanarella, 2012).

## APPLICATIONS OF NUCLEAR AND ISOTOPIC TECHNIQUES TO SUPPORT THE KNOWLEDGE OF SOIL ORGANIC MATTER AND CARBON SEQUESTRATION DYNAMICS

Isotopic techniques are high precision tools that can provide an insight into soil processes and help to better understand, define and quantify the decline in OM and the C sequestration processes occurring in soils. The delta carbon-13 ( $\delta^{13}\text{C}$ ) and delta nitrogen-15 ( $\delta^{15}\text{N}$ ) isotopic signatures of SOM fractions can be used to identify C stabilization mechanisms and determine how long C molecules persist in soil (Kayler *et al.*, 2011). Methods using the stable isotope  $^{13}\text{C}$  or the fall-out radionuclide carbon-14 ( $^{14}\text{C}$ ) are being used increasingly to study SOM turnover (Six and Jastrow, 2002). These methods are very useful in this field of research, as isotopes are the only tools which yield information over relatively short time periods (months and years), which is particularly relevant to study the impact of land-use management on SOM. The differences in the  $\delta^{13}\text{C}$  isotopic signatures between C3 (e.g. wheat) and C4 plants (e.g. corn) can be used as fingerprints with simple mixing models to estimate the respective contribution to SOM and the residence time in soil of each C organic input. For a specific ecosystem, the natural  $\delta^{13}\text{C}$  isotopic signature of the  $\text{CO}_2$  released from the soil provides information about the processes driving  $\text{CO}_2$  exchanges and the sources of the emissions (Pataki *et al.*, 2003).

## EUROPEAN SOIL PORTAL AND NETWORKS

### European soil portal

The SOIL Action manages the European Soil Portal (<http://eusoiis.jrc.ec.europa.eu>) which contributes to a thematic data infrastructure for soils in Europe and acts as a web platform for ESDAC. It presents data and information on the European soils and tries to provide links to national or global datasets. In the portal it is possible to find: an inventory and access to the soil data currently held by the JRC, a library of scanned maps and a collection of prepared maps derived

from the existing soil databases and user's applications to interact on-line with the data and all the SOIL Action activities.

### ESBN network

The European soil portal website serves as a vehicle to promote the activities of the European Soil Bureau Network (ESBN). It was created in 1996 as a network of national soil science institutions and is operated at the JRC by the SOIL Action. Its main tasks are to collect, harmonize, organize and distribute soil information for Europe.

The SOIL Action is involved in many other worldwide networks and collaborations such as the Global Soil Partnership, Harmonized World Soil Database, Intergovernmental Panel on Climate Change, Global Soil Map.net, Global Soil Biodiversity Initiative, Sino-EU Panel on Land and Soil, etc.

## CONCLUSIONS

The SOIL Action team and activities are very dynamic and influenced on one side by scientific passion and on the other side by the policy requests of the EC. The agenda of C sequestration has become a priority and the way to co-benefit soil protection and production with a cost-competitive mitigation potential is a new area of investigation.

## ACKNOWLEDGEMENTS

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# Global Monitoring of Soil Resources for Agriculture: Feasible Options for Early Implementation

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## ABSTRACT

Information on the status and trends in the condition of global soil resources for agriculture is urgently needed to drive policy for their protection and to support future food security. Current information on the extent and condition of global soil resources is inadequate and poorly collated. This paper reviews technical options and the feasibility of implementing a global soil monitoring system. Remote sensing methods are available for measuring indicators of change in the extent of soil resources (e.g. land area under arable production; area sealed by urban and infrastructure development). A soil monitoring network (SMN) based on physical sampling of strata corresponding to soil types and bio-geographical regions appears best suited to global ground-based soil monitoring. Reliable, agreed methods are available for estimating some forms of degradation (e.g. soil erosion by water, soil organic matter decline and salinisation). Thus there is an existing technical basis for establishing a global soil monitoring system and investment in this should be an urgent priority for the international community.

**Key words:** *soil degradation, food security, monitoring design*

## INTRODUCTION

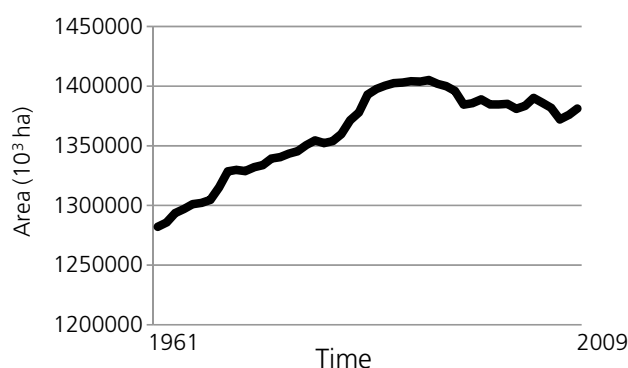
There is widespread and urgent concern in the scientific community (Lal, 2010) about the degradation of global soil resources and its impact on future food security, but this is not yet matched by concerted action by the international community to protect soil resources. This paper addresses the following questions: Why is soil monitoring a global priority? What needs to be monitored? Which options for a global monitoring programme are technically ready for implementation relatively quickly?

## Why is soil monitoring a global priority?

Soil resources underpin the productive capacity of land. Their natural regeneration takes longer than human life times and their degradation is not always reversible, e.g. when accompanied by desertification. Those soils that are most productive are also often those most threatened, either by non-sustainable agricultural practices or extension of the built environment, or both of these. Major expansion of

the agricultural land area allowed exploitation of new soil resources in the last century but there is now a net decrease in the land area in production, as the land available for new expansion has reduced and existing agricultural areas are degraded. Figure 1 shows the change in the total area of arable and permanent crops from 1961 to 2009. Simultaneously with the decline in the area of agricultural land, the demand for food and biofuels is rising and this is expected to encourage a long-term increase in food prices. The commoditization of food combined with established trading institutions and logistics support a global market in food. A consequence is that losses in supply due to soil degradation anywhere have the potential to affect global prices and so food prices for citizens who are far distant from the actual degradation.

Therefore there is a need for global governance of soil resources to ensure that these are conserved to support food availability and constrain food prices for all of humanity, especially the poor. A critical requirement for this governance is a global monitoring programme providing quantitative data of known and documented quality about trends in soil status. This programme is essential to identify which soil resources are most at risk from degradation, estimate the economic and social consequences of soil degradation, support arguments for appropriate policy measures and ensure that there is necessary investment in soil protection. Information from soil monitoring is a prerequisite for convincing politicians that unacceptable risks to food security are presented by the current lack of global soil protection. Furthermore, investment will not be targeted to where it is required without information on where soil resources are at most risk.



**FIGURE 1. Global area of arable and permanent crops — 1961 to 2009 (10<sup>3</sup> ha) (FAOSTAT).**

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## What should be monitored?

Information is needed both about the extent of productive soil resources and their condition. The spatial extent of productive soils may be assessed most directly from remote-sensed data on land cover and national returns on areas of land in agricultural production. Assessing the condition of soil resources means answering the question: "What is the state of the soil and how is it changing?". An integrative measure of soil status could be soil quality or soil health. Neither is ideal for global soil monitoring: soil quality (Karlen *et al.*, 1997) refers to "fitness for purpose" for a defined use, but soils can support a wide range of agricultural goods and services and over time different ones are prioritized so the perceived quality of soil may alter independently of any state change. Soil health is conceptually attractive as it refers to the functioning of the soil as a living system (Kibblewhite, Ritz and Swift, 2008). However, current knowledge is probably inadequate to identify reliable indicators of this functionality and particularly to relate these to agricultural productivity. An established and more promising approach focuses on indicators that quantify the rates and consequences of soil degradation processes, e.g. soil erosion by water and wind, decline of soil organic matter, compaction, contamination, salinization, loss of soil biodiversity and sealing. A comprehensive review of potential indicators for major types of soil degradation (Huber *et al.*, 2008) identified a core set of indicators (Table 1) that have mature, formal and agreed testing procedures and protocols or that are accessible via remote-sensing data. These could be employed with little or no further development. While not complete (e.g. for soil erosion by wind), the set is complete enough to support early implementation of soil monitoring, ideally with global, continental, regional and country reporting categories, and a reporting frequency of less than 10 years to match policy-making and review cycles.

## Options for soil monitoring

### *Spatial extent of soil resources*

Land cover data obtained through satellite-based remote sensing can provide accessible and reliable information on of soil resources in agricultural production systems and the changes to these resources over time as land is brought into agricultural production. Land cover data can also be used to provide information on soil resources being lost to surface sealing through extension of built environment (European Environment Agency, 2011). Therefore there is both a sound technical basis for monitoring the extent of productive soil resources and the availability of data to support this monitoring.

### *Condition of soil resources*

Some on-going estimates of soil degradation can be obtained using remote sensing. For example, soil erosion by water can be estimated

with land cover as the driving variable (Kirkby *et al.*, 2008), but most types of degradation require ground monitoring, not least because the soil surface is often obscured by vegetation. Different design options are available for soil monitoring networks (SMNs) (Arrouays *et al.*, 2012). They need to be sufficiently dense in space and time to provide information about soil state and change that has a low enough uncertainty to be useful. Rates of soil degradation may only be detectable across regions at multi-annual timescales even with higher spatial sampling densities. Many existing national and regional SMNs employ a regular grid-based design to provide an unbiased sample of land characteristics (soil types, land cover, etc.), but extending this approach globally could present insurmountable logistical problems (e.g. access to remote and contested regions) as well as high costs. Collection of data to support estimation of trends in the state of the soil at the continental scale may be achieved most efficiently by sampling strata defined by major soil groups and bio-geographical regions. As well as offering a more efficient sampling strategy, this approach should be able to accommodate logistical and other implementation problems more easily than a grid-based one. If sampling of some locations is not possible, data on trends can still be reported albeit with less confidence. Further investigation and agreement are probably required on within-site sampling protocols, including on whether to sample according to depth or pedogenic horizons.

Taxonomic definitions of soil types are contested but some internationally agreed ones are available (IUSS Working Group WRB, 2006), as well as data on their spatial distribution, the global coverage will hopefully be improved in the near future using digital soil mapping techniques (Sanchez *et al.*, 2009). This information is probably sufficient to identify sampling locations. Existing well-described sites should be used where this is possible and appropriate but investment will be needed to qualify sites with local data and by field-based investigation. The number of sites required, even with strata limited to soil type and bio-geographical region, will be substantial. In excess of 4 000 sites were found to be necessary to detect changes in topsoil organic carbon in Europe according to soil type and land use (Morvan *et al.*, 2008); by extrapolation, perhaps at least ten times this number of sites will be required for global monitoring.

## CONCLUSIONS

Soil resources underpin agricultural productivity and food security. There is an urgent requirement for a global soil monitoring system to provide reliable information to policy makers on trends in the state of soil. Without such information, protection of soil will not be given the correct priority relative to other natural resources.

Remote-sensed data are available now to support monitoring of the extent of soil resources and where these are being lost and at which rates.

**TABLE 1. Qualified indicators for degradation of soil**

| Degradation                | Indicator                                                 | Unit                                |
|----------------------------|-----------------------------------------------------------|-------------------------------------|
| Erosion by water           | Estimated soil loss by rill, inter-rill and sheet erosion | t·ha <sup>-1</sup> ·y <sup>-1</sup> |
| Soil organic matter status | Topsoil organic carbon content (measured)                 | g/kg                                |
| Soil organic carbon stocks | Soil organic carbon stocks (measured)                     | t/ha                                |
| Diffuse contamination      | Metal contents of soil                                    | %                                   |
| Soil sealing               | Sealed area                                               | ha; ha/y                            |
| Land consumption           | Land take                                                 | %; ha                               |
| Salinization               | Salt profile                                              | %; dS/m                             |

Indicators related to key soil degradation processes, erosion by water, decline in topsoil organic carbon, soil sealing are mature and supported by agreed testing procedures.

Early investment in the development of a design for a global SMN is required. Initial consideration indicates that a stratified design covering soil type and bio-geographical regions is preferable to one based on a regular grid. A possible estimate of the number of required sites is between 50 000 and 100 000.

Establishing a global soil monitoring system will take several years and information on trends in soil state will only become available following re-sampling. Realistically, it will be at least a decade and possibly two decades before a global soil monitoring system can start to report. Given the increasing pressure on soil resources for food production, the need for urgent action to establish this system is clear.

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# Legumes in Crop Rotations Reduce Soil Nitrous Oxide Emissions Compared with Fertilized Non-Legume Rotations

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## ABSTRACT

Soil nitrous oxide (N<sub>2</sub>O) emissions were measured from a range of dryland crops and crop rotations in the northern grains region of Australia. The objective was to compare N<sub>2</sub>O emissions associated with the growth and post-harvest residue decomposition of a nitrogen (N<sub>2</sub>)-fixing legume crop with that from N fertilized non-legume crops. From 2009 to 2012 a dryland crop rotation experiment was conducted on a black Vertosol (cracking clay soil) representative of the main soil type used for grain growing in the region. Crop rotation treatments were: canola + N\_wheat + N\_barley + N (CaWB), chickpea\_wheat + N\_barley (CpWB), chickpea\_wheat\_chickpea (CpWCp), and chickpea\_sorghum + N (CpS). Soil emissions of N<sub>2</sub>O were monitored in the field seven to eight times per day using an automated system of chambers connected to a gas chromatograph. Soil mineral N and plant N uptake were measured by regular field sampling. During the project, extremes of cold, hot, wet and dry weather were experienced that were often well below or above long-term averages for the site. Cumulative N<sub>2</sub>O emissions from the four rotations were in the order CaWB > CpS = CpWB > CpWCp. Emissions from CaWB (1 523 g-N<sub>2</sub>O-N-ha<sup>-1</sup>), where all crops were N fertilized, were more than twice those of CpWCp (614 g-N<sub>2</sub>O-N-ha<sup>-1</sup>), where legume N<sub>2</sub> fixation was the external N source. As a proportion of anthropogenic N input, legumes emitted less N<sub>2</sub>O than N fertilized non-legumes. Most emissions from N fertilized crops occurred during early crop growth, while most emissions from legumes occurred during post-harvest decomposition of crop residues. These differences should be taken into account when devising strategies to reduce N<sub>2</sub>O emissions from cropping.

**Key words:** nitrogen, chickpea, wheat, sorghum, barley.

## INTRODUCTION

The concentration of the nitrous oxide (N<sub>2</sub>O), a greenhouse gas in the atmosphere is increasing, largely as a consequence of increased anthropogenic nitrogen (N) inputs into the soil associated with animal and crop production. Emissions of N<sub>2</sub>O from the soil originate from the biological processes of nitrification and denitrification that utilize inorganic N from any source (Bremner, 1997). The susceptibility for

N<sub>2</sub>O loss is determined by the interaction between the rate of supply of inorganic N, the N demand from crops and environmental conditions (Dalal *et al.*, 2003).

There is little evidence that the process of biological N<sub>2</sub> fixation itself contributes directly to total N<sub>2</sub>O emissions (Jensen *et al.*, 2012), but legume crop residues are high in N and are readily mineralized in soil, albeit in a “slow-release” fashion. In contrast, inorganic N fertilizer adds N to the soil in an immediately-available form. Inclusion of N<sub>2</sub>-fixing legume crops into an otherwise cereal dominated crop rotation should reduce the potential for N<sub>2</sub>O losses through a reduction of soil inorganic N during the post-application period. Associated with reduced fertilizer N inputs will be reduced carbon dioxide (CO<sub>2</sub>) emissions from N fertilizer manufacture, transport and urea dissolution. This research aims to quantify the potential for reducing soil N<sub>2</sub>O emissions from dryland cropping by sourcing N from legume N<sub>2</sub> fixation rather than relying on fertilizer N.

## MATERIALS AND METHODS

The experiment was located on a cracking clay soil (Black Vertosol; Isbell, 2002) at the NSW Department of Primary Industries experimental station near Tamworth, NSW, Australia. The surface 0–0.1 m of the soil was 44 percent clay, pH 8.0 (1:5 soil:water), 1.9 percent soil organic carbon, and had a bulk density of 1.0 Mg/m<sup>3</sup>. The soil type is typical of the dominant soil used for dryland cropping throughout the northern grains region of Australia. The following crops were grown in the experiment: canola (*Brassica napus* cv. Hyola 50), chickpea (*Cicer arietinum* var. PBA Hatrick), wheat (*Triticum aestivum* var. Crusader), sorghum (*Sorghum bicolor* var. MR43), and barley (*Hordeum vulgare* var. Shepherd). Plot size was 6 m wide by 12 m long. All plots were sown using a zero-till planter at row spacings of 0.25 m (wheat, barley), 0.5 m (canola, chickpea) and 0.75 m (sorghum). Nitrogen fertilizer rates were based on the projected crop demand minus the soil mineral N supply indicated by pre-sowing soil core testing to 1.5 m. The N fertilizer used was urea, which was applied as a side band at planting at a depth of 0.5–0.1 m in the soil. The trial was a randomized complete block design with four replicates of six treatments (crop rotations). Greenhouse gas measurements were made in three replicates of four crop rotation treatments, namely:

- canola + N\_wheat + N\_barley+N [CaWB]
- chickpea\_wheat + N\_barley [CpWB]
- chickpea\_wheat\_chickpea [CpWCp]
- chickpea\_sorghum + N [CpS]

As sorghum is a summer season crop, the change from winter to summer crops within the rotation meant only two crops were possible in the three-year study period for that treatment. The “+N”

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indicates treatments that had N fertilizer added. Fertilizer N rates were: 80 kg-N-ha<sup>-1</sup> for canola and wheat, 40 kg-N-ha<sup>-1</sup> for sorghum, and 60 kg-N-ha<sup>-1</sup> for barley. After grain harvest by small plot harvester, all plant residue material from a plot was redistributed evenly across that same plot. Weeds, insect pests and plant diseases were controlled chemically using appropriate compounds at approved rates when necessary.

Above-ground whole plant samples (1 m row per plot) were collected monthly during crop growth by hand cutting, their dry matter determined, then analysed for total N concentration (percent N) by combustion analysis (EA1112, Thermo Finnigan). Total biomass N was calculated as biomass dry matter multiplied by biomass percent N. To determine the percentage of legume plant N derived from atmosphere (%Nd<sub>fa</sub>), samples collected at peak biomass were analysed for delta <sup>15</sup>N (δ<sup>15</sup>N) by mass spectrometry after combustion analysis for total N. The natural abundance of <sup>15</sup>N of above-ground plant tissues of legumes was compared with that of a non-legume crop growing nearby. Percent Nd<sub>fa</sub> and total N fixed were calculated according to Unkovich *et al.* (2008). Grain samples from the plot harvester were analysed for percent moisture by oven drying and percent N by combustion analysis. Grain protein was calculated by multiplying measured grain N by a factor of 5.7 and then standardizing results based on a moisture content of 8 percent for chickpea and canola, and 12 percent for wheat, sorghum and barley.

Automatic gas measuring chambers, as described by Scheer *et al.* (2011) were used to measure N<sub>2</sub>O emissions from soil seven to eight times per d. One chamber (0.5 m × 0.5 m × 0.2 m height) was deployed in each of three replicate plots of two treatments, CaWB and CpWB, from June 2009 until April 2010, when the system was upgraded to cover three replicates of four crop rotation treatments for the remainder of the experiment. Chamber height was increased using extensions to cover the erect crops as they grew. Chambers were secured to bases pushed 0.1 m into the soil.

Every three hours, the automatically-operated lids of all chambers in a replicate block were closed for one hour, during which time four separate samples of air were collected 15 min apart and analysed immediately by a gas chromatograph (8610C, SRI Instruments, California, USA) fitted with an electron capture detector for N<sub>2</sub>O measurement. After one hour, the closed chambers opened and all chambers in the next replicate block closed for the hour-long period of measurement, and so on. The N<sub>2</sub>O concentration in the four air samples collected from each chamber during each closure period was regressed against closure time. Patterns of accumulation of soil-emitted N<sub>2</sub>O within a closed chamber during a measurement period have been reported as being either linear or non-linear. A routine developed by Pedersen, Petersen and Schelde (2010) was used that selects the most appropriate model for flux estimation based on the actual data from each measurement period. The routine first fitted a non-linear model to the data. Where this fit was not statistically significant, the linear model was then attempted. If neither model was statistically significant, a slope of zero was assigned. The slope of the selected regression was integrated back to time zero, then used in the calculation of N<sub>2</sub>O flux as described by Scheer *et al.* (2011). Chamber temperature was measured using thermocouple probes, and barometric pressure data for Tamworth was obtained from the Australian Bureau of Meteorology.

Soil sampling for mineral N content (ammonium (NH<sub>4</sub><sup>+</sup>) and nitrate (NO<sub>3</sub><sup>-</sup>) was done by compositing five cores (0.05 m diameter) taken across each plot for surface and sub-surface samples (0–0.1 m and 0.1–0.2 m), and through compositing two cores per plot for deeper samples (0.2–0.3 m, 0.3–0.6 m, 0.6–0.9 m, 0.9–1.2 m, and 1.2–1.5 m). Surface and sub-surface samples were collected approximately monthly with a foot sampler, while deeper cores were

only collected pre-sowing and post-harvest using a vehicle-mounted hydraulic powered coring machine. Soil water content was determined by gravimetric analysis at each time of sampling. Ammonium and NO<sub>3</sub>-N in filtered (Whatman 42) soil extracts (2 M KCl) were determined by standard colorimetric analyses using a flow injection analyser (Lachat Instruments, Colorado, USA).

Nitrous oxide emissions factors (EFs) were calculated by dividing the amount of N lost through N<sub>2</sub>O emissions (output) by the amount of N input as fertilizer or fixed legume N. Where possible, these calculations took into account the background N<sub>2</sub>O emitted from the same soil with no anthropogenic additions of N.

Statistical comparisons of treatment results were carried using Genstat v 14, with individual means tested for difference using the least significant difference test at a probability level of five percent.

## RESULTS

### Crop production

Grain yields and proteins for each crop in each of the four rotation treatments are summarized in Table 1. These were comparable with district averages, especially given the erratic rainfall recorded during the study period (Figure 1). During the 35-month project period, there were nine months (26 percent) where monthly rainfall was less than half the monthly average for the site, and another five months (14 percent) where it was well above the long-term average. These extremes influenced crop production by reducing plant growth in dry times, by reducing yield potential when wet conditions delayed sowing and by reducing yield recovery when harvest was delayed by very wet periods.

In 2009, canola generally yielded more than chickpea (Table 1), but there was no difference in protein content between the oilseed and the legume. There was no significant effect of applied fertilizer N on grain yields of canola (CaWB vs canola with nil N in 2009; results not shown), wheat (CaWB and CpWB vs CpWCp in 2010), nor barley (CaWB vs CpWCp in 2011). However, the application of N fertilizer did increase significantly grain protein of canola (protein for canola with nil N was 22.4 percent), wheat, and barley (Table 1). The addition of N fertilizer also increased the concentration of N in crop biomass during the growing season and the N content of the crop residues remaining after harvest.

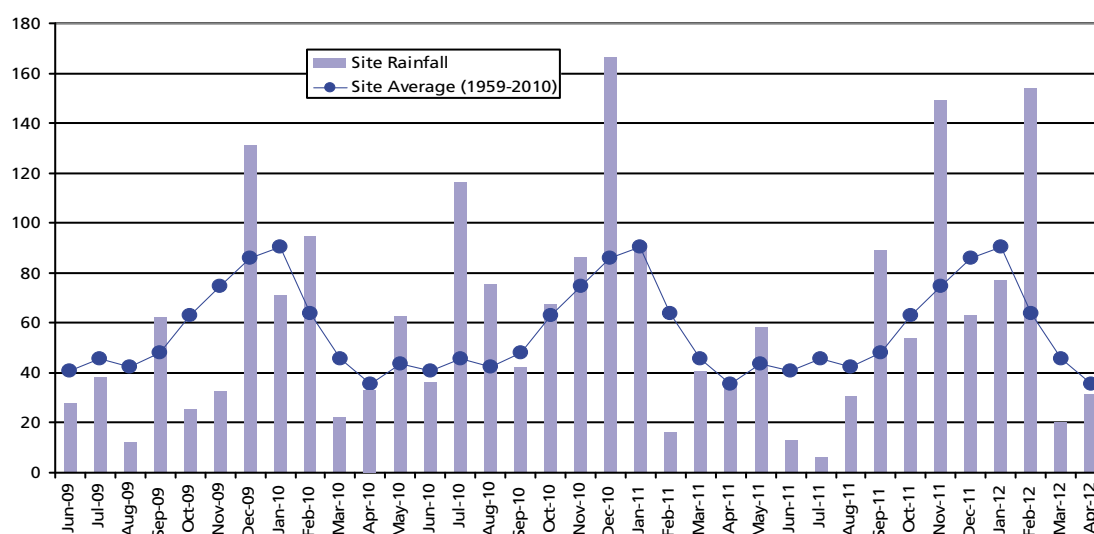
### Plant biomass N

The patterns of accumulation of above-ground (shoot) biomass N in all crops in the crop rotation trial between 2009 and 2012 are presented in Figure 2. In 2009, it is apparent that the N fertilized canola crop was N-rich; much more so than the chickpea crop. Biomass N decreased between the peak in October and the harvest in November because the November biomass mean does not include the harvested oilseed/grain N removed from the paddock. Grain N removal averaged 75 kg-N-ha<sup>-1</sup> for chickpea and 120 kg-N-ha<sup>-1</sup> for the canola. From Figure 2 it can be seen that the difference in biomass N between the peak for canola and the amount remaining in the plant biomass after grain harvest is greater than grain N offtake. Some of the N from the crop's peak biomass was leaf litter by the time of harvest as most canola leaves had fallen as the plant senesced. Some of the N from these fallen leaves had already mineralized in the soil as seen by the increase in mineral N seen in late 2009 in the canola soil results (Figure 3).

Plant N uptake in the 2010 wheat showed a similar level of N in the biomass as in the 2009 chickpea crop. There was no statistical difference in biomass N between fertilized and non-fertilized wheat plots (*p* > 0.05). Only at harvest time was biomass N in the fertilized

**TABLE 1. Grain yield (t/ha) and protein (%) in four-treatments, three-year crop rotation experiment. Means in a row followed by the same letter in brackets were not significantly different ( $p = 0.05$ ); n.s. indicates no significant treatment difference ( $p > 0.05$ )**

| Year | Rotation treatment / Crop / N fertilizer |              |                 |               |
|------|------------------------------------------|--------------|-----------------|---------------|
|      | CaWB                                     | CpWB         | CpWCp           | CpS           |
| 2009 | Canola+N                                 | Chickpea     | Chickpea        | Chickpea      |
|      | 1.8 t ha <sup>-1</sup> (b)               | 1.4 t/ha (a) | 1.3 t/ha (a)    | 1.7 t/ha (ab) |
|      | 23.8%                                    | 21.8%        | 21.6%           | 22.2% (n.s.)  |
| 2010 | Wheat + N                                | Wheat + N    | Wheat           |               |
|      | 3.1 t ha <sup>-1</sup>                   | 3.0 t/ha     | 3.1 t/ha (n.s.) |               |
|      | 14.2% (b)                                | 14.1% (b)    | 13.5% (a)       | Sorghum + N   |
| 2011 | Barley + N                               | Barley       | Chickpea        | 9.6%          |
|      | 3.7 t ha <sup>-1</sup> (b)               | 3.8 t/ha (b) | 2.0 t/ha (a)    |               |
|      | 13.3% (b)                                | 10.5% (a)    | 23.5% (c)       |               |



**FIGURE 1. Mean monthly rainfall (mm) at the Tamworth experimental site during the experimental period, together with the monthly site average taken from 50 years of historical data collected at the site.**

wheat following chickpea slightly greater than that in the fertilized wheat following canola ( $p < 0.05$ ). In 2011, there were much greater differences in biomass N between the chickpea (lowest), non-fertilized barley (after fertilized wheat), and the fertilized barley (highest). However, at harvest time, the N remaining in crop residues showed almost opposite trends with most in the chickpea plots and least in the non-fertilized barley plots where there was also less N to go into the grain, as evidenced by the lower protein results (Table 1).

### Legume N<sub>2</sub> fixation

Chickpea plants grown in 2009 had 18% Nd<sub>f</sub>a. This is low compared with the Australian average of 41 percent (Unkovich, Baldock and Peoples, 2010), probably due to dry conditions during late crop growth and abundant mineral N in the soil. Combining Nd<sub>f</sub>a with the measured plant biomass and multiplying by two to approximate the N in the below-ground biomass of the plant (Unkovich, Baldock and Peoples, 2010), total N fixed by the crop was 49 kg-N·ha<sup>-1</sup>. Since this is extra N to that already present in the soil, it can be counted as an anthropogenic input of N in the calculation of emissions factors (EF) for legume-derived N. The estimated %Nd<sub>f</sub>a for the 2011

chickpea was much higher at 37 percent. However, the plants grew less biomass than in 2009, so the total N input from N<sub>2</sub> fixation was just 41 kg-N·ha<sup>-1</sup>.

### Soil mineral N

Figure 3 shows the soil mineral N in the surface 0–0.1 m and 0.1–0.2 m layers sampled approximately at monthly interval throughout the project period. Nitrate and NH<sub>4</sub><sup>+</sup> N forms are combined in this figure. While NO<sub>3</sub><sup>-</sup> was generally the dominant soil N form, NH<sub>4</sub><sup>+</sup> was also high for certain samplings due to recent additions of fertilizer N or plant residues. In 2009, the high mineral N in the canola + N plots is due to the added fertilizer. From November 2009 to May 2010, mineral N increased through mineralization of the crop residues — more so in the canola + N treatment which had dropped its leaves much earlier than the chickpea. There is a dip in the mineral N of the canola + N plots in January 2010 after heavy rains which likely caused denitrification and NO<sub>3</sub><sup>-</sup> leaching. Mineral N increased subsequently as crop residues continued to mineralize, but decreased again in June 2010 after more heavy rains.

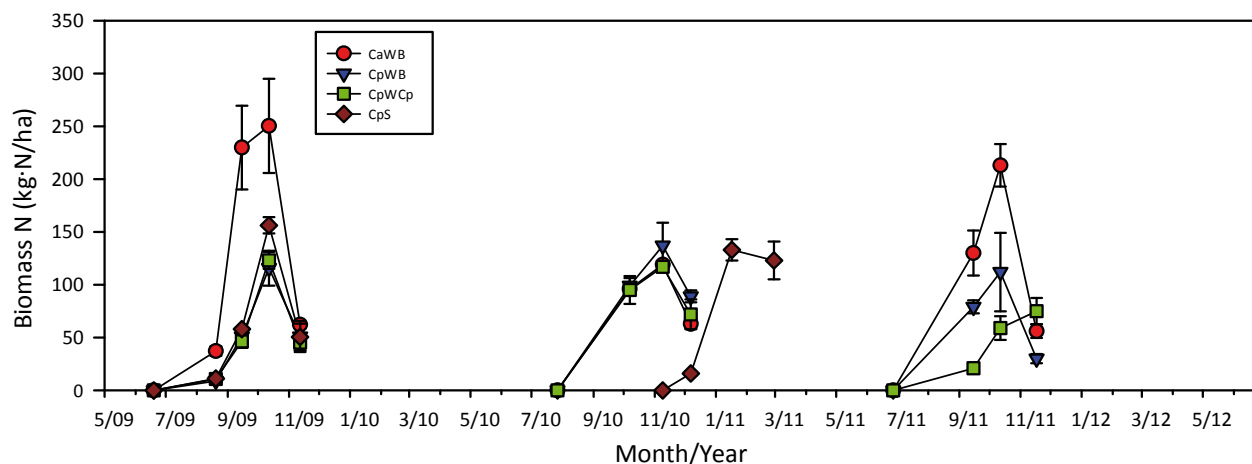


FIGURE 2. Above-ground biomass N in the crops of the four crop rotation treatments. Points are means of three replicate plots with bars indicating standard errors.

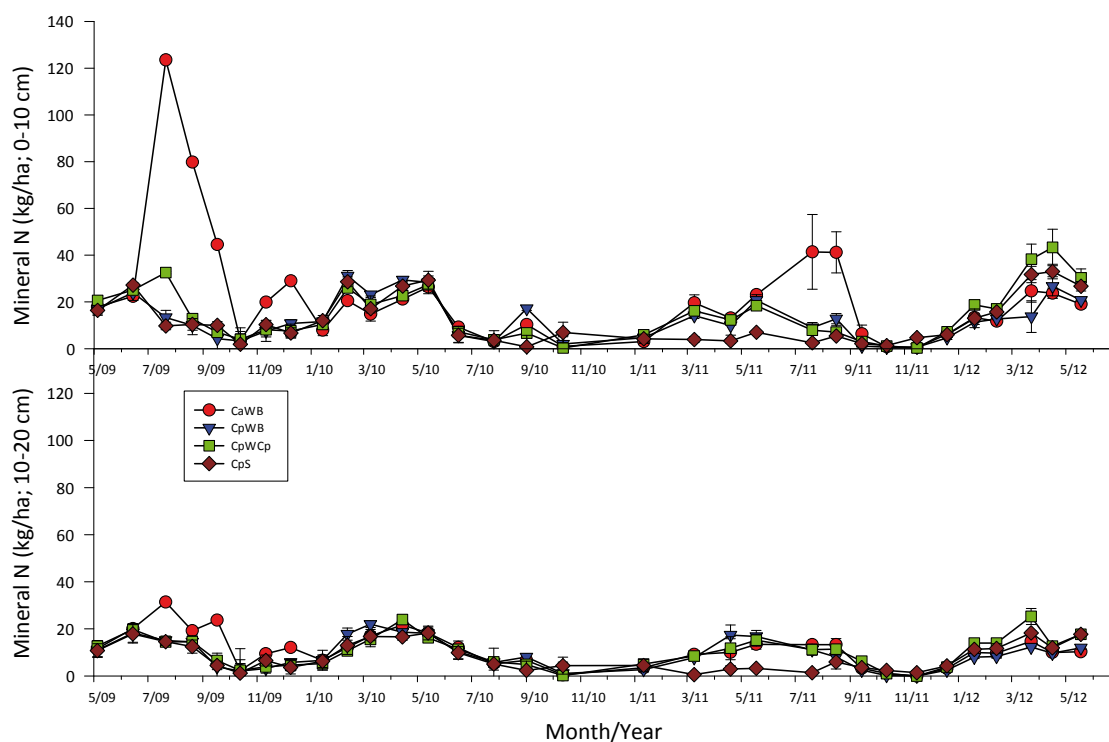


FIGURE 3. Changes in mineral N ( $\text{NO}_3^- + \text{NH}_4^+$  N) with time in the soil of the four crop rotation treatments. Points are means of three replicate plots with bars indicating standard errors.

The addition of fertilizer N to two of the three wheat treatments in late July 2010 led to only modest increases in mineral N in the 0–0.1 m sample depth. Again, heavy rainfall and saturated soil conditions probably led to N losses through denitrification and leaching beyond the sampling depths. Mineral N was barely detectable in the October 2010 sampling after these rains. Soil sampling was not possible in the next two months due to continued wet weather. Evidence for  $\text{NO}_3^-$  leaching during the prolonged wet conditions is shown by the significant difference in protein but not grain yield measured between the fertilized and non-fertilized wheat crops (Table 1). It is likely that some of the leached  $\text{NO}_3^-$  was accessed late in the wheat growing season when the plant was filling the grain.

After the wheat harvest in December 2010, all treatments except CpS (growing sorghum), increased in mineral N during late summer–early autumn as crop residues mineralized. Another increase occurred in July 2011 in the plots that were fertilized with N. However, the difference between N fertilized and non-fertilized treatments had disappeared by September 2011 due to plant uptake (see Figure 2). In 2012, mineral N again accumulated in the soil through continued mineralization of crop residues, with highest accumulation in the post-chickpea fallow.

Measurements of mineral N in the whole soil profile to 1.5 m (not shown) highlighted the strong accumulation of mineral N at the soil surface. Despite this, there was usually some N available throughout

the profile and sometimes a small accumulation below the surface due to  $\text{NO}_3^-$  leaching. Decomposing N-rich canola and chickpea residues, coupled with  $\text{NO}_3^-$  leaching led to mineral N differences down to 0.3 m depth.

### Nitrous oxide emissions

Cumulative emissions of  $\text{N}_2\text{O}$  for all crop rotations from June 2009 to May 2012 are presented in Figure 4. Daily rainfall at the site is shown on the same graph to highlight the strong influence that rainfall had on  $\text{N}_2\text{O}$  emissions in this dryland cropping system. Cumulative  $\text{N}_2\text{O}$  emissions from the four rotations were in the order  $\text{CaWB} > \text{CpS} = \text{CpWB} > \text{CpWCp}$ , with more than twice the emissions from CaWB ( $1523 \text{ g}\cdot\text{N}_2\text{O}\cdot\text{N}\cdot\text{ha}^{-1}$ ) where all crops in the rotation were fertilized with N, compared with CpWCp ( $614 \text{ g}\cdot\text{N}_2\text{O}\cdot\text{N}\cdot\text{ha}^{-1}$ ) where legume  $\text{N}_2$  fixation was the only external N source. The patterns of  $\text{N}_2\text{O}$  emissions across the measurement period were sporadic. Long periods of nil to barely detectable emissions were interspersed with brief periods of high emission activity after heavy rain on saturated soil with either freshly-decomposing crop residue or recently added N fertilizer.

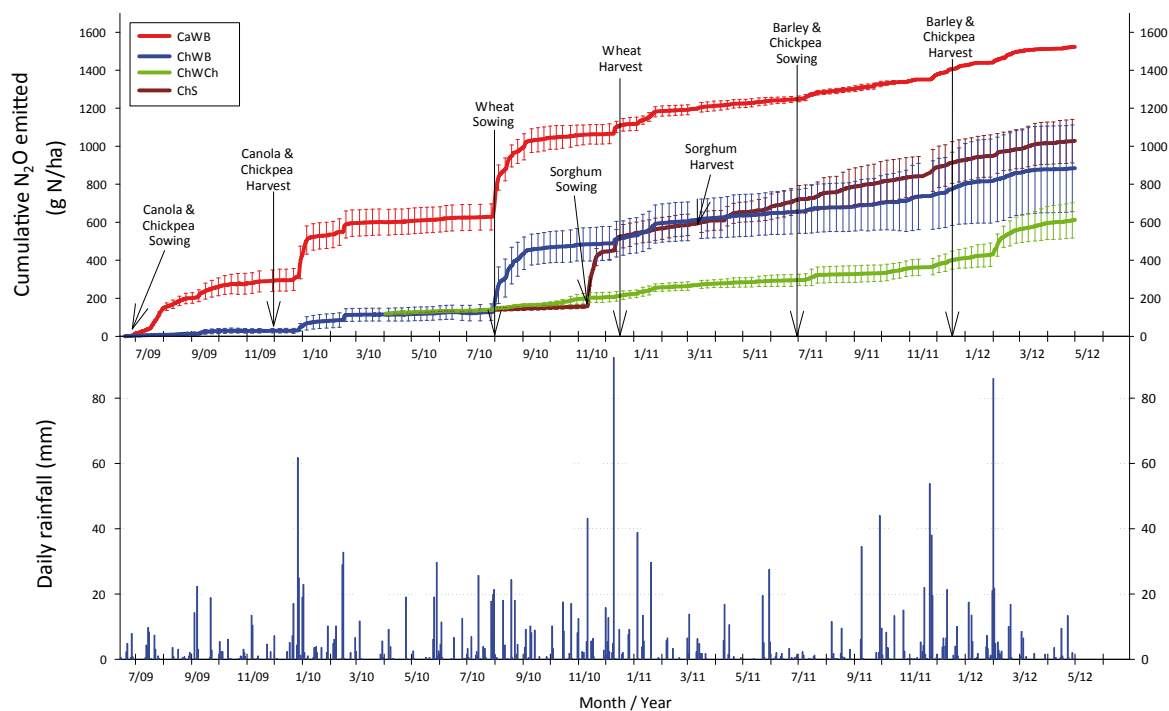
In 2009, emissions from the soil with chickpea were barely detectable during the growing season, while the soil with canola and fertilized with  $80 \text{ kg}\cdot\text{N}\cdot\text{ha}^{-1}$  responded to the N input and moist conditions by releasing  $\text{N}_2\text{O}$ . Between half and one third of the total emissions measured in the first 12 months occurred in just two heavy rainfall events during the summer post-harvest fallow (January–February 2010). The higher losses from the canola treatment compared with the chickpea treatment were likely due to mineralization of N from the leaves that the canola had dropped before harvest (see Figures 3 and 4). In contrast, chickpea, which had less biomass N than the canola to start with, only dropped its leaves around harvest time, after which it was mostly dry until the heavy rainfall in early

January and again in February. Between the early 2009 emissions and the sowing of the following winter crop there were very few emission events despite some very wet soil conditions during May–June 2010. The mineral N measurements in the soil surface (Figure 3) indicate some losses during this time, but these did not lead to significant  $\text{N}_2\text{O}$  emissions.

Emissions of  $\text{N}_2\text{O}$  were very high soon after wheat sowing and fertilizer N application in late July 2010. Sowing was followed by 68 mm of rain in the subsequent three d. The saturated soil conditions meant that the conversion of urea to  $\text{NH}_4^+$  then  $\text{NO}_3^-$  and its subsequent denitrification all occurred in a matter of h to d after the application of the urea. Despite identical amounts of N fertilizer applied to the CpWB and the CaWB plots,  $\text{N}_2\text{O}$  emissions in canola plots were higher, probably due to the earlier inputs of substantially greater amounts of residue N compared with chickpea.

Similarly, 60 mm of rainfall in the week following sorghum sowing in October 2010 led to significant  $\text{N}_2\text{O}$  emissions in the CpS treatment. Although only half the rate of fertilizer N was added to the sorghum ( $40 \text{ kg}\cdot\text{N}\cdot\text{ha}^{-1}$ ) compared with the  $80 \text{ kg}\cdot\text{N}\cdot\text{ha}^{-1}$  applied to the wheat, the magnitude of  $\text{N}_2\text{O}$  emissions from the sorghum plots were similar, so the rate of gaseous loss was double in the sorghum plots.

During 2011,  $\text{N}_2\text{O}$  emissions were consistently low throughout much of the year, with only isolated emission activity in response to rainfall. Of particular note was the lack of emissions following the addition of 60 kg fertilizer N/ha to barley in the CaWB treatment, sown in May 2011. This is despite the addition of the fertilizer clearly raising the concentration of mineral N in the surface soil (see Figure 3). It was a very dry start to the 2011 growing season (see Figures 1 and 4) with June, July and August all well below the long-term average rainfall. By September, when rainfall did return to normal, the wheat crop had taken much of the added fertilizer N into



**FIGURE 4.** Cumulative emissions of  $\text{N}_2\text{O}\cdot\text{N}$  for the four crop rotation treatments for the entire experimental period (top graph). Lines are means of three replicate plots with bars indicating standard errors. Bottom graph shows daily rainfall recorded at the site during the same period.

the biomass (see Figure 2), so that it was no longer susceptible to loss as N<sub>2</sub>O. Heavy rainfall in November 2011 did cause N<sub>2</sub>O emission activity, but the restricted supply of available NO<sub>3</sub><sup>-</sup> for denitrification meant losses were not large.

After the 2011 winter crop harvest in December, warm moist conditions continued to aid mineralization of the crop residues until heavy rainfall in early February led to pronounced emissions of N<sub>2</sub>O from the post-chickpea fallow. The N rich legume in this instance had mineralized more than the low N barley crop residues (see Figure 3), while the post-sorghum fallow had also mineralized more and consequently lost more than the barley residue plots. Again, there was no difference in losses between the post barley + N plots and those without N, as there was little difference in the harvest residue N contents (see Figure 2). This is likely because the + N treatment had channelled much of the additional N into grain protein which was removed from the paddock.

### Nitrous oxide emissions factors

Emissions factors for all the experimental crops are listed in Table 2. Since there were just six chambers for the first crop, measurement of background emissions from a non-N fertilized canola crop or a non N<sub>2</sub>-fixing chickpea crop was not possible in 2009. However, background emissions were measured for 2010 wheat and sorghum, and for the 2011 barley and chickpea crops. The smallest EF's were for the N fertilized barley in 2011 and N<sub>2</sub>-fixing chickpea in 2009 and 2011, while the largest EF was for N fertilized sorghum in 2010–11. The wheat and sorghum EFs were largely determined by saturated soil conditions immediately after sowing and N fertilizer application. In contrast, losses of N<sub>2</sub>O from chickpea plots were negligible during crop growth, but greater during the post-harvest fallow when

plant residues were decomposing. The 2011 barley results indicate that N<sub>2</sub>O emissions were reduced to a minimum when the mineral N from fertilizer was utilized by the crop and not subject to denitrifying conditions. These results showcase the range of emission scenarios possible in a highly variable rainfall environment coupled with a non-strategic fertilizer application programme.

The continuity of measurements during the three-year project period made it possible to compare total N<sub>2</sub>O emissions across the four crop rotation treatments. No background emissions were subtracted from the total emissions in calculating these factors, but the background would have been the same for all treatments. Table 3 shows that the whole of rotation EF's were the same for the three winter-crop rotations (CaWB, CpWB, CpWCp), whereas the EF for the mixed winter-summer rotation (CpS) was 70% higher than winter-crop rotation. The latter result is particularly significant because this rotation had a similar total N input as the CpWCp treatment, but had two-thirds more N<sub>2</sub>O emissions.

### DISCUSSION

There have been several studies of N<sub>2</sub>O emissions from N fertilized dryland wheat on cracking clay soils in Australia, with yearly total N<sub>2</sub>O emissions from crops and crop sequences similar to those in this experiment ranging from 0.50 kg-N<sub>2</sub>O-N-ha<sup>-1</sup> in western Victoria (Officer *et al.*, 2008) to 0.90 kg-N<sub>2</sub>O-N-ha<sup>-1</sup> in southern Queensland (Wang *et al.*, 2011). By comparison, 0.60 kg-N<sub>2</sub>O-N-ha<sup>-1</sup> was measured here for fertilized wheat. The EF's for wheat (0.46–0.59 percent) were less than the three-year average for no-till, stubble retained wheat figure of 0.77 percent measured by Wang *et al.* (2011), but greater than the figure of 0.14 percent for the N fertilized, non-irrigated treatment in the study by Officer *et al.* (2008). The figure

TABLE 2. N<sub>2</sub>O emissions factors for all crops in the four-treatment, three-year crop rotation experiment

| Crop                     | Total N added* | Crop N <sub>2</sub> O Emissions Factor (%) |                                     |
|--------------------------|----------------|--------------------------------------------|-------------------------------------|
|                          | (kg-N/ha)      | (crop growth only)                         | (crop growth plus post crop fallow) |
| canola (2009)†           | 80             | 0.36 ± 0.07                                | 0.78 ± 0.09                         |
| chickpea (2009)†         | 49             | 0.06 ± 0.02                                | 0.26 ± 0.08                         |
| wheat (after canola)     | 80             | 0.51 ± 0.05                                | 0.59 ± 0.07                         |
| wheat (after chickpea)   | 80             | 0.39 ± 0.07                                | 0.46 ± 0.11                         |
| sorghum (after chickpea) | 40             | 0.92 ± 0.12                                | 1.31 ± 0.24                         |
| barley (2011)            | 60             | 0.07 ± 0.03                                | 0.08 ± 0.02                         |
| chickpea (2011)          | 41             | -0.04 ± 0.05                               | 0.22 ± 0.16                         |

\* N added as urea fertilizer (canola, wheat, sorghum, barley) or N<sub>2</sub> fixation (chickpea)

† No background emissions data were available for calculation of canola 2009, or chickpea 2009

TABLE 3. N<sub>2</sub>O emissions factors for the four crop rotations

| Crop rotation treatment | Total N added*           | Total N <sub>2</sub> O-N emitted | Rotation N <sub>2</sub> O Emissions Factor† |
|-------------------------|--------------------------|----------------------------------|---------------------------------------------|
|                         | (kg-N-ha <sup>-1</sup> ) | (g-N-ha <sup>-1</sup> )          | (%)                                         |
| CaWB                    | 80 + 80 + 60 = 220       | 1 523 ± 5                        | 0.69 ± 0.00                                 |
| CpWB                    | 49 + 80 + 0 = 129        | 885 ± 228                        | 0.69 ± 0.18                                 |
| CpWCp                   | 49 + 0 + 41 = 90         | 614 ± 93                         | 0.68 ± 0.10                                 |
| CpS                     | 49 + 40 = 89             | 1 028 ± 114                      | 1.16 ± 0.13                                 |

\* N added as urea fertilizer (canola, wheat, sorghum, barley) or N<sub>2</sub> fixation (chickpea)

† No background emissions were subtracted

reported here came from a single year's measurement that was strongly influenced by heavy rainfall immediately after N fertilizer application into an already wet soil, so it is likely that more years of measurement would produce an average EF for wheat that is lower, as evidenced by the EF of 0.08 percent calculated for the barley crop grown in much drier conditions the following year.

Only Barton *et al.* (2011) have published Australian data on N<sub>2</sub>O emissions during and after a dryland legume crop growing season. They found effectively nil additional N<sub>2</sub>O emissions in the 12 months from planting a lupin crop (crop plus post-harvest fallow), once background emissions had been subtracted. However, their study was with lupins grown on a sandy soil in an arid Mediterranean climate (annual rainfall < 300 mm), whereas the present study was on a cracking clay soil in a subtropical, moderate rainfall region (annual average 680 mm). A review by Jensen *et al.* (2012) provided a mean N<sub>2</sub>O emission value for chickpea (crop plus post-harvest fallow) of 0.06 kg-N<sub>2</sub>O-N-ha<sup>-1</sup> (range 0.06–0.16), but they did not indicate whether these figures were corrected for background emissions. In comparison, 0.13 kg-N<sub>2</sub>O-N-ha<sup>-1</sup> was measured here for 2009 chickpea (uncorrected), and 0.32 (0.10 after correction) kg-N<sub>2</sub>O-N-ha<sup>-1</sup> for 2011 chickpea. In this study, N<sub>2</sub>O emissions during chickpea growth were negligible, but relatively high emissions were recorded during the post-harvest fallow period. For legume crops, the biologically-fixed N input into the system happens as the plant growth demands it. Any available soil mineral N is typically used by the legume first before it fixes additional N, hence during the growing season there is very little soil NO<sub>3</sub><sup>-</sup> N available for potential denitrification losses. For legumes it is the release of N from the decomposing crop residues that is potentially a significant contributor to N<sub>2</sub>O emissions (Jensen *et al.*, 2012), as seen in the 2010 summer fallow after 2009 chickpea, and the 2012 summer fallow after the 2011 chickpeas.

With the absence of Australian published data, the current Australian EF for N<sub>2</sub>-fixing crops and pastures remains at the Inter-Governmental Panel on Climate Change (IPCC) default of 1.25 percent of N fixed (Australian Government, 2012). The measurements here in two dryland chickpea crops have shown this figure to be a significant overestimate, with an EF of 0.26 ± 0.08 percent in 2009 and 0.22 ± 0.16 percent in 2011. The 2009 EF would actually be lower than this as there were no background measurements at the same time with which to correct it. If the mean annual background measurements from the other years in the project were used instead, then this EF would have been -0.10 ± 0.05 percent. However, background emissions during the first year would likely have been lower, given that year was much drier than the following two years. Nevertheless, it is considered that the current EF for dryland pulse crops needs to be lower, in line with the results obtained here and those of Barton *et al.* (2011).

## CONCLUSIONS

Both overall N<sub>2</sub>O emissions and emissions as a proportion of anthropogenic N inputs were reduced by incorporating chickpea, an N<sub>2</sub>-fixing legume, into an annual crop rotation in the place of non-legume crops receiving N fertilizer. Also, the timings of N<sub>2</sub>O loss from fertilizer application to winter cereal crops and of N input to the soil-plant system through legume N<sub>2</sub> fixation are different, and should be considered separately to mitigate emissions from the grains industry. For a winter cereal crop, the danger period is between application of the fertilizer at sowing and its uptake by the growing crop over the ensuing months of post-planting. In contrast, N<sub>2</sub>O emissions during legume growing season in a dryland crop rotation are negligible, as the growing plant uses the available mineral N in the soil before fixing additional N to meet crop N demand. For an

annual winter legume crop it is the period after grain harvest (during summer and autumn), the risk is high for N<sub>2</sub>O emission. Heavy rainfall and saturated soils during this period can trigger emission losses from the mineral N derived from the decomposition and mineralization of legume crop residues.

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# Nuclear Technologies in Global Warming: Assessing the Greenhouse Gas Effects Caused by Huge Biofuel Production in Indonesia

S.H. Waluyo<sup>1,\*</sup>

## ABSTRACT

Indonesia has developed a huge oil palm (OP) plantation industry and its contribution to global warming is unavoidable. Being the country with the largest OP production and the highest annual deforestation rate, Indonesia could contribute significantly to increasing greenhouse gas (GHG) emissions and exacerbate global warming. Oil palm production obviously puts heavy pressure on ecological processes such as land use, GHG balances, regional water and nutrient balances, erosion and biodiversity. A comprehensive understanding and proper assessment of the ecological changes arising from OP production are essential to ensure that OP cultivation has positive and sustainable impacts on climate. Measurements using stable isotopes of hydrogen (H), carbon (C), nitrogen (N), oxygen (O), and sulphur (S) and their ratios at natural abundances in the environment can address such large-scale ecological changes, detect the impacts at an early stage, assist in mitigation and give essential information that needs to be considered by government in formulating national biofuel policies. Studies of individual components of ecosystem C budgets and their environmental control can improve understanding of ecosystem function and its potential response to climate change. Measurement and analysis of C isotope ratios of leaf and atmospheric carbon dioxide (CO<sub>2</sub>) samples can provide integrated information about important plant physiological characteristics spatially and temporally, such as variation in C isotope discrimination in natural forest, OP monoculture and pasture or other agricultural practices. Further, employing an approach using multiple isotopes to assess simultaneously the ratios of deuterium:hydrogen (D:H), oxygen-18:oxygen-16 (<sup>18</sup>O:<sup>16</sup>O) and/or carbon-13:carbon-12 (<sup>13</sup>C:<sup>12</sup>C) in different compounds, and the delta hydrogen-2 (δ<sup>2</sup>H) and delta oxygen-18 (δ<sup>18</sup>O) of water (H<sub>2</sub>O), the delta carbon-13 (δ<sup>13</sup>C) and delta nitrogen-15 (δ<sup>15</sup>N) values of dissolved compounds in stream discharge from watersheds and in the tissues of organisms provide a unique means to investigate the coupling of H<sub>2</sub>O and C fluxes at various temporal and spatial scales, and to develop sustainable land-water management practices. Water scarcity is the key limiting factor for OP production in many contexts. Major allocations of water for OP production have the greatest impact on local water resource balances, disturbing stream flows and availability of fresh water. In addition, there will be loss of biodiversity and food-fuel competition as a result of PO development. Lastly, elimination of

natural food by land-use change will harm food security and increase food prices. Developing an isotope monitoring network and spatial modelling based on isotopic measurements of atmospheric inputs, ecosystem outputs, changes between inputs and outputs within ecosystems and sentinel organisms as integrators and indicators of ecological change are very useful to detect and understand ecological changes at a continental scale. Understanding the underlying or indirect causes of deforestation and the development of an isotope monitoring network are crucial for informing environmental policy makers in Indonesia about managing efforts for mitigation to cope with the global climate change within the Reduced Emissions from Deforestation and Forest Degradation (REDD) scheme in which Indonesia has pledged to reduce GHG emissions by 41 percent by 2020. Employment of nuclear isotopic techniques (NITs) in Indonesia could contribute greatly to better conservation of forest, agricultural and peat land resources through improving soil and water conservation practices and reducing GHG emissions.

**Key words:** oil palm production, greenhouse gases, climate change, forest and peat land conversion, isotope techniques, environmental and ecosystem effects.

## BIOFUEL DEVELOPMENT IN INDONESIA

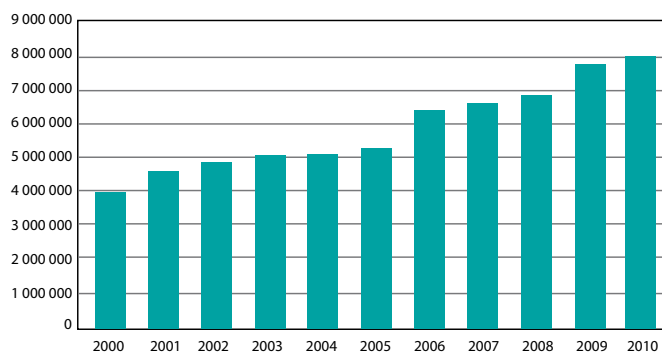
Indonesia is one of the new big emerging biofuel countries. The President of Indonesia has strongly supported the strengthening of national energy security by seeking alternative energy sources. One of the alternatives is development of biofuels, since the raw materials are abundantly available around the country (oil palm, jatropha, sugar, cassava, maize etc). The use of five percent biofuel in the national energy mix is targeted by 2025. This policy has already been started since 2006 with Presidential Instruction No. 1, 2006, Presidential Regulation No. 5, 2006 and Presidential Decree No. 10, 2006 (Caroko *et al.*, 2011).

Oil palm (OP) has the most potential for biofuel production in Indonesia. Indonesia is the largest producer of crude palm oil (CPO) in the world, producing almost half of the world's palm oil. Production in Indonesia increased sharply from 7 Mt in 2000 to around 23 Mt in 2011, with OP plantations expanding rapidly by 11.8 percent per year. Huge oil palm plantations were developed in the last decade on the islands of Sumatra, Kalimantan and Papua (Figure 1). Nearly 11 Mha was allocated in 2011 and it is planned to expand this up to 20 Mha (Colchester *et al.*, 2006; Schoneveld, 2010; Caroko *et al.*, 2011).

Two-thirds of the current expansion of OP cultivation is based on the conversion of rainforests and one third on previously cultivated or until now fallow land. Of the converted rainforest areas, one

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**FIGURE 1. Oil palm plantation areas (ha) in Indonesia (Ministry of Industry, Republic of Indonesia).**

quarter has peat soil with a high C content, and the expected share from peat soils is expected to be 50 percent by 2030 (Bringezu *et al.*, 2009).

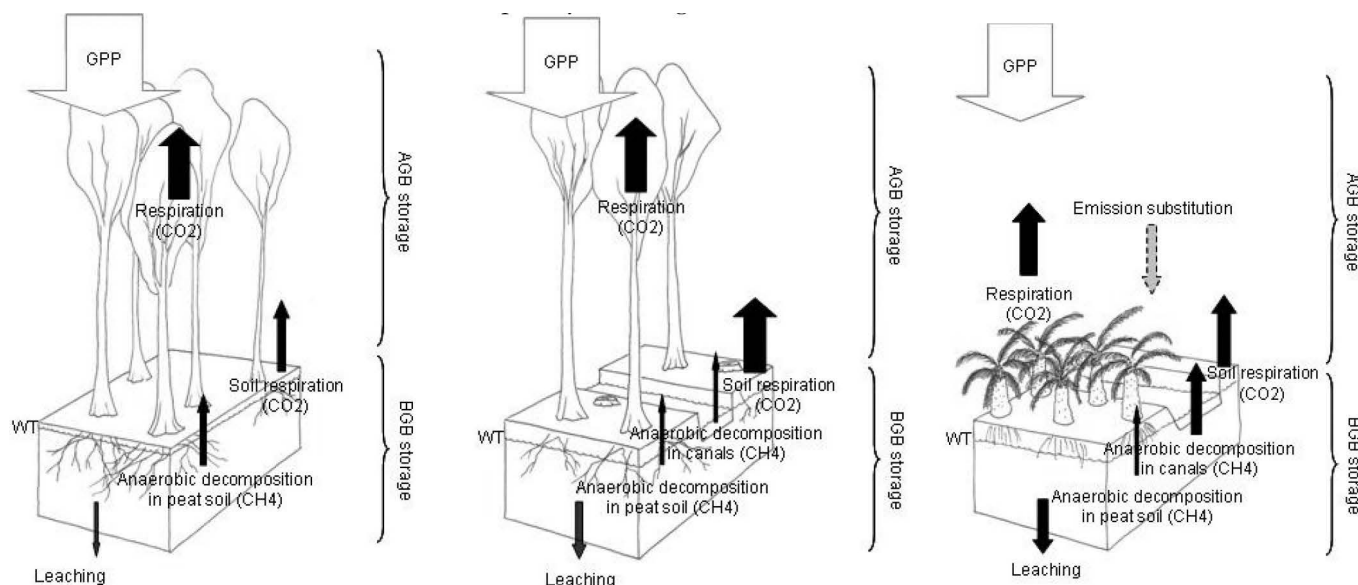
Indonesia has experienced extraordinary land cover changes over the past few decades. The deforestation rate was 2 percent ( $\approx 1.87$  Mha/yr) between 2000 and 2006, and this is believed to be the highest in the world. As a result, the forest area decreased from 119.7 Mha in 1985 to only 88.5 Mha in 2005. Assuming the current trend continues, the total forest area of Indonesia would be reduced by 29 percent compared with 2005, and would cover only about 49 percent of the original area in 1990 (Hooijer *et al.*, 2006; Bringezu *et al.*, 2009; Ekadinata *et al.*, 2011) leading to severe environmental consequences such as soil erosion and degradation, water pollution, loss of bio diversity and greenhouse gas emissions.

### Greenhouse gas emissions in oil palm production

Greenhouse gas emissions occur at all points in OP production cycle (Murdijarso *et al.*, 2010; Achten and Verchot, 2011). Carbon dioxide

is the most important GHG emitted from drained peat lands (Figure 2), contributing 98 percent or more of the total combined global warming potential (GWP). Significant amounts of stored C are lost at all stages of land use conversion and plantation management processes. Annual carbon dioxide ( $\text{CO}_2$ ) emissions from direct land conversion alone are estimated at around 1.83 Gt  $\text{CO}_2$ , and are even higher under OP plantation on peatland (Ravindranath *et al.*, 2009). Forest conversion on mineral soils to promote continued OP cultivation causes a net release of approximately 650 Mg  $\text{CO}_2\text{-eq}\cdot\text{ha}^{-1}$ . Peat forest conversions release over 1 300 Mg  $\text{CO}_2\text{-eq}$  per 25-yr cycle, and continuous decomposition augments the emission of 800 Mg  $\text{CO}_2\text{-eq}$  each cycle per ha (Germer and Sauerborn, 2007). Fargione *et al.* (2008) calculated that the conversion from forest peatland to palm oil releases 3 452 t  $\text{CO}_2\cdot\text{ha}^{-1}$  and that it would take  $>420$  yr to replenish C losses caused by habitat conversion.

Methane ( $\text{CH}_4$ ) fluxes in drained tropical peatland are insignificant relative to losses of  $\text{CO}_2$ , both in terms of the mass of C lost and overall climatic impact. Current research indicates that  $\text{CH}_4$  emissions can be very high in drainage canals and form a substantial part of the GHG emissions in tropical peat lands that are converted to plantations (canals make up 3–5 percent of total plantation areas). However, this potentially important source of  $\text{CH}_4$  remains to be quantified. Carbon lost through leaching seems to be prominent in OP plantations since particulate organic carbon (POC) and dissolved organic carbon (DOC) can easily flow away from the land. Rates of peat nitrous oxide ( $\text{N}_2\text{O}$ ) fluxes in OP plantations also remain uncertain, and there is limited data concerning the magnitude and dynamics of emissions, particularly following fertilizer application. Heavy fertilization may stimulate decomposition and soil temperatures which are generally higher in plantations than in forest, and may increase microbial activity influencing denitrification. According to the Intergovernmental Panel on Climate Change (IPCC) guidelines, one percent of fertilizer applied is emitted as  $\text{N}_2\text{O-N}$  with GWP of 296 times greater than  $\text{CO}_2$ . Based on a literature review, GHG emissions related to the use of artificial fertilizers and pesticides are in



**FIGURE 2. Land-based C fluxes on primary/and rained (left), on drained + partly logged peat swamp forest ecosystem (middle) and on oil palm plantation peat (right) (Verwer, van de Meer and Nabuurs, 2008).**

the order of 1 000–1 500 kg-CO<sub>2</sub>-eq-ha<sup>-1</sup>·yr<sup>-1</sup>. While based on average yields (3.2–4.0 t-PO-ha<sup>-1</sup>·yr<sup>-1</sup>), GHG emissions per t of CPO are in the order of 250–470 kg-CO<sub>2</sub>-eq. It is worth restating that it is only the C released from decomposition of historically accumulated peat that is of relevance to global C emissions and anthropogenic climate change (Germer and Saurborn; 2007; Brinkmann Consultancy, 2009; Page *et al.*, 2011).

Indonesia is the world's third largest emitter of GHGs after the United States and China. Emissions are heavily dominated by deforestation, and more than half of the emissions come from the land-use, land-use change and forestry (LULUCF) sector (USAID Indonesia, 2008; Amron *et al.*, 2010; Ekadinata and Dewi, 2011). Almost 84 percent (57.4 Gt C) of the peat land C resources are located in Indonesia, while conversion of tropical peat forest to OP plantation increases GHG emissions significantly (Page *et al.*, 2011). To mitigate global climate change, the Government of Indonesia has declared its commitment to reduce GHG emissions by 26 percent by 2020 by a business as usual approach and by a further 15 percent with international support. Most of the reduction is targeted to come from the LULUCF.

### Impacts of oil palm production on water resources

Large-scale deforestation will tend to change the regional water balance through reducing the evapotranspiration flux to the atmosphere. This will be accentuated as deforestation continues and an increasingly higher ratio of forest edge to undisturbed forest. A dense plant cover and relatively high surface air temperatures make the "biological water pump" very effective, returning a major part of precipitation back to the atmosphere. Deforestation makes the biological water pump weak, and will cause more water to run off to rivers and the local temperatures will rise. The effects of the reduction in the amount of water returned to the atmosphere will likely vary according to the size of the cleared area. However, from a meteorological point of view, the effects of deforestation may be somewhat greater in scope than the actual size of the deforested area (IAEA, 1990; Nguyen *et al.*, 2011).

Scarcity of water has proven to be the key limiting factor for OP production in many contexts. During the growing period, OP needs about 5.6 mm water·d<sup>-1</sup>·tree<sup>-1</sup> (equal to 150–200 L·d<sup>-1</sup>·tree<sup>-1</sup>) (Kartika and Sosiawan, 2010). Further, the processing of feedstocks into biofuels can use large quantities of water, mainly for washing plants and seeds and for evaporative cooling. Massive irrigation infrastructure has to be provided for commercial yield levels and major groundwater resources around the OP plantation of local communities have been diverted. Therefore, OP plantation decreases water resources, and mainly the groundwater resources which are required by local communities, although the potential for expansion of irrigated areas appears to be high in some areas on the basis of available water resources and land.

Oil palm plantations also cause deterioration of water quality. Converting forest to OP fields may exacerbate problems such as soil erosion, sedimentation and excess nutrient N (nitrogen) and P (phosphorus) run-off into surface waters and infiltration into groundwater from increased fertilizer application. Cultivation also requires a lot of pesticides for optimum production, which often leach into rivers, contaminating the water. Currently, around 25 different pesticides are used. The most commonly used weedkiller is paraquat-dichloride, which is very toxic and accumulates in soil with repeated applications. Its toxicity and accumulation negatively affect the ability to use the land as a source for food production. Water quality is also worsened by the overflow or dumping of untreated POME into

waterways, which threatens community health and reduces aquatic diversity (Colbran and Eide, 2008).

### Impacts of oil palm production on soil resources

Forest ecosystems have a finely balanced nutrient system and deforestation will affect the dynamics of soil resources of N, sulphur (S) and other nutrients. Increased biofuel production will be achieved through improved land productivity and through expansion of cultivated area using existing cropland as well as less productive land. However, it is more likely that biofuels will intensify the pressure on fertile lands where higher returns can be achieved.

Establishment of OP plantations by clearing vegetation and constructing roads and drainage canals will reduce the permeability of the land, cause a loss of soil fauna activity and compact the land, all of which increase top soil run-off and cause soil erosion and affect the fertility and quality of soils. The top soil of OP plantations are prone to erosion which reduces soil fertility, and the use of fertilizers and pesticides will contaminate water resources while sediment loads in rivers and streams will increase significantly (Soyka, Palmer and Engel, 2007; Williams *et al.*, 2007; Murdiyarso *et al.*, 2010). The real danger will ultimately fall on humans and arise from elimination of a natural food while major re-allocations of water will impact negatively on food security, food prices and availability of fresh water.

In addition, there will be loss of biodiversity mostly as a result of habitat loss, increased invasive species and nutrient pollution. Nutrient emissions to water and air resulting from intensive OP cropping will impact species composition in aquatic and terrestrial systems.

Social and economic pressures will also be increased to provide fresh water, food, fuel and wood products for subsistence use or for export, and soil degradation, erosion and leaching of nutrients may reduce the subsequent ability of the ecosystem to act as a C sink.

### Methods for assessing ecological changes caused by greenhouse gas effects

Forest ecosystems are controlled primarily by the interactions between water, oxygen, nutrients, carbon and microflora. These environmental structures can be modified by changes in land use due to OP plantation. Carbon dynamics are modulated primarily by the biota, especially through photosynthesis and respiration processes. It is essential to understand how plants affect the C cycle and ecosystem functioning (Prentice, 2001; Malhi, 2002). Quantifying and mitigating the potential of large-scale OP cultivation on carbon emissions is important to reduce its impact on climate change.

Information about the effects of deforestation on GHG emissions has basically come from methods of measurement such as flux chambers (Figure 3), eddy covariance, DOC and POC and subsidence monitoring for estimating C and GHG fluxes and budgets in tropical countries. It is important to note that each technique has its advantages and disadvantages, largely relating to the spatial and temporal scales of measurement. However, it is critical to have a clear understanding of exactly which components of the C and GHG budgets are measured by each method and, perhaps more importantly, which components are not measured or cannot be differentiated adequately (Page *et al.*, 2011).

One important approach is the measurement of stable isotope ratios present at natural abundances in the environment. Stable isotope measurements of key elements (H, C, N, O, and S) are an important component of ecological monitoring. Their advantages derive from their ability to integrate source and process information as well as their often greater sensitivity to ecological perturbations than elemental or compound concentrations or fluxes in nature.



**FIGURE 3.** Closed chamber measurements being made in the field; large static chamber (left) and dynamic chamber and CO<sub>2</sub> analyzer (Page *et al.*, 2011).

Indeed, stable isotope measurements can capture fundamentally different aspects and dimensions of ecosystem change that cannot be realized with the conventional types of environmental measurements (Williams *et al.*, 2007; Nguyen *et al.*, 2011).

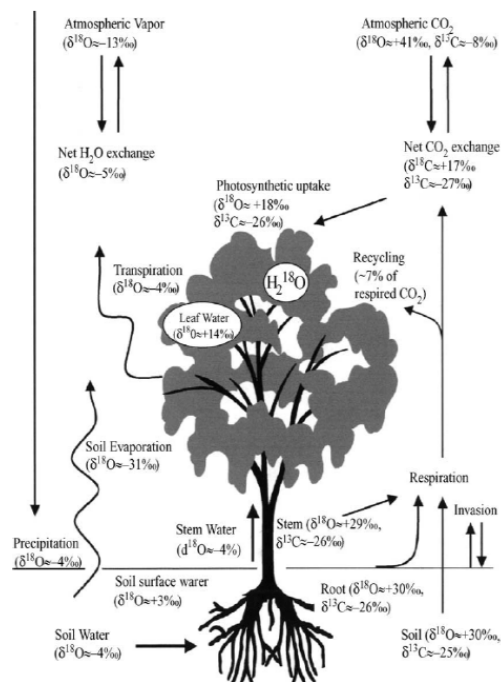
Natural variation in the stable isotope ratios of light elements in both biotic and abiotic components of ecological systems occur as a result of biological and physical fractionation events within an ecosystem. One important consequence of this is that sources of elements and material fluxes can be traced at large scales since different sources often have different isotope ratios based on natural fractionations in the environment. The isotope ratios of organic and inorganic substances also provide a temporal integration of significant metabolic and geochemical processes on the landscape. Further, the isotope ratios of well-mixed environmental reservoirs as reflected, for example, in the delta hydrogen-2 ( $\delta^2\text{H}$ ) and delta oxygen-18 ( $\delta^{18}\text{O}$ ) of water, the delta carbon-13 ( $\delta^{13}\text{C}$ ) and delta nitrogen-15 ( $\delta^{15}\text{N}$ ) of dissolved compounds in stream discharges from watersheds and in the tissues of organisms, represent an integration of source inputs that extend over large spatial scales and the processing of elements within ecosystems.

## NUCLEAR TECHNOLOGIES IN ASSESSING GREENHOUSE GAS EFFECTS

To date, deforestation for OP production in Indonesia is the overwhelming cause of ecosystem degradation (IAEA, 1990; Dawson and Siegewolf, 2007; Nguyen *et al.*, 2011). The major links between C, H and O in atmospheric and terrestrial ecosystems are shown in Figure 4.

One particularly powerful approach is to employ multiple isotope approaches to assess/measure simultaneously the deuterium:hydrogen (D:H), ( $^{13}\text{C}:^{12}\text{C}$ ) and/or ( $^{18}\text{O}:^{16}\text{O}$ ) ratios in different compounds to provide a unique means to investigate the coupling of water and C fluxes at various temporal and spatial scales. The use of stable isotopes has yielded significant knowledge breakthroughs such as partitioning of CO<sub>2</sub> (using carbon-13 [ $^{13}\text{C}$ ]) fluxes in terrestrial ecosystems between photosynthesis and respiration ( $^{13}\text{C}$  and  $^{18}\text{O}$ ), separating autotrophic and heterotrophic respiration in soils (using  $^{13}\text{C}$ ), and quantifying atmospheric N<sub>2</sub> inputs ( $^{15}\text{N}$ ) and their impacts on ecosystem functions (Ghosh and Brand, 2003; Martinelli *et al.*, 2007; Werner *et al.*, 2007).

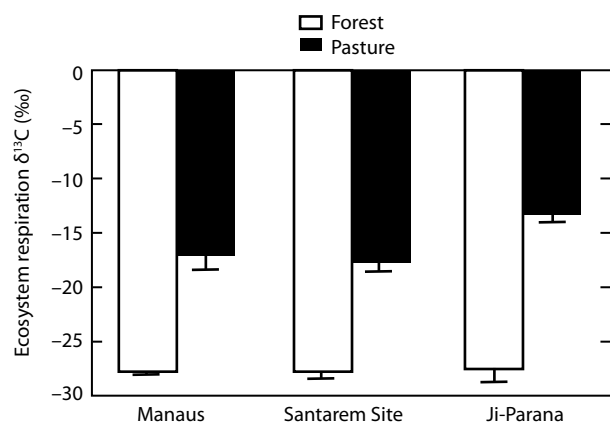
Isotope monitoring can also play an important role as an early warning of the effects of global warming. Analysis of C isotopes, for example, can help to explain what happens to the man-made GHGs in the atmosphere while N and S isotopes can reveal the connections between industrially produced oxides and acid rain. The  $^{18}\text{O}$  and D



**FIGURE 4.** Isotopic composition of C, O and H pools in terrestrial ecosystems. The values are approximations and will vary considerably with geographical location and environmental conditions. The actual data in the figure are from Israel (Ghosh and Brand, 2003).

isotopes in water are also very useful indicators of climate-related parameters such as surface air temperature, relative humidity and amount of precipitation.

Ecosystem C budgets are controlled by the balance between C uptake during photosynthesis and C loss during respiration (Prentice, 2001; Malhi, 2002; Kodama *et al.*, 2008; Brugnoli *et al.*, 2010). Within an ecosystem, both photosynthesis and respiration occur in a range of different species and functional groups, so the environmental control of C exchange processes is quite different in these distinct ecosystem components. The study of individual components of the ecosystem C budget and their environmental control would improve our understanding of ecosystem function and the system's potential response to climate variation. The measurement and analysis of C isotope ratios in leaf and atmospheric CO<sub>2</sub> samples can provide information that integrates important plant physiological characteristics spatially and temporally. Detailed mechanistic models have been developed that successfully explain the isotope effects occurring during photosynthetic gas exchange at the leaf level, making it is possible to interpret variation in C isotope discrimination resulting from differences in plant photosynthetic pathway and environmental conditions. For example, important physiological characteristics such as water use efficiency, stomatal limitation of photosynthesis, optimal stomatal behaviour and leaf N use efficiency are related directly to the value of leaf intercellular and ambient CO<sub>2</sub> ratios. Variation in environmental conditions causes change in the ratio of photosynthesis to stomatal conductance, and associated changes in leaf isotopic signatures ( $\delta^{13}\text{C}$ ) have also been documented. Using measurement of  $\delta^{13}\text{C}$  of leaf tissue and CO<sub>2</sub> released by respiration, Ometto *et al.* (2002) found that converting forest to pasture causes significant changes to ecosystem C isotope discrimination (Figure 5).



**FIGURE 5. Comparison of the average carbon isotope composition of total ecosystem respiration measured in forest and pasture sites at three locations in Brazilian Amazon Basin (Ometto *et al.*, 2002).**

Cernusak, Farquhar and Pate (2005) used  $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$  measurements to study post-photosynthetic variations of *Eucalyptus globulus* Labill. This approach is related quantitatively to plant photosynthetic performance, e.g. leaf physiological responses to environmental changes. Kodama *et al.* (2008) found a strong 24-h periodicity in  $\delta^{13}\text{C}$  of organic matter in leaf and twig phloem sap which was strongly dampened as carbohydrates were transported down the trunk. Periodicity reappeared in the  $\delta^{13}\text{C}$  of trunk-respired  $\text{CO}_2$ , which seemed to originate from respiratory fractionation rather than from changes in  $\delta^{13}\text{C}$  of the organic substrate. Ekblad and Hogberg (2001) found that the natural abundance of  $^{13}\text{C}$  in  $\text{CO}_2$  respired from forest soils demonstrated the speed of the link between tree photosynthesis and root respiration. It took 1–4 d for the C from canopy photosynthesis of 20–25 m trees to become available for root/rhizosphere respiration. Coletta *et al.* (2009) reported that C and N biogeochemical cycles in savannas are strongly regulated by the seasonal distribution of precipitation and pulses of nutrients released during the wetting of the dry soil, and are critical to the dynamics of microorganisms and vegetation. The  $\delta^{13}\text{C}$  values showed a consistent relationship with canopy height, revealing the importance canopy structure in determining the C isotopic signature of the vegetation. Carbon isotopic variations associated with the length of the dry season indicated the importance of recently fixed C to the integrated isotopic signature of the leaf organic C. Variations in foliar C and N isotope ratios were consistent with highly diverse vegetation with high energy availability but low availability of water and N (Matinelli *et al.*, 2007).

Isotope tracer techniques also show promise for quantifying the impacts of urban processes on the isotopic composition of the atmosphere and the partitioning the urban  $\text{CO}_2$  sources into their component parts (Pataki, Bowling and Ehleringer, 2003). Further, the isotope ratios of well-mixed environmental reservoirs such as the  $\delta^2\text{H}$  and  $\delta^{18}\text{O}$  of water and the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of dissolved compounds in stream discharge from watersheds and in the tissues of organisms, represent an integration of source inputs that extends over large spatial scales and the processing (mixing, losses, biogeochemical transformations) of elements within ecosystems. Therefore employing the approach in different compounds provides a unique means to investigate the coupling of water and C fluxes at various temporal and spatial scales.

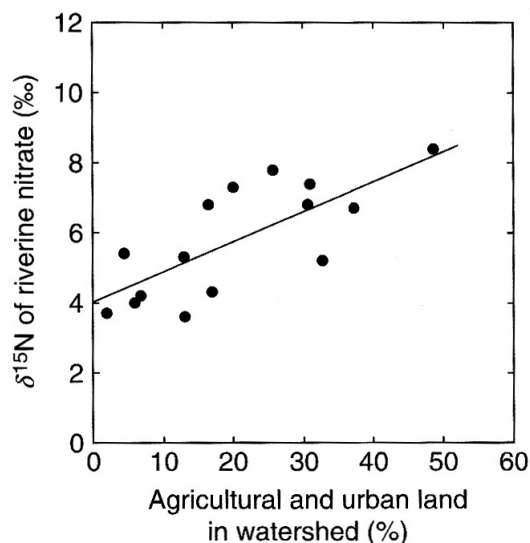
Carbon isotopes ( $^{12}\text{C}$ ,  $^{13}\text{C}$  and  $^{14}\text{C}$ ) may hold a key to determining the source of the increased C in the atmosphere by distinguishing the C cycles from deforestation, oceanic and fossil fuel. Plants prefer

$^{12}\text{C}$  to  $^{13}\text{C}$  and therefore photosynthetic  $\text{CO}_2$  is much lower in  $^{13}\text{C}$  than in the  $\text{CO}_2$  that comes from other sources (e.g. animal respiration). Declining  $^{14}\text{C}:$  $^{12}\text{C}$  and  $^{13}\text{C}:$  $^{12}\text{C}$  ratios parallel the reported increase of atmospheric  $\text{CO}_2$  and which are linked to the fact that fossil fuels, forests and soil C come from photosynthetic C which is low in  $^{13}\text{C}$ , while increased  $\text{CO}_2$  due to warming of the oceans would not be followed by reductions in the ratios of  $^{13}\text{C}$  and  $^{14}\text{C}$  to  $^{12}\text{C}$ . There are other clues that suggest the source of increased  $\text{CO}_2$  is not related to the warming of and subsequent release of  $\text{CO}_2$  from the ocean. For example, there has been a decline in the oxygen concentration of the atmosphere; therefore if ocean warming was responsible for the  $\text{CO}_2$  increase, an increase in atmospheric  $\text{O}_2$  should also be observed because  $\text{O}_2$  is also released as the water is warmed. The ocean is a sink for atmospheric C, and the C content of the oceans has increased by  $118 \pm 19 \text{ Pg C}$  in the last 200 yr. If atmospheric  $\text{CO}_2$  was the result of oceans releasing  $\text{CO}_2$  to the atmosphere, the  $\text{CO}_2$  in the ocean should not be rising as a result of ocean warming.

Global climate change will alter water availability in many ecosystems worldwide with marked impacts on biogeochemical cycles, as water represents one of the key factors constraining ecosystem productivity. Thus, a mechanistic understanding of the linkage between C and water cycles within the soil–plant–atmosphere continuum is needed to identify past and future climate and land-use change effects on ecosystem functioning (Heimann and Reichstein, 2008; Werner *et al.*, 2011).

Tritium is especially useful in studying the dynamics of water movement in different compartments of the hydrosphere, both on the local and global scales, while the heavy stable isotopes of deuterium and  $^{18}\text{O}$  can provide information about steady-state characteristics of the water cycle. Isotope data gathered from isotope monitoring of river water in main channels and in floodplains is expected to be very helpful for determining surface water routing, particularly to determine the proportions of water stored in the main channel and floodplains and to derive rates of transfer between them at various seasons and in each segment of the river valley. Preliminary isotope analyses suggest that up to 30 percent of water in the main channel is derived from water which has passed through the floodplain. Environmental isotope data also yield site-specific information about water transport and storage in the unsaturated zone. Numerous studies have shown that tritium is a very powerful tracer of  $\text{H}_2\text{O}$  movement in the unsaturated zone. In a study carried out in Brazil, it was possible to evaluate the average infiltration velocity and evapotranspiration flux for both the undisturbed forest and cleared area in the region. However, while isotope tracing provides information about the dynamics of  $\text{H}_2\text{O}$  movement in the unsaturated zone, the storage of water in the soil is commonly determined using neutron gauges (Nguyen *et al.*, 2011).

Sediment (and associated nutrients and chemicals) play important roles in degrading water quality and causing a range of other environmental problems. Sediment transport is the key to understanding the movement and fate of many nutrients (e.g. N and P), and of contaminants and organic C mobilization within the watershed (Figure 6). Soil erosion and associated deposition can cause redistribution in a differential manner of both soil particles and soil organic matter (SOM) along the landscape, and ultimately may result in C losses from the watershed as emissions of  $\text{CO}_2$  and  $\text{CH}_4$  and/or deposition/burial in sediment sinks and neighbouring aquatic systems. Fallout radionuclides (FRNs) such as caesium-137 ( $^{137}\text{Cs}$ ), lead-210 ( $^{210}\text{Pb}$ ) and beryllium-7 ( $^7\text{Be}$ ), naturally occurring radionuclides (NOR) such as potassium-40 ( $^{40}\text{K}$ ), radon-226 ( $^{226}\text{Ra}$ ) and thallium-232 ( $^{232}\text{Th}$ ), compound-specific stable isotope (CSSI) and conventional (modelling) techniques can be used to measure actual rates of soil erosion



**FIGURE 6.** Mean N isotope ratio of nitrate in rivers draining 16 watersheds (Williams *et al.*, 2007).

and sedimentation. Analysis of  $^{210}\text{Pb}$  and  $^{137}\text{Cs}$  stratigraphies in sediment cores has been used to determine the histories of sedimentation in floodplain lakes in Brazil (IAEA, 1990; Nguyen *et al.*, 2011).

The history and pathway of water in different parts of the hydrological cycle can be followed by the abundance of the stable heavy isotopes of  $^2\text{H}$  and  $^{18}\text{O}$ . In this way, water in different environments develops isotopic “fingerprints” with which it can be identified and its origins traced. Consequently, isotopic technique can determine the origins and ages of different water bodies; provide an estimate of the degree of mixing; determine the location and proportion of water recharge; measure recharge and indicate the velocity of groundwater flow. Recharge of groundwater is one critical aspect in water resource management, and isotopes can help determine both the area and the rate of recharge. The area can be identified by measuring  $^2\text{H}$  and  $^{18}\text{O}$  concentrations and correlating them with the altitude at which precipitation could have infiltrated the ground. The rate can also be measured by tracing levels of radioactive tritium in soil at various depths. The tritium peak method has been applied all over the world and in many different climates. In many instances, the tritium “peak” can be found at considerable depths, which indicates the distance travelled by the moisture since being deposited as tritium fallout in 1963. In moist climates where infiltration is high, artificial tritium can be injected as a tracer to determine the rate of recharge. Profiles of either environmental or artificial tritium can also give a measure of the movement of pollutants such as nitrates and pesticides from agriculture.

Polluted groundwater may remain in aquifers for centuries, even millennia, and is very difficult if not impossible to clean up. Isotope techniques can assess the vulnerability of groundwater to pollution from the surface by determining how rapidly it moves and where it is being recharged. Surface sources of pollution can then be determined, e.g. natural, industrial, agricultural or domestic. Isotope techniques can also identify incipient pollution, providing an early warning when the chemical or biological indicators do not give cause for concern (Williams *et al.*, 2007).

New instruments for sustainable use and conservation of peat forest such as REDD need more in-depth knowledge of C stocks and flows of peat land systems. Full ecosystem C balance data are needed to determine to what extent C accumulation still occurs in

peatland ecosystems and to what extent peat formation is limited under oil plantations. The full ecosystem C balance for different land use types such as undrained peat forest and disturbed forest and agricultural ecosystems can be estimated quite precisely with eddy covariance measurements above the forest canopy (incorporation of C fluxes of both respiration and decomposition). Soil  $\text{CO}_2$  fluxes are generally measured using the closed chamber method, while the eddy covariance tower can measure incoming and outgoing gas fluxes above the forest canopy and give more reliable estimation of ecosystem production and C dynamics. Leaching can be estimated using surface water samples from the catchment area and extracting the organic material.

Growing international concern over the adverse impacts of climate change and associated environmental stresses on sustainable development and poverty has ensured that climate change policy issues remain a central focus of most large funding agencies. Measurement of natural stable isotope ratios and the availability of large-scale isotope networks (Hemming *et al.*, 2007) are well placed to provide some of the key information required by policy makers, particularly in the field of C and water cycle management. It is therefore recommended that international agencies assist Indonesia to be a better conservator of its forest and peatland resources, either through forest conservation or improved soil and water management.

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# Effect of Elevated Carbon Dioxide on Nitrogen Dynamics and Greenhouse Gas Emissions in Grain Crop and Legume Pasture Systems: FACE Experiments and a Meta-Analysis

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## ABSTRACT

By 2070, atmospheric carbon dioxide concentration [ $\text{CO}_2$ ] is expected to be double that observed in 1950. In this higher [ $\text{CO}_2$ ] world, the sustainability of global crop production may be in jeopardy unless current nitrogen (N) management strategies are changed because of potential interactions between elevated atmospheric [ $\text{CO}_2$ ] and soil N dynamics. However, these interactions are poorly understood especially in semi-arid cropping systems. In this paper, experimental results are presented on the effects of elevated [ $\text{CO}_2$ ] on crop N demand, fertilizer N recovery, symbiotic  $\text{N}_2$  fixation and greenhouse gas emissions from cropping systems in southern Australia and northern China using free-air  $\text{CO}_2$  enrichment (FACE) facilities. Also discussed are the findings of a meta-analytic review of current literature which estimated quantitatively the effects of elevated [ $\text{CO}_2$ ] on soil N dynamics in grain crop and legume pasture systems. Results of experiments reported here and the meta-analysis suggest that under future elevated  $\text{CO}_2$  atmospheres (i) there will be an increase in crop demand for N, (ii) higher fertilizer N application rates and greater use of legume intercropping using locally appropriate agricultural management practices to meet the additional crop N demand, and (iii) increases in the terrestrial C sink may be less than expected since there will be a significant increase in greenhouse gas emissions ( $\text{CO}_2$ -equivalent) associated with an increase in atmospheric [ $\text{CO}_2$ ].

**Key words:** *elevated [ $\text{CO}_2$ ], fertilizer N recovery, grain N removal, meta-analysis, greenhouse gas emissions.*

## INTRODUCTION

Atmospheric carbon dioxide concentration ([ $\text{CO}_2$ ]) has increased from 280  $\mu\text{mol/mol}$  at the beginning of the Industrial Revolution to the current level of 397  $\mu\text{mol/mol}$  (NOAA, 2013). If  $\text{CO}_2$  emissions continue to rise at their present rate, [ $\text{CO}_2$ ] is estimated to reach about 550  $\mu\text{mol/mol}$  by 2050 and 700  $\mu\text{mol/mol}$  by the end of this century (IPCC, 2007). Elevated [ $\text{CO}_2$ ] can, in turn, affect agricultural greenhouse gas emissions via changes to carbon (C) and nitrogen (N) cycles in the plant–soil system (van Groenigen *et al.*, 2006). Agriculture accounted for 16 percent of net national emissions from Australia in 2009 (Department of Climate Change and Energy Efficiency, 2011), and 15 percent from China in 2005 (Wang, Huang and Rozelle, 2010). Elevated [ $\text{CO}_2$ ] has been estimated to increase the emission of nitrous oxide ( $\text{N}_2\text{O}$ ) and methane ( $\text{CH}_4$ ) from terrestrial ecosystems by a total of 1.12 Pg  $\text{CO}_2$  equivalent/yr due to enhanced C substrate availability (Hungate *et al.*, 1997; Baggs *et al.*, 2003; Inubushi, Cheng and Aonuma, 2003) and/or improved soil moisture (Leakey *et al.*, 2009), thereby offsetting 17 percent of the predicted increase in the entire terrestrial C sink (van Groenigen *et al.*, 2011).

Increasing [ $\text{CO}_2$ ] reduces stomatal conductance and transpiration and improves water use efficiency; it also stimulates photosynthetic processes, often resulting in increased crop growth and yield (Kimball, 1983; Drake *et al.*, 1997; Ainsworth and Long, 2005). When  $\text{C}_3$  plants are grown in an elevated [ $\text{CO}_2$ ], total N uptake and N removal in grain generally increase (Kimball, Kobayashi and Bindi, 2002). This increase in crop demand for N would be expected to gradually reduce soil N reserves unless replenished.

Depletion of soil N in agroecosystems can be compensated for by N fertilizer application although recovery by crops rarely exceeds 40 percent under ambient [ $\text{CO}_2$ ] (Chen *et al.*, 2008). Studies of the effect of elevated [ $\text{CO}_2$ ] on fertilizer N recovery in crops have generally been inconclusive and contradictory, showing positive (Martín-Olmedo, Rees and Grace, 2002; Weerakoon, Ingram and Moss, 2005), or neutral effects (Torbert *et al.*, 2004; Kim *et al.*, 2011). Symbiotically fixed  $\text{N}_2$  derived from crop and pasture legumes provides an alternative to additional fertilizer N use (Unkovich, Pate and Sanford, 1997; Chalk, 1998). Elevated [ $\text{CO}_2$ ] generally increases overall biomass and the amount of symbiotically fixed  $\text{N}_2$  due to increases in nodule size, nodule number per plant or, less likely, specific nitrogenase activity (Rogers *et al.*, 2009).

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This paper examines the effect of elevated  $[\text{CO}_2]$  on the fluxes of  $\text{N}_2\text{O}$ ,  $\text{CH}_4$  and  $\text{CO}_2$  from wheat-based cropping systems under semi-arid environments, such as in southern Australia and northern China and its interaction with agricultural practice, including the utilization of applied N fertilizer and symbiotic  $\text{N}_2$  fixation. It summarizes published data using free-air  $\text{CO}_2$  enrichment (FACE) facilities and extends the work with a meta-analysis of 108 published experiments examining N dynamics under elevated  $[\text{CO}_2]$ . This information provides new insight into the efficient management of fertilizer N and thus the sustainability of crop production under future high  $[\text{CO}_2]$ .

## MATERIALS AND METHODS

The field experiments were conducted at Horsham ( $36^\circ 45' \text{ S}$ ,  $142^\circ 07' \text{ E}$ ), Victoria, Australia from June to December in 2008 and 2009, and at Changping ( $40^\circ 10' \text{ N}$ ,  $116^\circ 14' \text{ E}$ ), Beijing, China from October 2008 to October 2009, using free-air  $\text{CO}_2$  enrichment (FACE) facilities. The targeted elevated  $[\text{CO}_2]$  was  $550 \mu\text{mol/mol}$  for both sites. Detailed methodology has been described elsewhere for the experiments on fertilizer  $^{15}\text{N}$  recovery (Lam *et al.*, 2012a, 2012c), symbiotic  $\text{N}_2$  fixation (Lam *et al.*, 2012d), greenhouse gas emissions (Lam *et al.*, 2011, 2013) and the meta-analysis (Lam *et al.*, 2012b). Briefly, to determine the recovery of fertilizer N in the wheat–soil system, nitrogen-15 ( $^{15}\text{N}$ )-labelled (10.22 atom %) granular urea was applied (at a rate according to local practice) to field micro-plots. Symbiotic  $\text{N}_2$  fixation in soybean was assessed using the  $^{15}\text{N}$  natural abundance method. The total C, total N and delta ( $\delta$ )  $^{15}\text{N}$  of plant and soil samples were analysed by isotope ratio mass spectrometry. The fluxes of  $\text{N}_2\text{O}$ ,  $\text{CO}_2$  and  $\text{CH}_4$  were measured by closed static chambers at various key growth stages of a wheat crop. The gas samples were analysed by gas chromatography.

Meta-analysis was conducted based on the natural log of the response ratio ( $r$  = response at elevated  $[\text{CO}_2]$ :response at ambient  $[\text{CO}_2]$ ) as a metric for analyses (Rosenberg, Adams and Gurevitch, 2000). Mean effect sizes and 95 percent confidence intervals were generated by bootstrapping (4 999 iterations) (Adams, Gurevitch and Rosenberg, 1997) using MetaWin 2.1 (Rosenberg, Adams and Gurevitch, 2000).

## RESULTS

### Fertilizer N recovery

The recovery of fertilizer N by the wheat crop or in the soil was not affected by elevated  $[\text{CO}_2]$  in the FACE experiments in Australia and China. The  $[\text{CO}_2]$ -induced increase in plant N uptake (18–44 percent) was satisfied mostly by increased uptake of indigenous N (19–50 percent) at both sites (Tables 1 and 2). Irrespective of  $[\text{CO}_2]$ , fertilizer N recovery by wheat grown under FACE was stimulated (13–609 percent) by supplementary irrigation (higher rainfall scenario) in Horsham (Lam *et al.*, 2012a), but reduced (47 percent) by high N application in Changping (Lam *et al.*, 2012c).

### Symbiotic nitrogen fixation

Under FACE conditions in Changping, elevated  $[\text{CO}_2]$  increased both the proportion (from 59 to 79 percent) and the amount (from 166 to  $275 \text{ kg}\cdot\text{N}\cdot\text{ha}^{-1}$ ) of shoot N derived from the atmosphere (Ndfa) by soybean cultivar Zhonghuang 13, but had no significant effect on either parameter for the other cultivar Zhonghuang 35 (Figure 1).

### Greenhouse gas emissions

Elevated  $[\text{CO}_2]$  increased the emission of  $\text{N}_2\text{O}$  (108 percent),  $\text{CO}_2$  (29 percent) and  $\text{CH}_4$  (from  $-0.14$  to  $3.45 \mu\text{g}\cdot\text{C}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ ) from soil in Horsham (Table 3), with changes being greater during the wheat vegetative stage than later in the growing season. Supplementary irrigation (higher rainfall scenario) reduced  $\text{N}_2\text{O}$  emission by 36 percent when averaged across  $[\text{CO}_2]$  treatments. Supplementary irrigation increased  $\text{CO}_2$  flux by 26 percent at ambient  $[\text{CO}_2]$ , but not at elevated  $[\text{CO}_2]$ , and had no impact on  $\text{CH}_4$  flux (Table 3). At the Changping site, elevated  $[\text{CO}_2]$  increased  $\text{N}_2\text{O}$  (60 percent) and  $\text{CO}_2$  (15 percent) emission, but had no significant effect on  $\text{CH}_4$  flux (Table 4).

### Meta-analysis

Elevated  $[\text{CO}_2]$  increased grain N removal (17 percent) of  $\text{C}_3$  non-legumes, legumes and  $\text{C}_4$  crops. This increase resulted from an overall increase (27 percent) in grain yield but a reduction (8 percent) in grain N concentration (Figure 2). The C:N ratio of residues from  $\text{C}_3$  non-legumes and legumes increased under elevated  $[\text{CO}_2]$  by 16 percent and 8 percent, respectively, but the increase in residue C:N ratio

**Table 1. The effect of elevated  $[\text{CO}_2]$  on the plant total N (kg/ha) derived from fertilizer N and soil N in Horsham, Australia, under different irrigation regimes (adapted from Lam *et al.*, 2012a)**

|                                        | Ndff   | Ndfs  | Total N | Ndff   | Ndfs  | Total N | Ndff   | Ndfs  | Total N |
|----------------------------------------|--------|-------|---------|--------|-------|---------|--------|-------|---------|
|                                        | 2008NS |       |         | 2008LS |       |         | 2009NS |       |         |
| Rainfed                                |        |       |         |        |       |         |        |       |         |
| Ambient [CO <sub>2</sub> ]             | 23.0   | 149.5 | 172.5   | 2.0    | 67.8  | 69.8    | 19.2   | 103.1 | 122.3   |
| Elevated [CO <sub>2</sub> ]            | 23.4   | 179.8 | 203.2   | 2.1    | 72.3  | 74.4    | 21.1   | 154.7 | 175.8   |
| Supplementary irrigated                |        |       |         |        |       |         |        |       |         |
| Ambient [CO <sub>2</sub> ]             | --     | --    | --      | 12.6   | 111.0 | 123.6   | 23.7   | 194.3 | 218.0   |
| Elevated [CO <sub>2</sub> ]            | --     | --    | --      | 16.5   | 129.1 | 145.6   | 22.1   | 195.6 | 217.7   |
| [CO <sub>2</sub> ]                     | ns     | *     | *       | ns     | ns    | ns      | ns     | 0.08  | 0.09    |
| Irrigation regime                      | --     | --    | --      | ***    | ***   | ***     | 0.06   | ***   | ***     |
| [CO <sub>2</sub> ] × Irrigation regime | --     | --    | --      | ns     | ns    | ns      | ns     | 0.09  | 0.09    |

Ndff — N derived from fertilizer; Ndfs — N derived from soil. 2008NS, 2008 normal sowing time; 2008LS, 2008 late sowing time; and 2009NS, 2009 normal sowing time

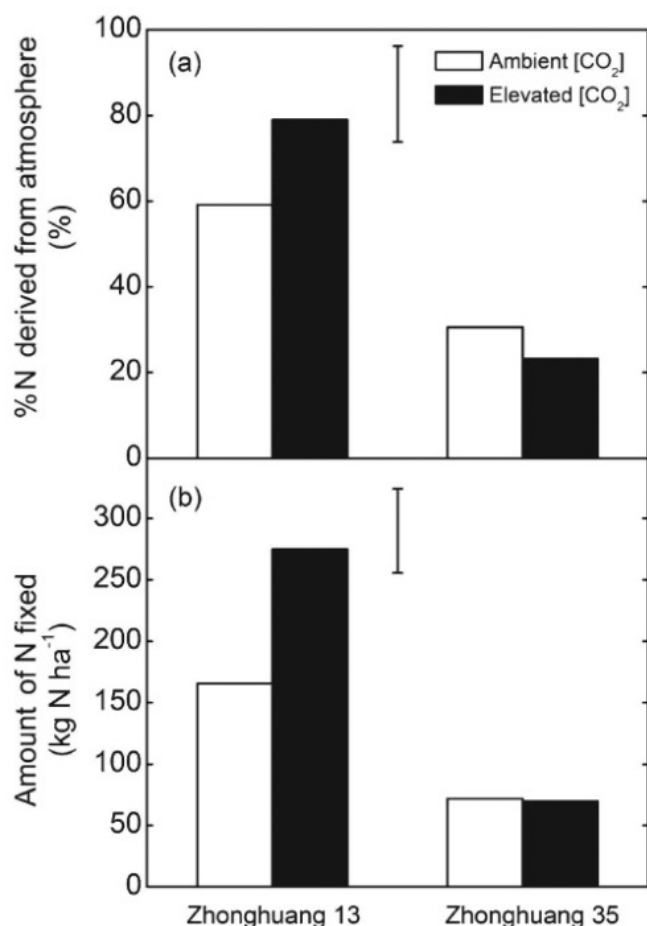
Significant effects are indicated as \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$ ; ns — not significant

**TABLE 2. The effect of elevated  $[\text{CO}_2]$  and N input on the plant total N (kg/ha) derived from fertilizer N and soil N at stem elongation in Changping, China (adapted from Lam *et al.*, 2012c)**

|                                          | Ndff | Ndfs  | Total N |
|------------------------------------------|------|-------|---------|
| Low N input (25 kg-N-ha <sup>-1</sup> )  |      |       |         |
| Ambient $[\text{CO}_2]$                  | 3.65 | 181.6 | 185.3   |
| Elevated $[\text{CO}_2]$                 | 3.88 | 187.4 | 191.3   |
| High N input (95 kg-N-ha <sup>-1</sup> ) |      |       |         |
| Ambient $[\text{CO}_2]$                  | 7.86 | 148.2 | 156.1   |
| Elevated $[\text{CO}_2]$                 | 8.06 | 168.0 | 176.0   |
| $[\text{CO}_2]$                          | ns   | 0.08  | 0.09    |
| N input                                  | *    | ns    | ns      |
| $[\text{CO}_2] \times \text{N input}$    | ns   | ns    | ns      |

Ndff = N derived from fertilizer; Ndfs = N derived from soil

Significant effects are indicated as \*  $p < 0.05$ ; ns = not significant



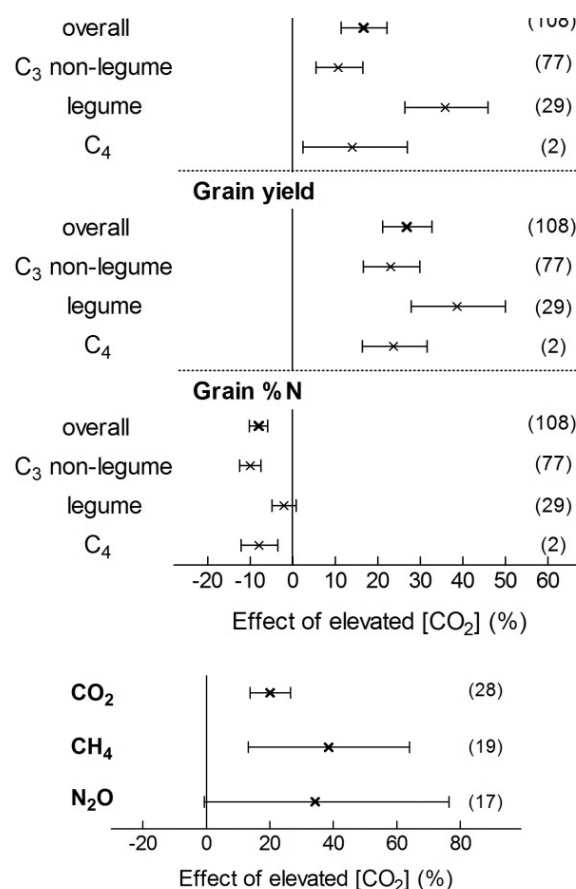
**FIGURE 1. Effect of elevated  $[\text{CO}_2]$  on (a) %Ndfa and, (b) amount of  $\text{N}_2$  fixed in the above-ground parts of two soybean cultivars (adapted from Lam *et al.*, 2012d). Each data point represents the mean of three replicates. Vertical bar indicates least significant difference (LSD) ( $p = 0.05$ ).**

of  $\text{C}_4$  crops (9 percent) was not significant. Elevated  $[\text{CO}_2]$  increased the recovery of fertilizer N in crops by 17 percent (Lam *et al.*, 2012b). Under elevated  $[\text{CO}_2]$ , there was a 38 percent increase in the amount of  $\text{N}_2$  fixed from the atmosphere, which was accompanied by greater whole plant nodule number (33 percent), nodule mass (39 percent), nitrogenase activity (37 percent) and %N derived from the atmosphere (10 percent; non-significant) (Lam *et al.*, 2012b). Elevated  $[\text{CO}_2]$  increased the emissions of  $\text{CO}_2$  (19 percent),  $\text{CH}_4$  from rice paddies (38 percent) and  $\text{N}_2\text{O}$  (27 percent) (Figure 3).

## DISCUSSION

### The need for N replenishment in cropping systems under elevated $[\text{CO}_2]$

The meta-analysis of current literature and FACE experiments indicate that elevated  $[\text{CO}_2]$  generally increases plant N uptake and grain N removal from cropping systems. Furthermore, elevated  $[\text{CO}_2]$  resulted in greater production of crop residues with a higher C:N ratio (especially for  $\text{C}_3$  non-legumes), leading to increased N immobilization in the soil (Torbert *et al.*, 2000). These  $[\text{CO}_2]$ -induced changes suggest that extra N will be required for cropping systems to maintain soil N availability and sustain grain yield (Lam *et al.*, 2012b). This is especially important in dryland cropping systems where water availability will often limit the cycling of N. In these systems, the “ $\text{CO}_2$  fertilization effect” on crop growth and N demand will most often be realized when rainfall is between average and moderately below average



**FIGURE 2. Effect of elevated  $[\text{CO}_2]$  on (a) grain N removal, (b) grain yield and (c) grain %N (adapted and modified from Lam *et al.*, 2012b). Means and 95 percent confidence intervals are depicted. Numbers of experimental observations are in parentheses.**

**TABLE 3. Interaction between [CO<sub>2</sub>] and irrigation on the emission of N<sub>2</sub>O, CO<sub>2</sub> and CH<sub>4</sub> averaged across the experimental period in Horsham, Australia (adapted from Lam *et al.*, 2013)**

|                             | N <sub>2</sub> O (μg N·m <sup>-2</sup> ·h <sup>-1</sup> ) | CO <sub>2</sub> (mg·C·m <sup>-2</sup> ·h <sup>-1</sup> ) <sup>a</sup> | CH <sub>4</sub> (μg·C·m <sup>-2</sup> ·h <sup>-1</sup> ) |
|-----------------------------|-----------------------------------------------------------|-----------------------------------------------------------------------|----------------------------------------------------------|
| Ambient [CO <sub>2</sub> ]  |                                                           |                                                                       |                                                          |
| Rainfed                     | 27.7                                                      | 259.7                                                                 | -0.56                                                    |
| Supplementary irrigated     | 15.6                                                      | 327.6                                                                 | 0.29                                                     |
| Elevated [CO <sub>2</sub> ] |                                                           |                                                                       |                                                          |
| Rainfed                     | 53.3                                                      | 379.7                                                                 | 7.06                                                     |
| Supplementary irrigated     | 36.5                                                      | 378.7                                                                 | -0.24                                                    |
| [CO <sub>2</sub> ] (C)      | **                                                        | ***                                                                   | ns                                                       |
| Irrigation regime (I)       | †                                                         | †                                                                     | ns                                                       |
| C × I                       | ns                                                        | †                                                                     | ns                                                       |

<sup>a</sup> CO<sub>2</sub> fluxes included both plant and soil respiration as plants were inside the measurement chambers

Significant effects are indicated as † *p* < 0.1, \*\**p* < 0.01 and \*\*\**p* < 0.001; ns — not significant

**TABLE 4. Emission of N<sub>2</sub>O, CO<sub>2</sub> and CH<sub>4</sub> as affected by ambient and elevated [CO<sub>2</sub>], averaged across N application rates and irrigation events in Changping, China (adapted from Lam *et al.*, 2011)**

|                             | N <sub>2</sub> O (μg N·m <sup>-2</sup> ·h <sup>-1</sup> ) | CO <sub>2</sub> (mg·C·m <sup>-2</sup> ·h <sup>-1</sup> ) <sup>a</sup> | CH <sub>4</sub> (μg·C·m <sup>-2</sup> ·h <sup>-1</sup> ) |
|-----------------------------|-----------------------------------------------------------|-----------------------------------------------------------------------|----------------------------------------------------------|
| Ambient [CO <sub>2</sub> ]  | 24.0                                                      | 37.3                                                                  | -5.6                                                     |
| Elevated [CO <sub>2</sub> ] | 38.4                                                      | 42.8                                                                  | -4.2                                                     |
| [CO <sub>2</sub> ]          | *                                                         | *                                                                     | ns                                                       |

Significant effects are indicated as \* *p* < 0.05; ns — not significant

(Lam *et al.*, 2012a). While [CO<sub>2</sub>]-induced increase in N uptake is associated with a slightly greater increase in grain yield under these rainfall conditions, improved N management practice (higher N application rate or using enhanced efficiency fertilizers) will be required to satisfy the increased demand for N.

### Measures to optimize the benefits of N replenishment strategy under elevated [CO<sub>2</sub>]

Using the <sup>15</sup>N labelling technique, it was demonstrated that both higher rates of fertilizer N application and greater use of legume intercropping can compensate for the enhanced rate of grain N removal under future elevated CO<sub>2</sub> environments (Lam *et al.*, 2012b). There are several strategies to optimize the benefits of N replenishment under elevated [CO<sub>2</sub>]. Firstly, while irrigation increases plant uptake of fertilizer N (Lam *et al.*, 2012a), irrigation is unavailable in dryland cropping systems in southern Australia and many other semi-arid farming systems. Farmers will need to take even greater account of likely seasonal rainfall conditions when making fertilizer management decisions to enhance recovery, e.g. using split fertilizer applications (Chen *et al.*, 2008). Secondly, an optimum N application rate should be determined. Excessive N application reduces wheat growth (van Herwaarden *et al.*, 1998) and the recovery of fertilizer N in the plant regardless of [CO<sub>2</sub>] (Lam *et al.*, 2012c). This defeats the purpose of compensating for the additional N removed in grains under elevated [CO<sub>2</sub>], and is therefore not recommended. Thirdly, legume species, cultivars and rhizobacteria should be selected according to their ability to fix more N<sub>2</sub> under elevated [CO<sub>2</sub>] (Lam *et al.*, 2012d; Matsunami *et al.*, 2009). Nonetheless, [CO<sub>2</sub>]-induced increases in grain N removal by pulse legumes should also be considered (Lam *et al.*, 2012b).

### Agriculture's potential contribution to future climate change

As a result of additional C input to soil under elevated [CO<sub>2</sub>], there will be a positive relation between the elevation of [CO<sub>2</sub>] and emission of greenhouse gases of CO<sub>2</sub> and N<sub>2</sub>O from semi-arid cropping systems (Lam *et al.*, 2011, 2012b, 2013). This will partly negate the expected increase in global terrestrial C sinks expected under elevated [CO<sub>2</sub>] (van Groenigen *et al.*, 2011).

During the vegetative stage of crop growth there are rapid changes in soil C and N. Under elevated [CO<sub>2</sub>] the extent of stimulation of greenhouse gas emissions may be reduced if this growth stage is shortened, e.g. by future warmer temperatures increasing the rate of crop maturation, or through choice of crop. This stimulation of greenhouse gas emissions may also be lower in irrigated systems (Lam *et al.*, 2013) if water supply is sufficient to facilitate the reduction of N<sub>2</sub>O to N<sub>2</sub> (Weier *et al.*, 1993; Ciarlo *et al.*, 2008). This process is favoured by the higher C substrate availability under elevated [CO<sub>2</sub>] (Baggs *et al.*, 2003). The greater [CO<sub>2</sub>] effect on N<sub>2</sub>O emission under high N compared with low N input (Lam *et al.*, 2011) reaffirms that excessive application of N fertilizer should be avoided in semi-arid cropping systems in future elevated CO<sub>2</sub> environments.

### CONCLUSIONS

Elevated [CO<sub>2</sub>] reduced grain N concentration, but increased N demand and removal in grain cropping systems. Extra N will be required to maintain soil N availability (avoid gradual decline of soil N) and sustain crop yield. The extra N could come from increased rates of fertilizer N application, or greater use of legume intercropping and legume cover crops. Increases in agricultural greenhouse gas

emissions will negate part of the predicted increase in the terrestrial C sink.

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