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# Biochar, Compost and Biochar-Compost: Effects on Crop Performance, Soil Quality and Greenhouse Gas Emissions in Tropical Agricultural Soils

Thesis submitted by

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in February 2017

for the degree of Doctor of Philosophy

College of Science and Engineering

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# **Dedicated to:**

My late parents, Agegnehu Jenberu and Shekmitu Wassie, who worked hard throughout their lives to provide me with a better education.

Farming communities, soil scientists, environmentalists and others who care about land, soil health and the environment.

# **ELECTRONIC COPY**

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17 February 2017

Date

# DECLARATION

I declare that this thesis is my own work and has not been submitted in any form for another degree or diploma at any university or other institution of tertiary education. Information derived from the published or unpublished work of others has been acknowledged in the text and a list of references is given.

17 February 2017

Date

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| Chapter | Details of publication                | Contribution of each author               |
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| number  |                                       |   |
| 1       | Introduction                          | • Getachew Agegnehu: conducted the        |
|         | Agegnehu, G., and Bird, M.I. 2016.    | literature search, prepared the database, |
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|         | and crop performance: A review.       | review.                                   |
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| 2       | Agegnehu, G., Bird, M.I., Nelson,     | • Getachew Agegnehu: conceived the        |
|         | P., and Bass, A. 2015. The            | research idea, designed and conducted     |
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Agegnehu, G., Nelson, P.N., Bird, M.I., 2016. Crop yield, plant nutrient uptake and soil physicochemical properties under organic soil amendments and nitrogen fertilization on Nitisols. *Soil and Tillage Research* 160: 1-13.

5

6

Agegnehu, G., Nelson, P.N., Bird, M.I., 2016. The effects of biochar, compost and their mixture and nitrogen fertilizer on yield and nitrogen use efficiency of barley grown on a Nitisol in the highlands of Ethiopia. *Science of the Total Environment* 569-570: 869-879.

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- Getachew Agegnehu: conceived and designed the study under field condition, conducted the experiment, collected and analyzed the data, and wrote the manuscript.
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#### Abstract

*Background and aims*: Chapter 1 highlights the introduction and summarizes the literature search in the area of the study. Soil fertility depletion, declining agricultural productivity because of reduction of soil organic matter (SOM), nutrient imbalance, and climate change due to increased greenhouse gases (GHG) emissions, are major constraints in most tropical agricultural soils. The aims of my PhD study were to: 1) investigate and compare the effects of different soil fertility treatments in relation to biochar, compost, and their mixture on soil physicochemical properties and soil fertility; 2) determine the response of crops to biochar, compost, and their mixture on different soil types; 3) evaluate the effect of different soil fertility treatments in relation to organic amendments and nitrogen (N) fertilizer on nutrient uptake, N recovery, and use-efficiency of barley; 4) determine nutrient retention and leaching under alternative organic amendments; and 5) investigate the impact of biochar, compost, and their mixture with respect to GHG emissions and carbon sequestration.

*Methods*: The present research comprised three separate experiments, which were conducted under two agro-ecosystems and soil types in north Queensland, Australia, using maize and peanut, as well as in the central highlands of Ethiopia using barley as test crops. Both greenhouse and field experiments were carried out to test the hypothesis that application of biochar improves soil fertility, fertilizer use efficiency, plant growth, and productivity, particularly when combined with compost. Treatments comprised: untreated control; mineral fertilizer; willow biochar (WB), acacia biochar (AB); compost (Com); WB + Com, AB + Com, and co-composted biochar-compost (COMBI). Mineral fertilizer was applied uniformly to all treatments. The treatments were laid out using a randomized design, each with three replicates. In the Ethiopian experiment, factorial combinations of five organic amendments (control, B, Com, B + Com and COMBI) and five levels of N fertilizer were investigated in a split-plot design with three replicates, with organic amendments as main plots and N levels as sub-plots.

*Results*: Chapter 2 presents results from a greenhouse pot trial which indicated that application of compost with fertilizer significantly increased plant growth, soil nutrient status, and plant nutrient concentration, with shoot biomass decreasing in the order F + Com (4.0) > F + WB + Com (3.6) > F+ WB (3.3) > F + AB + Com (3.1) > F + AB (3.1) > F (2.9) > control (1.0), with the ratio of the value to that of the control given in brackets. The shoot and root biomass exhibited significant positive correlations with soil water content, plant nutrient concentration, and soil nutrient concentration after harvesting. The results of a principal component analysis (PCA) showed that the first component (PRIN1) provided a reasonable summary of the data, accounting for about 84% of the total variance. As the plants grew, compost and biochar additions significantly reduced leaching of nutrients.

Chapters 3 and 4 present field trial results showing that application of biochar, compost, and their mixture increased maize yield by 10 - 29% and peanut yield by 17 - 24%, compared to fertilizer alone, in trials conducted in the Atherton Tablelands, Australia. Biochar, compost, and their mixture significantly improved the availability of plant nutrients, which appeared critical in improving crop performance. Soil organic carbon (SOC), soil water content (SWC), nitrate and ammonium concentrations were significantly higher in biochar treated plots than fertilizer alone, implying that potential exposure of nitrate and ammonium to the soil microbial community was significantly lower in biochar and COMBI treated plots compared to the fertilizer only and compost treatments. Emissions of CO<sub>2</sub> were highest in the fertilizer treatment and lowest in the COMBI treatment. Emissions of N<sub>2</sub>O were the highest in the fertilizer treatment and all biochar amended plots reduced N<sub>2</sub>O emission compared to the control.

Chapters 5 and 6 present field trial results from two sites in Ethiopia showing that the application of organic amendments and N fertilizer all significantly improved soil fertility and barley yield. In Ethiopia, the feedstock for biochar production was acacia (*Acacia spp.*), and compost was prepared from a mix of farmyard manure (FYM) and plant materials. Organic amendment by N fertilizer interaction significantly improved barley grain yield, with yield increments of 60% and 54% due to Com + B and 69 kg N ha<sup>-1</sup> and Com + 92 kg N ha<sup>-1</sup> at Holetta and Robgebeva, respectively, compared to the highest N rate only (92 kg ha<sup>-1</sup>). Organic amendments significantly improved soil properties through increases in SWC, SOC, cation exchange capacity (CEC) and pH. Addition of B, Com and B + Com increased SOC and CEC by 23 - 27% and 20 - 24% at Holetta and 26-34% and 19-23% at Robgebeya, compared to their respective initial values. Soil pH increased from the initial value of 5.0 to 5.6 at Holetta and from 4.8 to 5.4 at Robgebeya at harvest, due to biochar soil amendment. The highest total N uptake was obtained from  $\text{Com} + \text{B} + 92 \text{ kg N} \text{ ha}^{-1}$  at Holetta (138 kg ha<sup>-1</sup>) and Com+ 92 kg N ha<sup>-1</sup> at Robgebeya (101 kg ha<sup>-1</sup>). Application of organic amendments and N fertilizer improved significantly the agronomic efficiency (yield increase per unit of N applied, AE), apparent recovery efficiency (increase in N uptake per unit of N applied, ARE), and physiological efficiency (yield increase per unit of N uptake, PE). Mean AE and ARE were highest at  $B + 23 \text{ kg N} \text{ ha}^{-1}$  at Holetta and at B + 23 and B + 46 kg N ha<sup>-1</sup> at Robgebeya. The PE ranged from 19 - 33 grain kg<sup>-1</sup> N uptake at Holetta and 29 - 48 kg grain kg<sup>-1</sup> N uptake at Robgebeya. The effects of organic amendments and N fertilizer on AE, ARE and PE were greater at Robgebeya than at Holetta. The enhancement of N use efficiency through application of organic amendments emphasizes the importance of balanced

crop nutrition, ensuring that barley crops are adequately supplied with N and other nutrients. Overall, the integration of both organic and inorganic amendments may optimize N uptake efficiency and reduce the amount of N fertilizer required for sustainable barley production in the long-term.

*Conclusions:* The results indicate that applications of biochar and compost either singly, or in combination, could be adopted as a sustainable agronomic strategy, since they have demonstrated a strong potential to improve SOC, soil nutrient status, SWC, crop yield and reduce GHG fluxes on tropical agricultural soils. Moreover, the integration of both organic and inorganic nutrient sources may optimize NUE and reduce the amount of fertilizer required for the sustainable production of barley in the long term. However, the amount by which conventional fertilizer could be reduced, and the resultant economic benefit because of addition of these amendments, need further study for longer-term economic and environmental sustainability.

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# Abbreviations

| AB               | Acacia biochar                                  |
|------------------|---|
| AE               | Agronomic efficiency                            |
| Al               | Aluminum  |
| ANRE             | Apparent nitrogen recovery efficiency           |
| ARL              | Analytical Research Laboratory                  |
| AusAID           | Australian Agency for International Development |
| В                | Biochar   |
| С                | Carbon  |
| Cmol(+)/kg       | Centimole per kilogram                          |
| CH <sub>4</sub>  | Methane   |
| C: N             | Carbon to nitrogen ratio                        |
| CFI              | Carbon farming initiative                       |
| $CO_2$           | Carbon dioxide                                  |
| Com              | Compost   |
| COMBI            | Co-composted biochar-compost                    |
| CSA              | Central Statistical Authority                   |
| EC               | Electric conductivity                           |
| ECEC             | Effective Cation Exchange Capacity              |
| FYM              | Farmyard manure                                 |
| GHGs             | Greenhouse gases                                |
| GLM              | General Linear Model                            |
| Gt               | Gigaton   |
| GNC              | Grain nitrogen concentration                    |
| GNU              | Grain nitrogen uptake                           |
| GPC              | Grain protein content                           |
| ha <sup>-1</sup> | Per hectare                                     |
| HARC             | Holetta Agricultural Research Center            |
| HI               | Harvest index                                   |
| $\delta^{13}C$   | Isotope carbon                                  |
| $\delta^{15}N$   | Isotope nitrogen                                |
| JCU              | James Cook University                           |
| Κ                | Potassium                                       |

| Kg                              | Kilogram                        |
|---------------------------------|---------------------------------|
| LWC                             | Leaf water content              |
| Mg                              | Magnesium                       |
| Ν                               | Nitrogen                        |
| $N_2O$                          | Nitrous oxide                   |
| NH4 <sup>+</sup> -N             | Ammonium nitrogen               |
| NHI                             | Nitrogen harvest index          |
| NO <sub>3</sub> <sup>-</sup> -N | Nitrate nitrogen                |
| NUE                             | Nitrogen use efficiency         |
| Р                               | Phosphorus                      |
| PCA                             | Peanut Company of Australia     |
| PE                              | Physiological efficiency        |
| Pg                              | Petagram                        |
| PRIN                            | Principal component analysis    |
| r                               | Pearson correlation coefficient |
| r <sup>2</sup>                  | Coefficient of determination    |
| SLW                             | Specific leaf weight            |
| SNC                             | Straw nitrogen concentration    |
| SNU                             | Straw nitrogen uptake           |
| SOC                             | Soil organic carbon             |
| SOM                             | Soil organic matter             |
| SWC                             | Soil water content              |
| t                               | Ton                             |
| TNU                             | Total nitrogen uptake           |
| WB                              | Willow biochar                  |
| Zn                              | Zinc                            |

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# **Overview of Chapter 1**

This chapter is based on a paper submitted to *Applied Soil Ecology* with a moderate change in content and formatting.

In this chapter I conducted a detailed and comprehensive review of published scientific research and review articles on biochar and biochar-compost. I completed a systematic search (title, keywords and abstract) in the global scientific databases, identify papers focused on the biochar and biochar-compost in relation to their effects on soil quality, plant growth and crop yield, and conduct in-depth review of those papers. Key findings from this study have been interpreted to identify knowledge and information gaps on biochar and biochar-compost research.<sup>1</sup>

<sup>&</sup>lt;sup>1</sup> Agegnehu, G., and Bird, M.I. 2016. The role of biochar and biochar-compost in improving soil quality and crop performance: A review. *Applied Soil Ecology* (accepted).

## **Chapter 1 Introduction**

#### 1.1. Background

Soil fertility depletion, declining agricultural productivity because of reduction of soil organic matter (SOM), nutrient imbalance, and climate change due to increased greenhouse gases emissions, are major constraints in most tropical agricultural soils (Lal, 2015; Pender, 2009; Sanchez, 2002). Globally, acidic soil occupies 30% of ice-free land (Von Uexküll and Mutert, 1995), and 20% of irrigated global land has been seriously affected by high salinity levels (Pitman and Läuchli, 2002). More than half of all African people are affected by land degradation, making this an urgent development issue for the continent. For example, an estimated US \$42 billion in income and 6 million ha of productive land are lost every year due to land degradation and declining agricultural productivity (Bationo et al., 2006). Moreover, Africa is saddled with a US \$9.3 billion annual cost associated with desertification. Research has shown that nutrient losses are only partially compensated by natural and synthetic inputs; thus, the nutrient balance across the whole of Sub Saharan Africa appears to be negative, by 26 kg N, 3 kg P, and 19 kg K ha<sup>-1</sup> yr<sup>-1</sup> (Drechsel et al., 2001). Many tropical soils are lacking in inorganic nutrients and rely on the recycling of nutrients from soil organic matter (SOM) to maintain fertility. An extremely nutrient-poor Amazonian soil, for example, showed no potential for agriculture beyond the three-year lifespan of the forest litter mat inputs, once biological nutrient cycles were interrupted by slash-burning (Tiessen et al., 1994).

In the 21<sup>st</sup> century, agriculture faces various challenges; i.e., it has to meet food and industrial needs of the growing population, while concurrently protecting the environment. In 2015, the world population was 7.35 billion, while projections show that population would reach 9.72 billion by 2050 (UN, 2015). Thus, world food production must increase by ~70% from its current level to satisfy food needs by 2050 (FAO, 2009). The majority of agricultural production depends on synthetic fertilizers, and one of the major problems is over-application of fertilizers, especially N fertilizers, such as urea. For instance, intensification of agricultural and human activities, such as the increased use of synthetic fertilizer (103 million ton of N worldwide in 2010), and inefficient use of N fertilizers are blamed for the increased N<sub>2</sub>O emissions of 17.7 T g of N per year to the atmosphere (IFA, 2010; Zaman et al., 2012). The total global demand for NPK fertilizer was 180 million tons in 2012, of which nitrogen fertilizer constituted 109.9 million tons (~61%). The world nitrogen fertilizer demand is expected to be around 116.0 million tons in 2016 at an annual growth rate of 1.3%. Of the overall increase in demand for 6 million tons of nitrogen, between 2012 and 2016, 60% would be in Asia, 19% in

America, 13% in Europe, 7% in Africa and 1% in Oceania (FAO, 2012). In monetary value, the annual loss in N fertilizer has been estimated to be \$18.8 billion with the 33% N recovery efficiency (Raun et al., 2002) and \$USD 255 per ton (World Bank, 2015).

The benefits of inorganic fertilizers, which have played a significant role in increasing agricultural production and productivity over the last half century, have been widely demonstrated since the green revolution. However, the productivity boost provided by fertilizer technology may be reaching a point of diminishing returns (Gruhn et al., 2000a; Rosset et al., 2000), with industrial fertilizer use becoming increasingly unsustainable, environmentally damaging, and unlikely to keep up with demand (Barrow, 2012). The increase in human population pressure has decreased the availability of arable land and it is no longer feasible to use extended fallow periods to restore soil fertility in the tropics (Bationo et al., 2007; Nandwa, 2001). Shrinking land area per capita and declining soil quality have led to a sustained increase in the rate of inorganic fertilizer application from year to year, to enhance or maintain agricultural productivity, where this is an option. However, the application of fertilizer alone is not a sustainable solution to improve soil fertility and maintain yield increases (Gruhn et al., 2000a); rather, it has been widely realized that application of excessive inorganic fertilizer, especially N, may cause soil deterioration and other environmental problems due to more rapid organic matter mineralization (Liu et al., 2010; Palm et al., 2001).

Soil degradation is the most serious biophysical constraint limiting agricultural productivity in many parts of the world, particularly in the tropics (Lal, 2015; Pender, 2009). Figure 1.1 shows the extent of soil global soil degradation, where soils are becoming degraded in many areas worldwide (UNEP, 2002). The long-term benefit of assigning more land to agriculture will not offset the negative environmental impacts of land degradation in the future (Tilman et al., 2002). Instead, a more promising approach to ensuring food security is to increase yield from currently cultivated land where productivity is low (Foley et al., 2011). Sustainable agricultural intensification, i.e., increasing productivity per unit land area, is thus necessary to secure the food supply for the increasing world population (Godfray et al., 2010; Tilman et al., 2011). In most tropical environments, sustainable agriculture faces significant constraints due to low nutrient status and rapid mineralization of SOM (Zech et al., 1997). Decline in SOM content leads to decreased cation exchange capacity (CEC), and under such circumstances, the efficiency of applied mineral fertilizers is low. In addition, most small-scale farmers cannot afford to apply fertilizers regularly due to high costs. Thus, nutrient deficiencies are prevalent in many crop production systems of the tropics and this constrains productivity (Glaser et al., 2002).

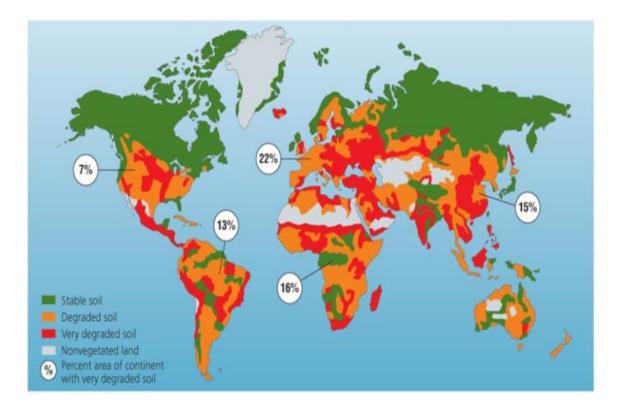


Figure 1.1. Soil degradation: A global concern. Source: UNEP (2002).

#### 1.2. Biochar and soil properties

Soil management actions can affect sustainable use of soil resources through their influence on soil quality, soil stability and soil resilience. Soil quality indicators are a composite set of measurable biophysical and chemical attributes which relate to functional soil processes and can be used to evaluate soil health status, as affected by management and climate change drivers (Allen et al., 2011; Dalal et al., 2011; McNeill and Penfold, 2009). According to Murphy (2015), soil properties include aggregate stability, bulk density, water-holding capacity, soil erodibility, soil color, soil strength, compaction characteristics, friability, nutrient cycling, CEC, soil acidity and buffering capacity, capacity to form ligands and complexes, salinity, and the interaction of SOM with soil biology. With reference to agricultural land use, soil quality refers to its capacity to sustain and support the growth of crops and animals while also maintaining or improving the quality of the environment (Lal, 2011b; Powlson et al., 2011). Soil quality is a strong determinant of agronomic yield. Soil stability refers to the susceptibility of soil to change under natural or anthropogenic perturbations, while soil resilience refers to soil's ability to restore its life support processes after being stressed (Lal, 2015; Nciizah and Wakindiki, 2015).

Although food and nutritional insecurity are global issues, the decline in the actual and potential productivity of soil is a major threat to agricultural sustainability and environmental quality, owing to inappropriate land-use (Doran and Zeiss, 2000; Lal, 2011b). The problem is particularly critical in developing countries due to demographic pressure and a shortage of prime agricultural land, water and other farm resources (Lal, 2015; Ray et al., 2015). The challenge is especially overwhelming due to the changing and uncertain climate (Lobell and Field, 2007) and the associated increase in the threat of soil degradation (Bai et al., 2008). The sustainability of agronomic practices and increases in production are, thus, essential to meeting the goals of sustainably increasing food supply. Soil organic carbon (SOC) is vital for sustainable yields as it is able to retain water and nutrients, provide a habitat for soil biota, and improve soil structure (Lal, 2009a; Lorenz et al., 2007). According to Lal (2004) soil is a major reservoir of soil C, where the global soil C pool of 2,500 Gt includes about 1,550 Gt of SOC and 950 Gt of soil inorganic C. The soil C pool is 3.3 times the size of the atmospheric pool (760 Gt) and 4.5 times the size of the biotic pool (560 Gt). Land use change and farming practices have led to a marked reduction in SOC, and with the increased temperatures expected with climate change, SOC stocks are likely to fall further, in turn reducing soil fertility and exacerbating climate change (Raich et al., 2002).

Maintaining an appropriate level of SOM, and ensuring the efficient biological cycling of nutrients, is crucial to the success of soil management and agricultural productivity strategies (Bationo et al., 2007; Diacono and Montemurro, 2010; Murphy, 2015; Vanlauwe et al., 2010). The decline in SOM contributes to several soil degradation processes including erosion, compaction, salinization, nutrient deficiency, loss of biodiversity, and desertification, all of which are accompanied by a reduction in soil fertility (Lal, 2015). Soils depleted of SOC not only reduce yield but also lead to low use efficiency of applied inputs, and are able to sequester less atmospheric CO<sub>2</sub> (Lehmann et al., 2006). Research has shown the application of the positive effects of mulches, composts and manures on soil fertility, agronomic use efficiency of nutrients, and crop productivity (Vanlauwe et al., Bationo et al., 2007; 2010). However, accelerated mineralization is a limitation to the practical application of organic fertilizers under tropical conditions, as only a small portion of the applied organic compounds are stabilized in the soil in the long term, with most released back to the atmosphere as carbon dioxide (CO<sub>2</sub>) (Fearnside, 2000). Thus, in addition to repeated application at high doses and the cost of application of organic materials, their rapid decomposition and mineralization may make a significant contribution to global warming (Barrow, 2012). Realizing such environmental and soil degradation problems, biochar research has progressed considerably with important key findings on agronomic benefits, carbon sequestration, greenhouse emissions, soil quality, soil acidity, soil fertility, soil

salinity, etc. (Lehmann et al., 2003; Van Zwieten et al., 2014), along with research into biocharcompost mixes and co-composted-biochar-compost as soil amendments (Schmidt et al., 2014; Schulz et al., 2013).

"Biochar is a solid material obtained from thermochemical conversion of biomass in an oxygenlimited environment" (IBI, 2014, p. 9). Biochar as a soil amendment has potential benefits in improving the quality of soils, including the physicochemical properties of degraded or nutrient depleted soils, thereby making more agricultural land available, while increasing crop yields, so that the need for expansion of agricultural land area decreases (Barrow, 2012; Luo et al., 2016). The application of biochar to infertile soils decreases soil bulk density, and increases total pore volume and water holding capacity (Abel et al., 2013; Andrenelli et al., 2016; Omondi et al., 2016). Soil with a high CEC has the ability to retain cations on the surface of biochar particles, humus, and clay, so nutrients are retained and more available for uptake by plants rather than leached below the root zone (Ding et al., 2016; Glaser et al., 2002; Laird et al., 2010). Other studies indicate significant changes in soil quality, including increases in pH, organic carbon, exchangeable cations, and N fertilizer use efficiency as well as a reduction in aluminum toxicity (Chan et al., 2007; Hewage, 2016; Lehmann et al., 2003; Steiner et al., 2007). Recent studies have also indicated that the simultaneous application of biochar and compost resulted in enhanced soil fertility and water holding capacity, crop yield, and C sequestration potential (Luo et al., 2016; Olmo et al., 2016; Plaza et al., 2016; Schulz and Glaser, 2012).

The pyrolysis conditions and biomass type affect the composition and structure of biochar (Crombie et al., 2013; Ippolito et al., 2015; Ronsse et al., 2013; Subedi et al., 2016), resulting in significant differences in the characteristics of the biochar that can be correlated with changes in nutrient content and retention (DeLuca et al., 2009; Granatstein et al., 2009). Biochars derived from manure and animal-product feedstock are relatively rich in nutrients compared with those derived from plant materials and especially those derived from wood (Alburquerque et al., 2014; Singh et al., 2010a). However, biochar in general may be more important for use as a soil amendment and driver of nutrient transformation than as a primary source of nutrients (DeLuca et al., 2009). Although both rapid and slow decomposition of biomass-derived biochars have been reported (Bird, 1999; Lehmann et al., 2006), biochar in soil could persist longer and retain cations better than other forms of SOM, such as mulches, crop residues, composts, and manures (Lone et al., 2015; Sohi et al., 2010; Tiessen, 1994)).

Biochar may improve the physical and chemical environment in soil due to its porous nature, high surface area, and ability to adsorb soluble organic matter and inorganic nutrients (Thies and Rillig,

2009). This provides microbes with a suitable habitat instead of supplying them with a primary source of nutrients (Gomez et al., 2014; Lehmann et al., 2011). Microbial communities in biochar can change over time once it has been added to the soil. Such attributes are beneficial for agriculture in nutrient cycling or mineralization of organic matter which may develop over time following biochar addition (Rousk et al., 2010; Wang et al., 2016). Biological nitrogen fixation (BNF) significantly decreases if available nitrate concentrations in soils are high, and if available Calcium (Ca), Phosphorus (P), and micronutrient concentrations are low (Giller, 2001). However, in soils with appreciable concentrations of biochar, available nitrate concentrations are usually low and available Ca, P, and micronutrient concentrations are high, which is ideal for maximum BNF (Lehmann et al., 2003). Studies have shown that combined biochar and fertilizer application increased microbial biomass compared to mineral fertilizer alone (Birk et al., 2009; Burger and Jackson, 2003; Ding et al., 2016). Microbial immobilization is an important mechanism to retain N in soils affected by leaching, where increased carbon availability stimulates microbial activity, resulting in greater N demand, promoting immobilization and recycling of nitrate (NO<sub>3</sub>) (Burger and Jackson, 2003). Application of biochar has also increased crop yield, soil microbial biomass, plant tissue potassium (K) concentration, soil P and K, total soil C and N (Biederman and Harpole, 2013; Galvez et al., 2012), nodulation, and biological N fixation by common beans (Rondon et al., 2007), red clover (Mia et al., 2014), soybean (Mete et al., 2015) and faba beans (Van Zwieten et al., 2015).

Nutrients are retained and remain available for plants in soils, mainly by adsorption mechanisms to minerals and organic matter. Biochar has a greater ability, than other SOM (e.g. compost and manure), to adsorb cations per unit C due to its greater surface area, greater negative surface charge, and greater charge density (Jaafar et al., 2015; Sombroek et al., 2003). The immediate beneficial effects of biochar applications for nutrient availability are largely due to the availability of higher K, P and zinc (Zn), and to a lesser extent, Ca and copper (Cu) (Lehmann et al., 2003; Steiner et al., 2007; Zhao et al., 2014a). The long-term benefits for nutrient availability include greater stabilization of organic matter, slower nutrient release from added organic matter, and improved retention of cations due to a greater CEC (Lehmann, 2007; Liang et al., 2006). Applications of biochar to soil have shown obvious increases in total SOC and N concentrations, the availability of major cations, and P (Biederman and Harpole, 2013; Steiner et al., 2007), improved soil CEC, and pH (Peng et al., 2011; Yuan and Xu, 2011). Leaching of nutrients from soils can deplete soil fertility, hasten soil acidification, increase cost of fertilizer for farmers, reduce yield of crops, and most notably threaten environmental health (Lehmann et al., 2003; Major et al., 2012). The application of biochar has been demonstrated to significantly reduce the leaching of nitrate (Haider et al., 2017), ammonium and phosphate (Yao et

al., 2012), N, P and magnesium (Mg) (Laird et al., 2010), and Ca and Mg (Major et al., 2012). Biochar also helps reduce the leaching of N into groundwater, while reducing the need for fertilizers that are the source of excess N (Glaser et al., 2015; Lehmann, 2007; Zhang et al., 2016). Table 1.1 shows a summary of the effects of biochar application on soil biophysical and chemical properties.

 Table 1.1. Summaries of the effects of biochar applications on soil biophysical and chemical properties.

| <b>Biochar source</b> | Soil type  | Effect on soil properties/soil quality         | References       |
|-----------------------|------------|--|------------------|
|                       |            | changes  |                  |
| Different             | Different  | Increase in soil pH, CEC, available K, Ca and  | (Glaser et al.,  |
| feedstock types       | soil types | Mg, total N and available P; decrease in       | 2002; Schulz and |
|                       |            | aluminum (Al) saturation of acid soils.        | Glaser, 2012)    |
| Wood charcoal         | Anthrosol  | Increase in soil C content, pH value and       | (Chan et al.,    |
|                       | and        | available P; reduction in leaching of applied  | 2007; Lehmann    |
|                       | Ferralsol  | fertilizer N, Ca and Mg and lower Al           | et al., 2003)    |
|                       |            | contents.                                      |                  |
| Eucalyptus            | Clay-      | Significant increase in total N derived from   | (Güereña et al., |
| logs, maize           | loam       | the atmosphere (NdfA) by up to 78% in          | 2012; 2015;      |
| stover                | Oxisol;    | common beans (Phaseolus vulgaris L.); high     | Rondon et al.,   |
|                       | silt loam  | recovery of applied N (39%) due to greater N   | 2007)            |
|                       |            | retention through lower gaseous or erosion N   |                  |
|                       |            | losses with biochar addition.                  |                  |
| Charcoal site         | Haplic     | Increase in total porosity from 46% to 51%     | (Oguntunde et    |
| Soil                  | Acrisols   | and saturated soil hydraulic conductivity by   | al., 2008)       |
|                       |            | 88% and reduction in bulk density by 9%.       |                  |
| Peanut hulls,         | Loamy      | Biochars produced at higher pyrolysis          | (Novak et al.,   |
| pecan shells,         | sand       | temperature increased soil pH, while biochar   | 2009b)           |
| poultry litter        |            | made from poultry litter increased available P |                  |
|                       |            | and sodium (Na).                               |                  |
| Wood and              | Different  | Increase in P availability from 163-208%, but  | (Madiba et al.,  |
| peanut shell          | soil types | decreased AMF abundances in soils from 43-     | 2016; Warnock et |
|                       |            | 77%.   | al., 2010)       |

| Wood and        | Different  | Increase the soil's saturated hydraulic                    | (Abel et al.,      |
|-----------------|------------|--|--------------------|
| manure-derived  | soil types | conductivity and plant's water accessibility,              | 2013; Ajayi et     |
| biochars        |            | as well as boost the soil's total N                        | al., 2016;         |
|                 |            | concentration and CEC, improving soil field                | Atkinson et al.,   |
|                 |            | capacity, and reduce NH <sub>4</sub> -N leaching on soils, | 2010; Lehmann      |
|                 |            | such as loamy sand soil and Ferralsols.                    | et al., 2003;      |
|                 |            |  | Stavi, 2012)       |
| Manure, corn    | Alfisol    | Tissue N concentration and uptake decreased                | (Rajkovich et al., |
| stover, woods,  |            | with increasing pyrolysis temperature and                  | 2012)              |
| food waste      |            | application rate, but increased K and Na                   |                    |
|                 |            | content.   |                    |
| Different       | Different  | Increased crop yield, improved microbial                   | (Biederman and     |
| biochar sources | soil types | habitat and soil microbial biomass, rhizobia               | Harpole, 2013;     |
|                 |            | nodulation, plant K tissue concentration, soil             | Ding et al., 2016; |
|                 |            | pH, soil P, soil K, total soil N, and total soil C         | Thies et al.,      |
|                 |            | compared with control conditions.                          | 2015)              |
| Peanut hull     | Ultisols   | Increased K, Ca, and Mg in the surface soil (0-            | (Gaskin et al.,    |
|                 |            | 15 cm). Increased K was reflected in the plant             | 2010)              |
|                 |            | tissue analysis.   |                    |

#### 1.3. Biochar and crop productivity

Biochar has been reported to both increase (Blackwell et al., 2015; Chan et al., 2007; Yamato et al., 2006) and decrease (Deenik et al., 2010) plant growth and crop yield. Crop growth and yield increases with biochar have, in most cases, been attributed to optimization of the availability of plant nutrients (Abiven et al., 2015; Gaskin et al., 2010; Lehmann et al., 2003), reduction of exchangeable  $Al^{3+}$  (Glaser et al., 2002; Qian et al., 2013), and changes in soil microbial dynamics (Lehmann et al., 2011; Wang et al., 2016). Thus, if pyrolysis for energy production is combined with biochar addition to soil, biochar can have a significant effect on retention of cations due to its longevity, enhancing agricultural productivity, drawing CO<sub>2</sub> from the atmosphere, and abating environmental pollution. However, biochar effects on plant growth and yield tend to be reduced or lacking in temperate regions (Borchard et al., 2014; Schmidt et al., 2014).

The effect of biochar on the productivity of crops depends on the rate of application. Progressive growth improvement at higher biochar application rates is seen with relatively low levels of biochar, with significant improvements ranging from 20 - 220% in productivity over the control at low rates of 0.4-8 t C ha<sup>-1</sup> (Lehmann and Rondon, 2006). However, negative responses of crops to biochar application can result from changes in soil properties and pH induced micronutrient deficiency (Xu et al., 2015). The study of Macdonald et al. (2014b) indicated that negative growth effect correlated with increasing electrical conductivity in the acidic Arenosol, while in the acidic Ferralsol a small rate-dependent increase in pH correlated with relatively large gains in biomass, possibly due to improved phosphorus nutrition and alleviated Al toxicity. This is probably more relevant and immediate than lower micro-nutrient availability decreases through immobilization by microbial biomass at high C:N ratios, although other growth-limiting factors may be responsible as well (Lehmann et al., 2003; Sigua et al., 2016). Table 1.2 shows the summary of responses of crops to different sources of biochar applications.

| <b>Biochar source and</b>            | Crop    | Reasons and responses of crops to biochar       | References      |
|--------------------------------------|---------|---|-----------------|
| application rate                     | type    | applications                                    |                 |
| Mango wood (0, 8,                    | Maize   | Increase in biomass from 30-43% and yield       | (Rajkovich et   |
| 16 t ha <sup>-1</sup> ), corn stover |         | by 22% due to improvements in soil pH, CEC,     | al., 2012;      |
| (2.6 - 91 t ha <sup>-1</sup> )       |         | nutrient availability and water retention.      | Rondon et al.,  |
|                                      |         |   | 2006)           |
| Acacia bark                          | Maize,  | Twofold increase in maize and peanut yields     | (Yamato et al., |
| (10 L m <sup>-2</sup> )              | peanut  | due to higher N and exchangeable bases and      | 2006)           |
|                                      |         | low Al.   |                 |
| Teak and rose wood                   | Rice    | Improved plant growth and 2-3 times yield       | (Asai et al.,   |
| biochars (4-16 t ha <sup>-1</sup> )  | and     | increment as biochar improved crop response     | 2009; Steiner   |
|                                      | sorghum | to NP fertilizer.                               | et al., 2007)   |
| Green waste, poultry                 | Radish  | Increase in yield (42 - 96%) due to improved    | (Chan et al.,   |
| litter (0, 10, 25, 50,               |         | soil physicochemical properties, N-             | 2007; 2008)     |
| 100 t ha <sup>-1</sup> )             |         | availability and use efficiency and decrease in |                 |
|                                      |         | exchangeable Al.                                |                 |

Table 1.2. Summaries of responses of crops to different sources of biochar applications.

| Macadamia nut shell                  | Maize,  | Biochar with high volatile matter (225 g kg <sup>-1</sup> ) | (Deenik et al.,  |
|--------------------------------------|---------|---|------------------|
| (0, 5, 10, 20%)                      | lettuce | decreased plant growth and soil $NH_4^+$ –N                 | 2010)            |
|                                      |         | compared to low-VM (63 g kg <sup><math>-1</math></sup> ).   |                  |
| Paper-mill biochar                   | Wheat,  | Increase in biomass by 250% attributable to                 | (Van Zwieten     |
| $(10 \text{ t ha}^{-1})$             | radish  | improved fertilizer use efficiency on Ferrosol,             | et al., 2010)    |
|                                      |         | but reduced biomass on Calcarosol.                          |                  |
| Wood, cow manure                     | Maize   | Increase in yield from 14 - 150% due to                     | (Major et al.,   |
| (0, 10, 15, 20 t ha <sup>-1</sup> )  |         | increases in water use efficiency, pH and                   | 2010; Uzoma      |
|                                      |         | available Ca and Mg, and decrease in                        | et al., 2011)    |
|                                      |         | exchangeable acidity  |                  |
| Waste water sludge                   | Cherry  | Increment in yield by 64% over the control                  | (Hossain et al., |
| biochar (10 t ha <sup>-1</sup> )     | Tomato  | due to increased NP availability.                           | 2010)            |
| Coppiced trees (30,                  | wheat   | Increase in seed germination by 4-9%, with                  | (Solaiman et     |
| 60 t ha <sup>-1</sup> ), wood,       |         | yield improvement of 30% and sustained yield                | al., 2012;       |
| wheat chaff $(10 \text{ t ha}^{-1})$ |         | for two consecutive seasons.                                | Vaccari et al.,  |
|                                      |         |   | 2011)            |
| Cassava stem, farm-                  | Maize,  | Increase in yield due to improvements in soil               | (Abiven et al.,  |
| yard manure, maize                   | cassava | organic C, N, P, CEC, K and water                           | 2015; Islami et  |
| cob.                                 |         | availability.   | al., 2011)       |

# 1.4. Biochar and greenhouse gas emissions (GHG)

Applying biochar to soils has shown enormous potential to reduce emissions of greenhouse gases (Cayuela et al., 2014; Shackley et al., 2010). Biochar incorporation into soils is the crucial step in making biomass pyrolysis sustainable and carbon negative. The durability of biochar carbon in soil is such that net carbon emissions for the process are negative, for centuries to millennia (Lehmann et al., 2006; Wang et al., 2016). Soil emissions, in the form of CO<sub>2</sub>, methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O), can be reduced by 1.8 Pg CO<sub>2</sub>-C equivalent per year (12% of current anthropogenic CO<sub>2</sub>-C equivalent emissions) and total net emissions over the course of a century, by 130 Pg CO<sub>2</sub>-C equivalent. This would create a meaningful sink, in comparison to current fossil fuel emissions, of 8.7 Pg C per year (Woolf et al., 2010). Biochar holds promise as an amendment for soil quality improvement and sequestration of atmospheric CO<sub>2</sub>. Emissions of GHGs, such as N<sub>2</sub>O which is more than 300 times as potent as CO<sub>2</sub>, have been shown to be significantly reduced from soils amended by biochar (Felber et al., 2012; Lentz et al., 2014; Martin et al., 2015; Mukherjee et al., 2014). Biochar

may also be an ideal bulking agent for composting N-rich materials. Emissions from organic wastes and crop residues may be avoided by preventing its natural decomposition in soil. On-site pyrolysis may reduce the mass of wet waste by 20 to 30%, minimizing transportation costs and waste to landfill (McHenry, 2009). Biochar has been shown to act as an absorber of NH<sub>3</sub> and water-soluble ammonium ions (NH<sub>4</sub><sup>+</sup>), with ammonia (NH<sub>3</sub>) concentrations in the emissions decreasing by up to 64% if poultry litter was mixed with biochar, and total N losses being reduced by up to 52% during composting with biochar (Steiner et al., 2010). Hua et al. (2009) also, reported that total N loss at the end of composting with biochar decreased by 64%, and mobility of Cu and Zn in the sludge composting material could be reduced by 44% and 19%, respectively.

### 1.5. Problem statement

The mitigation of soil fertility depletion is currently a pressing issue and major concern in most countries of the world, especially in the tropical and sub-tropical regions. Significant demographic pressure and shrinking land area per capita, overgrazing, soil erosion, and the associated decline in soil fertility and organic matter are developing into major constraints for agricultural production. Agricultural production must increase substantially to feed the growing global population and satisfy the increasing demand of raw materials for industries. It is apparent that increases in crop production could be achieved through yield increases and system intensification. This will require a more holistic approach to agricultural production, which includes improved soil fertility management, attention to crop varieties, improved agronomic, soil and water management practices, in aggregate leading to reduced negative impacts and improved sustainability, with these new approaches being accessible to small-scale farmers as well as commercial producers. This will not only sustain increased food production, but also preserve the environment (Barrow, 2012).

Although one of the factors limiting the more widespread adoption of organic fertilizers is the high labor costs, escalating fertilizer costs combined with the reduced need for frequent application on biochar enhanced soils may catalyze a conversion to sustainable organic methods of fertilization (Glaser et al., 2015). Healthy, biochar-enriched soils may give farmers more options for crop selection. The increased fertility of the soil will also help farmers adapt to the changing climate, while widespread use of biochar will reduce the intensity of climate change (Lehmann et al., 2006; Zhang et al., 2016). Application of biochar to soils is now considered as a potential valuable input and remedy to restore soil fertility and reduce reliance on industrial fertilizers. This, consequently, raises

and sustains agricultural productivity while storing carbon for long-term, especially in the tropics, and to mitigate climate change.

Moreover, the information on biochar-compost mixtures, and their interaction with N fertilizer on soil fertility and crop performance in agricultural soils, is inadequate. Therefore, to maximize the favorable impact of biochar, this study was conducted to investigate the use of biochar in the particular context of three crops, maize, peanut and barley, with these crops subjected to a number of amendments - biochar, compost, biochar-compost mixture, co-composted biochar (COMBI), and inorganic fertilizer. Figure 1.2 shows the conceptual framework of the study.

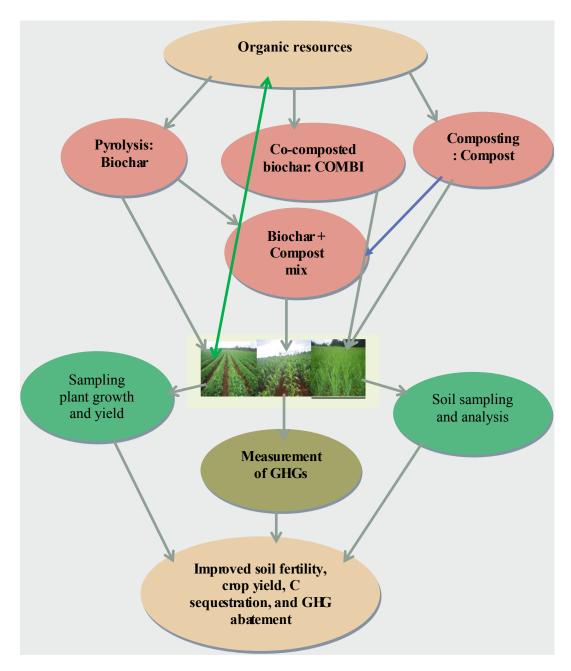


Figure 1.2. Conceptual frameworks for organic amendments and plant-soil relationships.

# 1.5.1. Aim and objectives

The general aim of this study was to investigate compost, biochar and their mixture as tools to enhance and sustain agricultural productivity, as well as reduce greenhouse gas emissions through improved soil quality. The specific objectives were to:

- i. Investigate and compare the effects of different soil fertility treatments in relation to biochar, compost and their mixture on soil physicochemical properties, and improvement in soil fertility.
- ii. Determine the response of several crops to biochar, compost, and their mixture on different soil types.
- iii. Evaluate the effect of different soil fertility treatments in relation to organic amendments and nitrogen fertilizer on nutrient uptake, nitrogen recovery, and use-efficiency of barley.
- iv. Investigate the impact of biochar, compost, and their mixture with respect to greenhouse gas emissions, carbon sequestration, and environmental benefits.

# 1.5.2. Research hypotheses

Specifically, I aimed to test the following hypotheses:

- i. The application of biochar, compost, and their mixture would enhance plant growth and yields of maize, peanut and barley relative to yields obtained from fertilizers alone.
- ii. The application of biochar, compost, and their mixture would improve soil properties, such as water holding capacity, soil pH, nutrient availability, and uptake by plants in comparison to fertilizer alone.
- iii. The application of biochar, compost, biochar-compost mixture, and N fertilizer would improve nitrogen use efficiency of barley.
- iv. The application of biochar, compost, and their mixture would reduce nutrient leaching relative to mineral fertilizer alone.
- v. The addition of biochar and biochar-compost mixture would reduce greenhouse gas emissions.

# **Overview of Chapter 2**

This chapter is based on a paper published in *Soil Research* with minimal formatting change. I have investigated the effects of biochar, compost, and biochar-compost mix, applied to an infertile tropical soil, on growth and nutrient uptake of maize; soil water content and chemical characteristics; and nutrient retention and leaching under greenhouse condition.

The study was a pot trial, designed to determine whether soil fertility and plant productivity could be enhanced by biochar and compost, applied singly or together and in the presence of inorganic fertilizers. I used willow biochar (WB; Earth Systems Pty Ltd, Melbourne, Vic.) and Acacia biochar (AB; Renewable Carbon Resources Australia Pty Ltd, Charleville, Qld) produced at a temperature of 500°C as soil amendments. I collected rigorous plant and soil data throughout the study period. I organized, analyzed and interpreted the data, and wrote the manuscript.<sup>2</sup>

<sup>&</sup>lt;sup>2</sup> Agegnehu, G., Bird, M., Nelson, P., and Bass, A. (2015). The ameliorating effects of biochar and compost on soil quality and plant growth on a Ferralsol. *Soil Research* **53**, 1-12.

# Chapter 2

# The Ameliorating Effects of Biochar and Compost on Soil Quality and Plant Growth on a Ferralsol

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#### Abstract

Deteriorating soil fertility and the concomitant decline in agricultural productivity are major concerns in many parts of the world. A pot experiment was conducted with Ferralsol to test the hypothesis that application of biochar improves soil fertility, fertilizer use efficiency, plant growth, and productivity, particularly when combined with compost. Treatments comprised: untreated control; mineral fertilizer at the rate of 280 mg N pot<sup>-1</sup>, 70 mg P pot<sup>-1</sup> and 180 mg K pot<sup>-1</sup> (F); 75% fertilizer + 40 g pot<sup>-1</sup> compost (F + Com); 100% F + 20 g pot<sup>-1</sup> willow biochar (WB) (F + WB); 75% F + 10 g pot<sup>-1</sup> willow biochar + 20 g pot<sup>-1</sup> compost (F + WB + Com); 100% F + 20 g pot<sup>-1</sup> acacia biochar (AB) (F + AB); and 75% F + 10 g pot<sup>-1</sup> AB + 20 g pot<sup>-1</sup> compost (F + AB + Com). Application of compost with fertilizer significantly increased plant growth, soil nutrient status, and plant nutrient content, with shoot biomass decreasing in the order F + Com (4.0) > F + WB + Com (3.6) > F + WB (3.3) >F + AB + Com(3.1) > F + AB(3.1) > F(2.9) > control (1.0), with the ratio of the value to that of the control given in brackets. Maize shoot biomass was positively, significantly correlated with chlorophyll content, root biomass, plant height, and specific leaf weight (r = 0.99, 0.98, 0.96 and 0.92, respectively). The shoot and root biomass had significant correlations with soil water content, plant nutrient concentration, and soil nutrient content after harvesting. The results of principal component analysis (PCA) showed that the first component (PRIN1) provided a reasonable summary of the data, accounting for about 84% of the total variance. As the plants grew, compost and biochar additions significantly reduced leaching of nutrients. In summary, the separate or combined application of compost and biochar together with fertilizer increased soil fertility and plant growth. Application of compost and biochar improved the retention of water and nutrients by the soil and thereby uptake of water and nutrients by the plants, but there was little or no observed synergistic effect.

**Keywords:** Biochar, carbon sequestration, compost, mineral fertilizer, nutrient leaching, soil quality

#### 2.1. Introduction

Soil degradation, by erosion, organic matter and plant nutrient depletion, and/or nutrient imbalances, are among the major challenges affecting agricultural productivity and food security (Foley et al. 2005; Lal, 2009b; Sanchez, 2002). The productivity of some lands has declined by 50% due to soil erosion and desertification (Eswaran et al., 2001). Annual global soil loss is estimated at 75 Gt, costing the world about US\$400 billion per year (Eswaran et al., 2001). The deterioration of soil fertility is exacerbated by nutrient mining, unsuitable land use and management, competing uses of

resources, and application of insufficient external inputs. For example, in Australia, crop production has led to a substantial loss of soil organic matter (SOM) from the cereal belt, where the long-term SOM loss often exceeds 60% from the top 0-10 cm of soil after 50 years of cereal cropping (Dalal and Chan, 2001). Loss of labile components of SOM, microbial biomass, and mineralizable N has been higher, resulting in greater decline in soil productivity (Dalal and Chan, 2001).

In the future, the long term benefit of allocating more land to agriculture will not offset the negative environmental impacts of land degradation (Tilman et al., 2002). Instead, a more promising approach to ensuring food security is to increase yield from currently cultivated land where productivity is low (Foley et al., 2011). Sustainable agricultural intensification, i.e., increasing productivity per unit land area, is thus necessary to secure the food supply for the increasing world population (Tilman et al., 2011). In most tropical environments, sustainable agriculture faces significant constraints due to low nutrient status and rapid mineralization of SOM (Zech et al., 1997). Decline in SOM content results in decreased cation exchange capacity (CEC). Under such circumstances, the efficiency of applied mineral fertilizers is low (Glaser et al., 2002; Troeh and Thompson, 2005). In addition, most small-scale farmers cannot afford to apply mineral fertilizers regularly due to high costs. Therefore, nutrient deficiencies are prevalent in many crop production systems of the tropics and this constraints productivity.

Soils fertilized with compost or manure have higher contents of SOM and soil microorganisms than mineral fertilized soils, and are more enriched in P, K, Ca and Mg in the top soil and NO<sub>3</sub>-N, Ca and Mg in sub-soils (Edmeades, 2003; Quilty and Cattle, 2011). Well-made composts are known to improve soil structure, resulting in improved air exchange, water infiltration and retention (Fischer and Glaser, 2012). Soils amended with organic fertilizers also have lower bulk density and higher porosity, hydraulic conductivity, and aggregate stability than mineral fertilized soils (Edmeades, 2003; Lal, 2009b). Labile forms of SOM are of prime importance as a reserve and source of plant nutrients in tropical soils poor in minerals that can be weathered (Zech et al. 1997). However, the amount of the labile SOM is smaller, and the turnover rate and release of nutrients in humid tropical soils is faster, than temperate soils. Accelerated mineralization of SOM limits the practical application of organic fertilizers in the tropics (Kaur et al., 2008; Zech et al., 1997).

Biochar is charcoal produced by controlled pyrolysis for use as a soil amendment or carbon sequestration technique (Lehmann and Rondon, 2006). A range of studies have shown that application of biochar to soil can improve soil biophysical and chemical properties, and nutrient supply to plants (Glaser et al., 2002; Sohi et al., 2010), enhance plant growth and yield (Chan et al. 2007; Lehmann

and Rondon, 2006; Major et al. 2010) and reduce greenhouse gas emissions through C sequestration (Ippolito et al. 2012; Van Zwieten et al. 2010; Zhang et al. 2012a). Biochar helps improve agricultural productivity by reducing soil acidity, and enhancing CEC and fertilizer use efficiency (Chan and Xu, 2009; Lehmann et al., 2003; Steiner et al., 2008), water retention capacity (Downie, 2011), plant available water content (Tammeorg et al., 2014), and creating a habitat for beneficial soil microorganisms (Thies and Rillig, 2009). Biochar can be used to rejuvenate depleted soils, making more agricultural land available and increasing crop yields so that the need for expansion of agricultural land area is decreased (Barrow, 2012; Blackwell et al., 2009). Biochar has significantly improved the efficiency of N fertilizers and increased plant growth and yield (Lehmann et al., 2003; Steiner et al., 2008). The long-term benefits of biochar for nutrient availability include a greater stabilization of SOM, slower nutrient release from added organic matter, and better retention of cations due to a greater CEC (Lehmann et al., 2003; Steiner et al., 2008). The resultant change in soil nutrient status may affect both plant growth and productivity. Responses to biochar application will depend on the type and rate of biochar applied, as well as soil physicochemical characteristics.

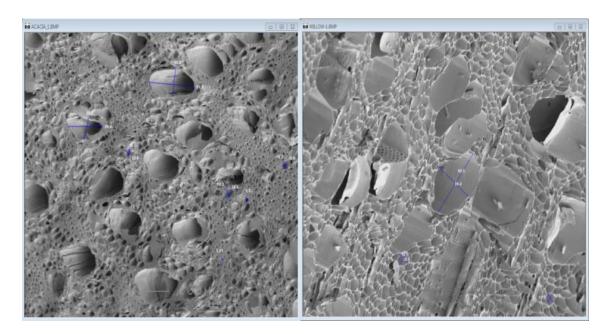
Some recent studies have indicated that the simultaneous application of biochar with compost could lead to enhanced soil fertility, improved plant growth and C sequestration potential (Fischer and Glaser, 2012; Schulz and Glaser, 2012). Liu et al. (2012) showed that the combined application of compost and biochar had a synergistic positive effect on SOM content, nutrient contents, and water holding capacity of soil under field conditions. Overall, information on the combined effects of biochar and compost on soil fertility and crop performance in tropical soils is not adequate. Different ways of producing and applying compost and biochar are hypothesized to differ in their effects on soil bio-physical and chemical properties, plant growth, and yield. Therefore, the objectives of this study were to determine the effect of compost and biochar applied to an infertile tropical soil, on: 1) growth and nutrient uptake of maize; 2) soil water content and chemical characteristics; and 3) nutrient retention and leaching.

## 2.2. Materials and methods

## 2.1.1. Experimental set-up

The study was a pot trial, designed to determine whether soil fertility and plant productivity could be enhanced by biochar and compost, applied singly or together. Willow biochar (WB; Earth Systems Pty Ltd, Melbourne, Vic.) and Acacia biochar (AB; Renewable Carbon Resources Australia Pty Ltd, Charleville, Qld) produced at a temperature of 550 °C and 500 °C, respectively were selected for the

soil amendments. Both biochar types were characterized using JSM-6300 Scanning Microscope at the Advanced Analytical Center of James Cook University before application (Figure 2.1). Prior to imaging, biochar samples were sputter coated with gold. The operating accelerating voltage was 5 KV, and the software used was "Semafore." WB had more pore spaces than AB. Compost was produced from a mix of bagasse, poultry litter and municipal waste following the standard windrow procedures for compost preparation by King Brown Technologies.



**Figure 2.1.** Microscopic images of acacia (left) and willow (right) biochars; pore sizes in micro meter.

The chemical characteristics of compost, WB, and AB, which were used as soil amendments in this study, are given in Table 2.1. They were screened through a 4-mm sieve before being mixed with the soil. Soil used in the trial was taken from a sugarcane field (17°1.23' S latitude and 145° 24.21' E longitude, 0-10 cm depth) in north Queensland, Australia. The international nomenclature of Ferralsols (FAO Soil Classification, IUSS Working Group WRB, 2014) is known as Oxisols (Soil Taxonomy, USA), Latosols (Brazil), Soils ferallitiques (France), Lateritic soils, Ferralitic soils (Russia), and Ferrosols (Australian Soil Classification). The reference soil group of the Ferralsols, developed on Quaternary basalt, represents the most highly weathered soils in the classification system (Brady and Weil, 2014). Soil chemical properties of the experimental soil were determined for samples taken before planting (Table 2.1). The pot trial was conducted in a greenhouse from the 7<sup>th</sup> of June to the 9<sup>th</sup> of August 2013 at James Cook University, Cairns, Australia. During the growing period, the temperature ranged between17.6 and 27.1°C, and the mean relative humidity was 62.5%.

The plastic pots used had 16 cm upper and 14 cm lower diameters, a height of 16 cm, and a total volume of  $\approx 2750$  cm<sup>3</sup>. A 2kg subsample of air-dried soil, screened through a 4-mm sieve, was placed into each pot after mixing thoroughly with the amendments. All pots were then watered to approximately field capacity.

The experiment comprised the following seven combinations of compost, biochar, and mineral fertilizer treatments: 1) untreated control; 2) mineral fertilizer only (F) at the rate of 280 mg N pot<sup>-1</sup> as urea, 70 mg P pot<sup>-1</sup> as triple superphosphate and 180 mg K pot<sup>-1</sup> as KCl-muriate of potash, which is equivalent to 140 kg N ha<sup>-1</sup>, 35 kg P ha<sup>-1</sup> and 90 kg K ha<sup>-1</sup>; 3) 75% fertilizer + 40 g pot<sup>-1</sup> compost (F + Com); 4) 100% F + 20 g pot<sup>-1</sup> willow biochar (WB) (F + WB); 5) 75% F + 10 g pot<sup>-1</sup> willow biochar + 20 g pot<sup>-1</sup> compost (F + WB + Com); 6) 100% F + 20 g pot<sup>-1</sup> acacia biochar (AB) (F + AB); and 7) 75%  $F + 10 \text{ g pot}^{-1} \text{ AB} + 20 \text{ g pot}^{-1} \text{ compost} (F + \text{AB} + \text{Com})$ . All amendments were thoroughly mixed with the experimental soil before potting. The experiment was arranged in a randomized complete block design with four replications. Compost was applied at the rates of 20 and 10 t ha<sup>-1</sup>, and biochar at the rates of 10 and 5 t ha<sup>-1</sup>. The N content of the compost was considered and fertilizer N rate was decreased by 25% when applied together with compost, assuming only 10-20% of the total N content of the compost is mineralized in the first year (Fischer and Glaser 2012). Nitrogen was applied in two applications: 156 mg N pot<sup>-1</sup> at planting and 124 mg N pot<sup>-1</sup> as top-dressing, four weeks after planting. Eight maize (Zea mays L.) seeds were planted in each pot at a depth of 3 cm one week after the amendments had been mixed with the soil. After emergence, four plants per pot were maintained until harvesting. Each pot was given 150 mL of water daily for 14 days after planting and 200 mL afterwards until the end of the experiment. The bottom of each pot had four holes and was lined with gauze to minimize the loss of particulate matter but allow leaching of soil solution. Each pot was equipped with a sealable plastic bag at the bottom for leachate collection.

|  | Unit                | Compost | Willow  | Acacia  | Experimental |
|--|---------------------|---------|---------|---------|--------------|
| Parameter                              |                     |         | biochar | biochar | soil         |
| pH-H <sub>2</sub> O                    | units               | 8.1     | 9.5     | 8.0     | 6.5          |
| pH-CaCl <sub>2</sub>                   | units               | -       | 8.3     | 7.5     | 5.6          |
| Total carbon (C)                       | g kg <sup>-1</sup>  | 233     | 924     | 590     | 11.4         |
| Total nitrogen (N)                     | g kg <sup>-1</sup>  | 11      | 0.36    | 0.43    | 0.80         |
| Extractable phosphorus (Colwell P)     | g kg <sup>-1</sup>  | 3.3     | 1.96    | 0.02    | 0.031        |
| Exchangeable potassium (K)             | g kg <sup>-1</sup>  | 8.9     | 15.4    | 0.09    | 0.30         |
| Exchangeable calcium (Ca)              | g kg <sup>-1</sup>  | 14      | 15.2    | 3.9     | 0.86         |
| Exchangeable magnesium (Mg)            | g kg <sup>-1</sup>  | 7.0     | 4.4     | 0.06    | 0.11         |
| Exchangeable sodium (Na)               | g kg <sup>-1</sup>  | 2.1     | 1.04    | 0.09    | 0.01         |
| ECEC                                   | g kg <sup>-1</sup>  | nd      | nd      | nd      | 1.28         |
| Total sulfur (S)                       | g kg <sup>-1</sup>  | 1.5     | 0.07    | 0.05    | nd           |
| Ammonium nitrogen (NH <sub>4</sub> -N) | mg kg <sup>-1</sup> | nd      | nd      | nd      | 11.3         |
| Nitrate nitrogen (NO <sub>3</sub> -N)  | mg kg⁻¹             | nd      | nd      | nd      | 14.0         |
| Copper (Cu)                            | mg kg⁻¹             | 72.0    | 239.7   | 9.4     | nd           |
| Zinc (Zn)                              | mg kg <sup>-1</sup> | 181.0   | 2571    | 38.0    | nd           |
| Manganese (Mn)                         | mg kg <sup>-1</sup> | 421.5   | 14599   | 22.0    | nd           |
| Iron (Fe)                              | mg kg <sup>-1</sup> | 17877   | 125898  | 47.0    | nd           |
| Boron (B)                              | mg kg <sup>-1</sup> | 17.0    | nd      | 9.3     | nd           |
| Molybdenum (Mo)                        | mg kg <sup>-1</sup> | 2.2     | nd      | < 0.3   | nd           |
| Cobalt (Co)                            | mg kg <sup>-1</sup> | 9.8     | nd      | < 0.4   | nd           |
| Aluminum (Al)                          | mg kg <sup>-1</sup> | 7596    | 3175    | 400     | nd           |

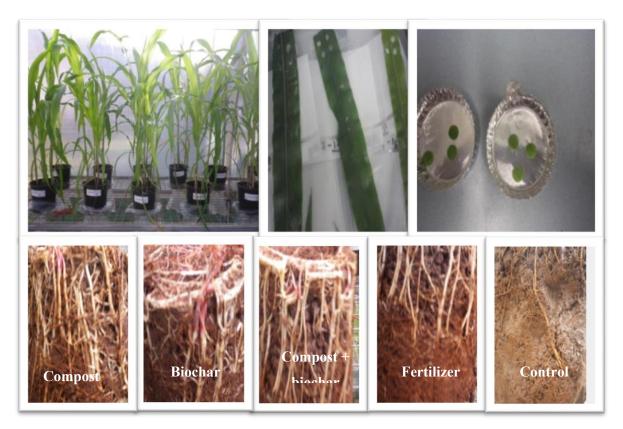
**Table 2.1.** Chemical characteristics of compost, willow biochar, acacia biochar and experimental soil before planting.

Notes: Cu, Zn, Mn and Fe are DTPA extractable micronutrients; nd: Not determined

# 2.1.2. Measurements

Plant height was recorded just prior to harvesting. Chlorophyll content (greenness) was measured on the 34<sup>th</sup>, 48<sup>th</sup> and 52<sup>th</sup> day after planting, with four replicate measurements on three leaves (youngest fully expanded leaves) per plant, using a chlorophyll meter (Minolta SPAD 502). Leachate was collected every two weeks, its volume was measured, and subsamples were kept frozen for analysis.

Soil water content (SWC) was measured every week after planting using a HydroSense II probe (Campbell Scientific, INC). Specific leaf weight (SLW, expressed as dry weight per cm<sup>2</sup>) of plants was measured. One leaf from each plant of the same age and position was harvested one week before harvesting. Leaves were transferred immediately to individual, sealed plastic bags that were kept in an insulated box above ice packs until all leaves were harvested. In the laboratory, a hole-punch with a diameter of 8 mm was used to take a leaf disc from the middle of the leaf lamella (Figure 2.2). Fresh leaf discs were weighed and dried at 70°C for 48 h before reweighing them. SLW was calculated as dry weight of leaf disc per area of hole-punch.



**Figure 2.2.** Growing maize plants in pots, leaf samples and leaf discs for the determination of specific leaf weight (upper), and root growth under different soil fertility treatments (lower).

The above-ground parts of four plants were harvested from each pot at soil level 62 days after planting. The root part of each plant from each pot was also separated judiciously from the soil, cleaned, and weighed. The fresh shoots and roots were dried separately at 70°C for 72 h, and the shoot/root ratio (SRR) was determined. The shoot part of plants was used to determine nutrient content and uptake of nutrients by shoots. Total shoot N and C concentrations were determined using an elemental analyzer (ECS 4010 CHNSO Analyzer; Costech Analytical Technologies INC, CA,

USA) fitted with a Zero Blank Auto-sampler (Costech Analytical Technologies, INC). Total P, K and NO<sub>3</sub>-N concentrations in plants were quantified at the Analytical Research Laboratories (ARL) in New Zealand. Nitrate-N in plant tissue was determined using 2% acetic acid (Miller, 1998). Plant K concentration was determined after wet digestion with sulfuric acid by atomic absorption spectroscopy (Watson et al., 1990). Plant P concentration was determined photometrically with the molybdenum blue method (Mills and Jones, 1996).

Total soil C and N concentrations were determined in the same way used for plants. Exchangeable cations and P contents, electrical conductivity (EC), NO<sub>3</sub>-N, and NH<sub>4</sub>-N were determined by the ARL in New Zealand. The EC was determined by a conductivity meter on a 1:2.5 soil: water suspension (Rayment and Lyons, 2010). Available soil P was determined according to Colwell (1963). Exchangeable K, Na, Ca and Mg were determined using 1M ammonium acetate extraction method buffered at pH 7 for 30 minutes (Rayment and Lyons, 2010). Soil NH<sub>4</sub>-N and NO<sub>3</sub>-N were determined colorimetrically after extraction with 1M KCl (Rayment and Lyons, 2010). The chemical properties of compost, WB, and AB were determined in the laboratory following similar methods for soil analysis (Table 2.1). Pot leachate samples were analyzed for NO<sub>3</sub>-N, P, K, Mg, Ca, and Na concentration. Nitrate concentration of the leachate was measured using a Nitrate-Meter, (B-743, Horiba Scientific, Japan). Phosphorus concentration was determined photometrically using the molybdate blue method, and K, Ca, Na and Mg concentrations were measured using inductively coupled plasma atomic emission spectroscopy at the Advanced Analytical Center, James Cook University.

#### 2.1.3. Data analysis

The data were subjected to analysis of variance using the general linear model procedure (PROC GLM) of SAS statistical package version 9.1 (SAS Institute, Cary, NC, USA). The total variability for each trait was quantified using the following model.

$$Y_{ij} = \mu + R_i + T_j + RT_{ij} + e_{ij}$$

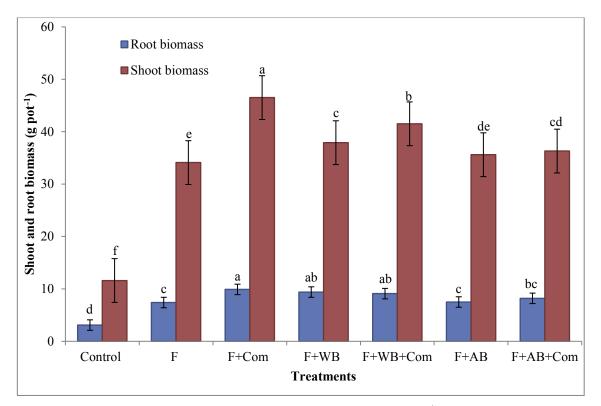
Where  $Y_{ij}$  is total observation,  $\mu$  = grand mean,  $R_i$  is effect of the i<sup>th</sup> replication,  $T_j$  is effect of the j<sup>th</sup> treatment,  $RT_{ij}$  is the interaction, and  $e_{ij}$  is the variation due to random error. Means for the treatments were compared using the MEANS statement with the least significant difference (LSD) test at the 5% probability level. To perform the multivariate approach of correlation and principal component analysis (PCA), the data were standardized by removing treatment mean character values followed by dividing by the corresponding character standard deviations. The correlation coefficients (r) were,

then, calculated among plant parameters, nutrient uptake and soil nutrient contents using the SAS CORR procedure, and the PCA was performed using the SAS PRINCOMP procedure to distinguish the treatments as a function of the soil management and determine the most important parameters to characterize them.

#### 2.2. Results

#### 2.2.1. Shoot nutrient concentration

The different soil fertility treatments significantly (p < 0.05 and p < 0.001) increased above- and below-ground biomass, shoot/root ratio, specific leaf weight (SLW), chlorophyll content, and plant height of maize (Figure 2.3, Table 2.2). Shoot and root biomass were greater with F + Com than with other treatments. The shoot biomass recorded from F + Com and F + WB + Com treated soil was 4 and 3.6 times those of the control and 1.4 and 1.2 times those of fertilizer only treatments, respectively (Figure 2.3). Root biomass was highest in the F + Com treatment, but the differences between F +Com, F + WB, and F + WB + Com treatments were not statistically significant. Applications of F +Com and F + WB produced root biomass 3.2 and 3.0 times those of the control and 1.3 and 1.3 times those of the fertilizer only treatments, respectively. When the two biochar types were compared, WB addition increased root biomass over AB by 20%. However, applications of F + Com, F + WB + Com or F + AB + Com resulted in similar shoot to root ratios. The shoot and root parts of plants constitute 73-79% and 21-27% of plant biomass, respectively. Plants grown with F + Com or F + WB + Com had higher chlorophyll content and plant height than the other treatments. Chlorophyll contents in the F, F + Com, and F + WB + Com treatments were 1.3, 1.5 and 1.4 times that of the control (Table 2.2). Fertilizer + WB + Com resulted in the highest SLW of plants, with the lowest being from the control treatment.



**Figure 2.3.** Effects of treatments on shoot and root biomass (g pot<sup>-1</sup>) of maize. Least significant difference (LSD) = 2.1 and 1.1, and Coefficient of variation (CV) = 4.1and 9.7 for root and shoot biomass, respectively. Columns with the same letter are not significantly different at p < 0.05. Error bars represent ± 1SE.

The treatments significantly (p < 0.01) increased shoot uptake of total N, C, P and resulted in plants with a higher shoot nitrate concentration (Table 2.2). Fertilizer + Com, F + WB, and F + WB or AB with Com significantly increased plant N uptake compared to the fertilizer alone treatment. Plant C, N, P and K uptake ranged from 4.4-19.7 g pot<sup>-1</sup>, and 150-1,117, 15-87 and 354-1,746 mg pot<sup>-1</sup>, respectively, with the highest values from F + Com and the lowest from the control treatment (Table 2.2). However, there was no statistically significant difference between F + Com and F + WB + Com for N, NO<sub>3</sub>-N and P uptake. Nitrogen and P contents of plants in the F + Com treatment were 7.4 and 5.9 times those of the control and 1.7 and 1.5 times those of the fertilizer only treatments, respectively. Applications of biochar and compost resulted in optimal N and P uptake by plants (within the sufficiency range, i.e. 3.5-5.0% for N and 0.35-0.80% for P). Fertilizer + Com resulted in the highest plant NO<sub>3</sub>-N accumulation (41 mg pot<sup>-1</sup>), and the lowest plant NO<sub>3</sub>-N content was in the control (Table 2.2). But significant differences were not observed in NO<sub>3</sub>-N uptake between organic amendments and mineral fertilizer, except the F + Com treatment. Plants treated with mineral fertilizer alone, F + Com, F + WB + Com, and F + AB + Com had increased NO<sub>3</sub>-N of 9.6, 14, 11.8

and 10 times that of the control treatment. Overall, application of compost and biochar in conjunction with F increased plant growth, and plant N content with F in particular and then F + Com with biochars. While shoot biomass decreased in the order F + Com (4.0) > F + WB + Com (3.6) > F + WB (3.3) > F + AB + Com (3.1) > F + AB (3.1) > F (2.9) > control (1.0), with the ratio of the value to that of the control (given in brackets).

|                | SLW            | Chlorophyll   | Plant height | С                      | Shoot nutrient content (mg pot <sup>-1</sup> ) |                    |        |         |  |
|----------------|----------------|---------------|--------------|------------------------|--|--------------------|--------|---------|--|
| Tractmont      | $(mg cm^{-2})$ | content (SPAD | (cm)         | content –              | N  | NO <sub>3</sub> -N | Р      | K       |  |
| Treatment      |                | unit)         |              | $(g \text{ pot}^{-1})$ | IN   | INO3-IN            | P      | K       |  |
| Control        | 1.98 c         | 26.9 d        | 78.2 b       | 4.41 e                 | 150 d  | 2.9 c              | 14.8 c | 354 d   |  |
| F              | 2.34 b         | 36.3 c        | 111.0 a      | 14.19 d                | 648 c  | 27.9 b             | 60.1 b | 1148 c  |  |
| F + Com        | 2.58 ab        | 40.6 a        | 115.0 a      | 19.66 a                | 1117 a   | 40.8 a             | 86.9 a | 1746 a  |  |
| F + WB         | 2.58 ab        | 39.0 ab       | 112.0 a      | 16.00 c                | 943 b  | 28.6 b             | 69.1 b | 1330 bc |  |
| F + WB + Com   | 2.66 a         | 40.5 a        | 114.3 a      | 17.35 b                | 990 ab   | 34.3 ab            | 86.1 a | 1470 b  |  |
| F + AB         | 2.56 ab        | 37.6 bc       | 110.5 a      | 14.68 d                | 771 c  | 29.1 b             | 60.6 b | 1219 c  |  |
| F + AB + Com   | 2.36 b         | 38.1 bc       | 112.5 a      | 14.89 d                | 904 b  | 29.4 b             | 69.6 b | 1316 bc |  |
| <i>p</i> level | **             | ***           | ***          | **                     | ***  | ***                | ***    | **      |  |
| LSD (0.05)     | 0.28           | 2.1           | 5.8          | 0.98                   | 129.4  | 7.9                | 10.5   | 226.0   |  |
| CV (%)         | 7.7            | 4.0           | 3.6          | 4.59                   | 11.2   | 10.4               | 11.1   | 12.4    |  |

Table 2.2. Effects of treatments on plant parameters and nutrient uptake into shoot parts of plants.

Significant at \*p < 0.05, \*\*p < 0.01, \*\*\*p < 0.001; NS: Not significant. SLW: Specific leaf weight; LSD: Least significant difference; CV: Coefficient of variation. Within each column, means with different letters are significantly different at p < 0.05.

#### 2.2.2. Soil characteristics

The treatments with organic components significantly (p < 0.001) improved soil water content (SWC) and decreased leachate volume (Table 2.3). Differences in leachate volume among treatments increased during the growing period, as the demand for water by plants depended on treatments. Therefore, the cumulative leachate volume was inversely related with the above- and below-ground biomass. Soil water content was highest in compost- and biochar-treated soil. Following harvest, total C, total N, C: N ratio, NO<sub>3</sub>-N and NH<sub>4</sub>-N, available P, exchangeable cations, ECEC, and EC significantly responded to the treatments (Tables 2.3 and 2.4). Soil nutrients were higher when compost and biochar were added to soil than in the control and fertilizer only treatments. The highest SOC content (33g pot<sup>-1</sup>) was obtained from F + WB addition, which is in agreement with the initial C content of WB. In F + WB amended soil, SOC content increased by a factor of 1.4 and 1.5 compared to the control and fertilizer only treatments (Table 2.3). There was a linear relationship between the amount of C added in the amendments and the SOC content at the end of the experiment. The maximum Colwell P value (108 mg pot<sup>-1</sup>) was obtained from Com + F treated soil. The highest soil NO<sub>3</sub>-N and NH<sub>4</sub>-N contents were from F + Com and F + WB + Com treated soil (Table 2.3). Fertilizer + Com, and F + WB + Com treated soil had higher contents of available nutrients, such as K. Ca and Mg, than the other treatments (Table 2.4). Fertilizer + WB + Com addition especially increased soil exchangeable K, Ca and ECEC after harvesting, while F + Com addition resulted in the highest exchangeable Mg and Na. The ratios of Ca to Mg for the different treatments ranged from 4.0 - 5.4 (Table 2.4), which is within the recommended range (4 - 6).

|                | Cumulative              | SWC Total C and N |        | C: N ratio | Available nutrients (mg pot <sup>-1</sup> ) |         |                    |        |
|----------------|-------------------------|-------------------|--------|------------|---|---------|--------------------|--------|
|                | leachate                | (%)               | (g pot | ·1)        |   |         |                    |        |
| Treatment      | (mL pot <sup>-1</sup> ) |                   | TC TN  |            |   | Р       | NO <sub>3</sub> -N | NH4-N  |
| Control        | 715 a                   | 17.6 c            | 23.2 c | 1.40 c     | 16.6 a                                      | 49.0 d  | 1.01 e             | 35.6 c |
| F              | 189 b                   | 20.0 c            | 22.4 c | 1.42 c     | 15.8 ab                                     | 68.4 c  | 4.02 d             | 45.0 c |
| F + Com        | 58 e                    | 23.7 b            | 30.0 b | 2.20 a     | 13.6 c                                      | 107.6 a | 17.6 a             | 85.2 a |
| F + WB         | 68 e                    | 26.3 ab           | 33.4 a | 2.0 ab     | 16.7 a                                      | 95.0 ab | 10.8 c             | 61.4 b |
| F + WB + Com   | 54 e                    | 26.9 a            | 28.7 b | 2.02 ab    | 14.2 b                                      | 97.6 ab | 15.8 ab            | 86.0 a |
| F + AB         | 140 c                   | 26.5 ab           | 27.4 b | 1.80 b     | 15.2 ab                                     | 88.6 b  | 14.8 b             | 77.8 a |
| F + AB + Com   | 81 d                    | 28.1 a            | 27.8 b | 2.01 ab    | 13.8 c                                      | 94.2 ab | 8.6 c              | 61.8 b |
| <i>p</i> level | ***                     | ***               | ***    | **         | **  | ***     | ***                | ***    |
| LSD (0.05)     | 18.1                    | 3.1               | 3.4    | 0.22       | 1.8   | 15.5    | 2.2                | 13.6   |
| CV (%)         | 6.5                     | 8.8               | 8.1    | 8.4        | 8.0   | 11.3    | 14.3               | 14.2   |

Table 2.3. Effects of treatments on cumulative leachate, soil water content and soil chemical properties after harvesting.

Significant at  ${}^{**}p < 0.01$ ,  ${}^{***}p < 0.001$ ; NS: Not significant. SWC: Soil water content; TC: Total carbon; TN: Total nitrogen; P: Colwell phosphorus. Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation.

|                | Ι      | Ca/Mg   | EC     |          |         |        |                       |
|----------------|--------|---------|--------|----------|---------|--------|-----------------------|
| Treatment      | К      | Ca      | Mg     | Na       | ECEC    | ratio  | (dS m <sup>-1</sup> ) |
| Control        | 202 c  | 1590 d  | 190 c  | 40.2 d   | 2020 d  | 8.4 a  | 0.05 c                |
| F              | 210 c  | 1660 cd | 192 c  | 43.6 cd  | 2104 cd | 8.7 a  | 0.07 b                |
| F + Com        | 306 ab | 1885 ab | 284 a  | 72.4 a   | 2516 a  | 6.5 c  | 1.0 ab                |
| F + WB         | 304 ab | 1800 b  | 216 bc | 56.4 bc  | 2378 b  | 8.4 a  | 1.11 a                |
| F + WB + Com   | 342 a  | 1950 a  | 242 b  | 64.4 ab  | 2600 a  | 8.1 ab | 0.08 b                |
| F + AB         | 232 c  | 1680 cd | 202 c  | 43.8 cd  | 2158 c  | 8.3 ab | 0.08 b                |
| F + AB + Com   | 288 b  | 1760 bc | 236 b  | 51.8 bcd | 2334 b  | 7.5 b  | 1.0 ab                |
| <i>p</i> level | ***    | ***     | ***    | **       | **      | **     | **                    |
| LSD (0.05)     | 41.6   | 119.0   | 31.0   | 13.0     | 127     | 0.85   | 0.02                  |
| CV (%)         | 10.4   | 4.6     | 9.3    | 16.5     | 3.5     | 7.2    | 17.4                  |

Table 2.4. Effects of treatments on soil chemical properties after harvesting.

Significant at \*\*p < 0.01, \*\*\*p < 0.001; NS: Not significant; ECEC: Effective cation exchange capacity; EC: Electrical conductivity. Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation.

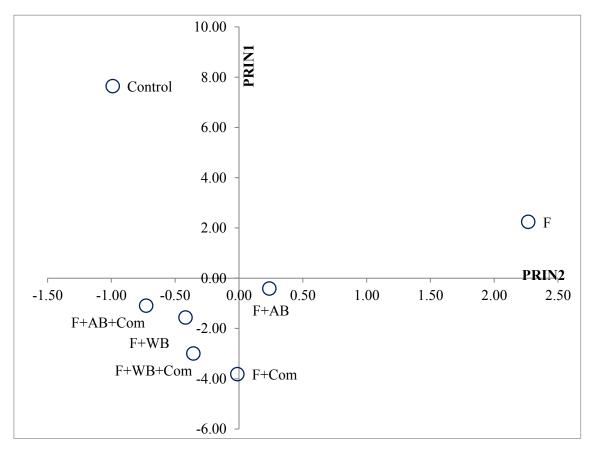
Significant positive correlations were observed between plant growth parameters, plant nutrient concentrations, SWC, and soil chemical properties (p < 0.05, p < 0.01 and p < 0.001). Shoot biomass was positively correlated with leaf chlorophyll content, root biomass, plant height, SLW, and SWC (r = 0.99, 0.98, 0.96, 0.92 and 0.74, respectively) under different soil fertility treatments (Table 2.5). However, the amount of leachate collected as percolated water was negatively correlated with shoot and root biomass, leaf chlorophyll content, SLW, and SWC, but not significantly (data not shown). Shoot and root biomass, chlorophyll content, and plant height were positively, significantly correlated with plant and soil contents of N, NO<sub>3</sub>-N and P (Table 2.5). Plant N content was positively correlated with SWC, soil N, P and K contents, and plant NO<sub>3</sub>-N, P and K concentrations, but soil K content was not significantly correlated with shoot biomass, plant height, SLW, plant NO<sub>3</sub>-N concentration and SWC.

|                          |                    |         |                    | Shoot              | Shoot              |                    |            |            |             |         |         |         |
|--------------------------|--------------------|---------|--------------------|--------------------|--------------------|--------------------|------------|------------|-------------|---------|---------|---------|
| Parameters               | Soil K             | Soil P  | Soil N             | NO <sub>3</sub> -N | Κ                  | Shoot P            | Shoot N    | SWC        | SLW         | PHT     | CHLC    | RB      |
| SB                       | 0.73 <sup>ns</sup> | 0.93**  | $0.80^{*}$         | 0.99***            | 0.99***            | 0.98***            | 0.98***    | 0.74*      | 0.92**      | 0.96*** | 0.99*** | 0.98*** |
| RB                       | $0.79^{*}$         | 0.94**  | 0.83*              | 0.96***            | 0.97***            | 0.97***            | 0.98***    | $0.75^{*}$ | 0.92**      | 0.95**  | 0.98*** |         |
| CHLC                     | $0.77^*$           | 0.94**  | $0.81^{*}$         | $0.97^{***}$       | 0.98***            | 0.98***            | 0.98***    | $0.78^*$   | 0.94**      | 0.97*** |         |         |
| PHT                      | 0.63 <sup>ns</sup> | 0.85*   | 0.68 <sup>ns</sup> | 0.96***            | 0.94**             | 0.94**             | 0.93**     | $0.74^{*}$ | $0.88^{**}$ |         |         |         |
| SLW                      | 0.75 <sup>ns</sup> | 0.89**  | $0.77^{*}$         | 0.89**             | 0.89**             | 0.91**             | 0.91**     | $0.76^{*}$ |             |         |         |         |
| SWC                      | 0.72 <sup>ns</sup> | 0.83*   | 0.79*              | 0.66 <sup>ns</sup> | 0.69 <sup>ns</sup> | 0.72 <sup>ns</sup> | $0.78^{*}$ |            |             |         |         |         |
| Total shoot N            | 0.83*              | 0.98*** | 0.90**             | 0.96***            | 0.98***            | 0.98***            |            |            |             |         |         |         |
| Total shoot P            | 0.81*              | 0.93**  | $0.82^{*}$         | 0.98***            | 0.98***            |                    |            |            |             |         |         |         |
| Total shoot K            | $0.75^{*}$         | 0.95**  | $0.84^{*}$         | 0.99***            |                    |                    |            |            |             |         |         |         |
| Plant NO <sub>3</sub> -N | 0.69 <sup>ns</sup> | 0.91**  | $0.77^{*}$         |                    |                    |                    |            |            |             |         |         |         |
| Soil TN                  | 0.89**             | 0.96*** |                    |                    |                    |                    |            |            |             |         |         |         |
| Soil P                   | 0.85*              |         |                    |                    |                    |                    |            |            |             |         |         |         |

Table 2.5. Correlation coefficients among plant parameters, soil water content, soil nutrient concentration and shoot nutrient content.

Significant at  ${}^{*}p < 0.05$ ,  ${}^{**}p < 0.01$ ,  ${}^{***}p < 0.001$ ; ns: Soil TN: Total nitrogen; Soil P: Colwell phosphorus; Soil K: Exchangeable potassium; Not significant; SSWC: Soil water content; SLW: Specific leaf weight; PHT: Plant height; CHLC: Chlorophyll content; RB: Root biomass; SB: Shoot biomass.

The results of principal component analysis (PCA) revealed that the first two principal components (PRIN1 and PRIN2) accounted for  $\approx$ 91% of the total variation of the treatments, of which 84% was contributed by the first principal component (Table 2.6). The first eigenvector had similar weights on all of the characters. Thus, most characters in PRIN1 individually contributed comparable effects (0.186 - 0.255) to the total variation of the treatments (Table 2.6). The second eigenvector had positive loadings on the variables soil N and K contents, SOC, SWC and effective CEC. Each vector corresponded to one of the analysis variables and was proportional to its component loading. For instance, the biplot of PRIN1 and PRIN2 showed that the variables F + Com, F + WB + Com, F + WB, F+ AB + Com and controlled load heavily on the first component, while the variables F and F +AB loaded heavily on the second component (Figure 2.4).



**Figure 2.4.** Plot of principal component one and principal component two (PRIN1 and PRIN 2) in 7 treatments. F: Fertilizer; AB: Acacia biochar; Com: Compost; WB: Willow biochar.

| Parameter                              | PRIN1        | PRIN 2 | PRIN3  | PRIN4  |
|--|--------------|--------|--------|--------|
| Eigenvalue                             | 15.1         | 1.17   | 0.65   | 0.51   |
| %variance                              | 84.00        | 6.49   | 3.62   | 2.84   |
| Cumulative                             | 84.00        | 90.49  | 94.11  | 96.95  |
| Character                              | Eigenvectors |        |        |        |
| Plant N concentration                  | 0.255        | -0.037 | 0.015  | 0.130  |
| Soil P content                         | 0.254        | 0.119  | 0.015  | 0.011  |
| Chlorophyll content                    | 0.252        | -0.169 | -0.040 | 0.087  |
| Plant P concentration                  | 0.251        | -0.145 | 0.101  | 0.060  |
| Plant K concentration                  | 0.251        | -0.160 | 0.103  | 0.026  |
| Shoot biomass                          | 0.250        | -0.214 | 0.040  | 0.019  |
| Root biomass                           | 0.249        | -0.142 | -0.116 | 0.200  |
| Plant C content                        | 0.249        | -0.217 | 0.020  | 0.021  |
| Plant NO <sub>3</sub> -N concentration | 0.245        | -0.256 | 0.125  | -0.032 |
| Specific leaf weight                   | 0.240        | -0.111 | -0.328 | -0.231 |
| Plant height                           | 0.234        | -0.324 | -0.002 | 0.206  |
| Soil N content                         | 0.234        | 0.358  | 0.059  | 0.048  |
| Soil NO <sub>3</sub> -N content        | 0.230        | 0.115  | -0.056 | -0.589 |
| Soil NH <sub>4</sub> -N content        | 0.228        | 0.125  | 0.064  | -0.603 |
| Soil K content                         | 0.217        | 0.345  | -0.022 | 0.176  |
| Soil water content                     | 0.208        | 0.256  | -0.111 | 0.192  |
| SOC content                            | 0.194        | 0.394  | -0.549 | 0.200  |
| ECEC                                   | 0.186        | 0.341  | 0.717  | 0.113  |

**Table 2.6.** Percentage, cumulative variances and eigenvectors on the first four principal components for 18 characters in 7 treatments.

SOC: Soil organic carbon; ECEC: Effective cation exchange capacity.

#### 2.2.3. Nutrient leaching

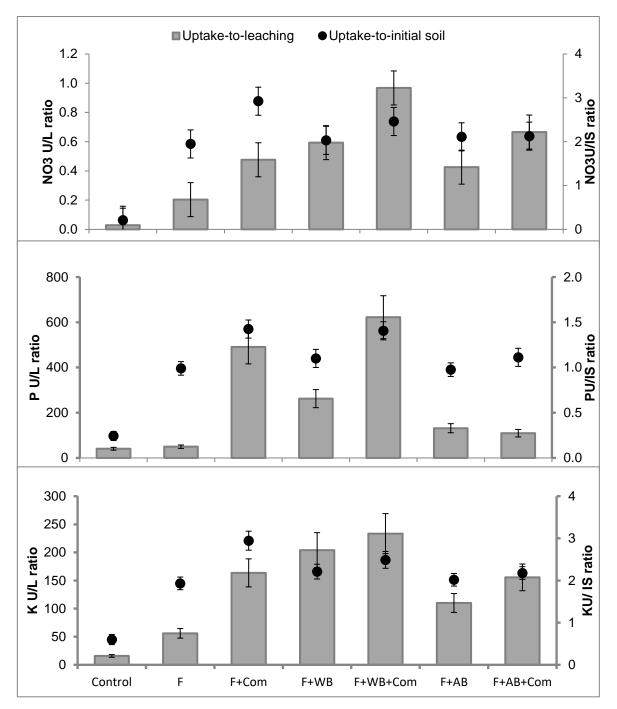
Leaching of NO<sub>3</sub>-N, P, K, Ca, Mg and Na significantly (p < 0.01) differed among the treatments (Table 2.7). Applications of F + WB + Com, and F + WB, significantly reduced the cumulative leaching of NO<sub>3</sub>-N, P, exchangeable K, Ca and Mg (Table 2.7). The greatest NO<sub>3</sub>-N and P leaching was recorded from mineral-fertilized soil. On the other hand, leaching of K, Ca, Mg and Na were the highest for the control treatment, followed by mineral-fertilized soil (Table 2.7). Fertilizer + WB +

Com, and F + WB, had less cumulative leaching of nutrients than other treatments. Most native available NO<sub>3</sub>-N was leached from the control during early crop establishment period or until 21 days after crop emergence. Significantly higher NO<sub>3</sub>-N uptake, P-uptake and K uptake-to-leaching ratios were obtained from F + WB + Co treated soil (Figure 2.5). In contrast, F + Co resulted in the maximum NO<sub>3</sub>-N uptake and K uptake-to-initial soil ratios, while F + Com and F + WB + Com treatments had similar P uptake-to-initial soil ratios. Nutrient uptake-to-leaching and initial soil ratios were the lowest for the control treatment followed by mineral-fertilized soil (Figure 2.5). There was a positive, significant linear correlation between the amount of leaching of nutrients and the volume of leachate. Marked differences were observed among treatments in the magnitude of leaching of nutrients. Overall, as the growth of plants progressed, the leaching of nutrients was markedly reduced because of higher nutrient uptake by the plants and smaller amounts left to leach.

|      | P<br>0.37 d                         | K  | Ca  | Mg   | Na   |
|------|-------------------------------------|--|---|--|--|
|      | 0.37 d                              | 22.0   |   |  |  |
|      |                                     | 22.8 a   | 32.7 a  | 7.2 a  | 4.96 a   |
| .6 a | 1.23 a                              | 20.7 b   | 24.1 b  | 5.6 b  | 2.34 b   |
| .7 c | 0.18 f                              | 10.8 c   | 23.5 b  | 4.4 c  | 2.57 b   |
| 7 ef | 0.26 e                              | 6.5 e  | 10.3 d  | 2.3 d  | 0.75 d   |
| .6 f | 0.14 g                              | 6.4 e  | 9.4 d   | 2.2 d  | 1.04 cd  |
| .4 d | 0.46 c                              | 11.0 c   | 16.5 c  | 3.7 c  | 1.37 c   |
| .9 e | 0.63 b                              | 8.4 d  | 10.9 d  | 2.6 d  | 1.22 cd  |
| ***  | ***                                 | **   | ***   | **   | **   |
| 0.1  | 0.04                                | 1.7  | 3.0   | 0.86   | 0.41   |
| 9.2  | 5.8                                 | 8.9  | 11.1  | 14.5   | 13.7   |
|      | 6.6 f<br>.4 d<br>.9 e<br>***<br>0.1 | 6.6 f       0.14 g         .4 d       0.46 c         .9 e       0.63 b         ***       ***         .0.1       0.04 | 6.6 f       0.14 g       6.4 e         .4 d       0.46 c       11.0 c         .9 e       0.63 b       8.4 d         ***       ***       **         0.1       0.04       1.7 | 6.6 f       0.14 g       6.4 e       9.4 d         .4 d       0.46 c       11.0 c       16.5 c         .9 e       0.63 b       8.4 d       10.9 d         ****       ***       **       ***         0.1       0.04       1.7       3.0 | 6.6 f       0.14 g       6.4 e       9.4 d       2.2 d         .4 d       0.46 c       11.0 c       16.5 c       3.7 c         .9 e       0.63 b       8.4 d       10.9 d       2.6 d         ***       ***       ***       **         0.1       0.04       1.7       3.0       0.86 |

**Table 2.7.** Effect of treatments on cumulative loss of nutrients by leaching at the end of the experiment.

Significant at  $p^* < 0.01$ ,  $p^* < 0.001$ . Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation.



**Figure 2.5.** The ratios of NO<sub>3</sub>-N, P and K uptake to their respective cumulative leaching, and initial soil contents as influenced by the treatments (n = 7). Note: NO<sub>3</sub>, P and K U/L Nitrate, phosphorus and potassium uptake to leaching ratio; NO<sub>3</sub>-N, P and K U/IS Nitrate, phosphorus and potassium uptake to initial soil ratio. Error bars represent  $\pm 1$ SE.

#### 2.3. Discussion

#### 2.3.1. Plant growth and nutrient uptake

Our results showed significant increases in plant growth and biomass production because of F + Comand F + WB + Com additions in comparison to the control and fertilizer only; the effects were due to improved availability of water and nutrients. In contrast, F + AB treatment was no better than fertilizer treatment alone in biomass production as well as N, P and K uptake. Shoot and root biomass increments were higher as a result of compost addition compared to the biomass obtained with either biochar type, indicating that the amount applied and the nutrients supplied by compost were adequate. Other studies have shown that application of compost increased oat biomass (Schulz and Glaser 2012), shoot and root biomass of ryegrass (Khan and Joergensen, 2012), and rice and cowpea biomass (Lehmann et al., 2003). There were also significant effects due to biochar type; higher total biomass was obtained from WB than AB. This may be due to WB having a higher nutrient retention capacity because of greater pore spaces and the ability to supply plants with more nutrients than AB, which may have contributed to the plant growth differential. For instance, the addition of 20 g WB to the pot added an extra 40 mg per pot P, nearly doubling the quantity of P applied as fertilizer. The effect of biochar on soil physical and chemical properties depends on feed-stock type and pyrolysis conditions (Novak et al., 2009b). Singh et al. (2010b) indicated that wood biochars had higher total C, but lower contents of ash, total N, P, K, S, Ca, Mg, Al, Na, Cu and CEC, than manure-based biochars.

Both positive and negative yield responses have both been reported for a wide variety of crops as a result of biochar application to soils (Chan and Xu, 2009; Tammeorg et al., 2014). For instance, maize yield increased by 98-150%, and water use efficiency by 91-139%, due to manure biochar addition (Uzoma et al., 2011). Furthermore, there was a 250% increase in wheat plant biomass following charred paper mill waste addition (Van Zwieten et al., 2010) and 18% increase in wheat grain yield from the use of oil mallee biochar (Solaiman et al., 2010). Plant growth and yield increases with biochar additions have been, in most cases, attributed to the optimization of the availability of plant nutrients (Gaskin et al., 2010; Glaser et al., 2002; Lehmann et al., 2003), increases in soil microbial biomass and activity (Biederman and Harpole, 2013; Thies and Rillig, 2009), and reduction of exchangeable Al<sup>3+</sup> (Glaser et al., 2002; Steiner et al., 2007). Likewise, wood biochar addition increased wheat yield by up to 30%, with no differences in grain N content, and sustained yield for two consecutive seasons without biochar addition in the second year (Vaccari et al., 2011). Major et al. (2010) reported that maize grain yield did not increase significantly in the first year following

addition of 20 t ha<sup>-1</sup> biochar (biomass-derived black carbon), but increased by 28, 30 and 140% over the control for the three consecutive years. Gathorne-Hardy et al. (2009) also reported that biochar addition alone did not show a significant effect on barley yield, but applications of 50 t biochar ha<sup>-1</sup> and 80 kg N fertilizer ha<sup>-1</sup> increased barley grain yield by 30%, which could be attributed to increased N use efficiency.

The application of compost and biochar singly or in combination with fertilizer enhanced chlorophyll content and specific leaf weight over mineral fertilizer alone. This could also be an indication of increased nutrient availability, vigorous plant growth, and healthier plants, resulting in higher plant biomass. Chlorophyll content, an indicator of photosynthetic activity, is related to the N concentration in green plants and serves as a measure of the response of crops to N fertilizer application and soil nutrient status (Minotta and Pinzauti, 1996). Hua et al. (2012) reported that application of bamboo biochar increased chlorophyll contents of ryegrass by 20-32% compared to the control. Application of F + WB + Com resulted in the production of leaves with higher specific leaf weight (+0.68 and  $0.32 \text{ mg cm}^{-2}$ ) than those produced under the control and mineral fertilizer only, respectively. Specific leaf weight is related to leaf resistance or susceptibility to insect attack, with higher specific leaf weight conferring resistance (Steinbauer, 2001).

Without amendment, the nutrient contents of the soil used in this study was extremely low. Leaf P concentration in the control and fertilizer only treatments was lower than the established sufficiency range (0.20 - 0.50%). Sufficiency ranges are established standard values which are used as a reference to compare nutrient concentrations of plant parts collected at a specific stage of crop development. The sufficiency range for each nutrient is based on a particular stage of growth and a specific plant part of each crop. Phosphorus concentration of maize plants in the F + WB + Com treatment was not higher than that in the F + Com treatment, thereby WB had no effect on plant P concentration. Fertilizer + Com and F + WB + Com considerably improved nutrient content of maize plants per unit of root biomass. Higher P uptake by the crop implies that a higher concentration of P was maintained in the soil solution and available to the plants. It could also imply that due to a greater sink for P the pool was just depleted and replenished faster. The organic amendments in this study may have made the soil more porous and friable, which potentially enhanced root growth and development. This could have improved the root-nutrient contact in the soil and optimized nutrient availability and uptake by plants, as the effect of the amendments was particularly noticeable on root growth. Higher plant nutrient uptake was accompanied by increased shoot and root biomass.

#### 2.3.2. Soil characteristics

Applications of biochar and compost had a significant influence on soil water content and chemical characteristics. Post-harvest SOC content of the mineral fertilized soil was lower than in the control, suggesting that application of mineral fertilizer alone may exacerbate the depletion of SOM through accelerated decomposition and mineralization relative to organic inputs. Soils treated with biochar had higher SOC, and SOC remained more stable, than soils treated with manure (Sukartono et al., 2011). Similarly, charcoal amended soil lost 8 and 4% SOC for mineral fertilized and unfertilized plots compared to losses of SOC from compost amended (27%) and control plots (25%), as well as reduced exchangeable Al (Steiner et al., 2007). While F + Com, F + WB + Com, and F + AB + Comhad a slight effect on soil C: N ratio, F + WB increased the C: N ratio by the largest margin compared to the initial value (14.2:1), which was reflected in the high SOC in the soil treated by F + WB. This might possibly be due to insignificant mineralization during the growing period, so the increase in SOC could be proportional to the amount added. In contrast, mineral fertilizer had negative effect on the C: N ratio compared to the initial value. Steiner et al. (2008) reported that biochar and compost amendment increased the soil C: N ratio after two consecutive harvests. In this study, compost and biochar additions significantly improved soil quality, including increases in SOC, exchangeable cations and water retention.

Biomass yield was significantly lower in the F + WB + Com treatment than in the F + Com treatment. However, the SOC increase in F + Com and F + WB + Com treated soil improved nutrient contents and nutrient availability compared to other treatments, which was reflected in plant growth and biomass yield. A possible explanation for this could be the fact that increasing total SOC by compost or compost and biochar additions might have increased reactive surfaces and stimulated microbial growth, which may lead to a short-term immobilization of plant-available nutrients. In the course of plant growth, these nutrients might be released through mineralization of compost and dead microorganisms, thus leading to improved plant growth over the course of the experiment. Schulz and Glaser (2012) reported that application of biochar with compost resulted in better plant growth and C-sequestration than biochar with mineral fertilizer. In biochar and biochar + Com, a significant part of their initial total C content remained after the second harvest, whereas only 58% remained in the biochar + F treatment. Nevertheless, in contrast to total C, biochar C contents remained almost constant during two crop growth periods without further biochar additions, but the mineral fertilizer only reduced the black C content to 75% of the original amount (Schulz and Glaser 2012).

Compost and biochar additions directly influenced the availability of native or applied nutrients, which significantly contributed to the increase in available soil K, Mg, ECEC, NO<sub>3</sub>-N and NH<sub>4</sub>-N after harvest, compared to the control and fertilizer only. Increase in total N would be simply due to

the N in the compost. Enhanced crop growth in the compost treated soil in this study was largely because of improved nutrient availability and greater uptake of N and P. Higher soil macro- and micro-nutrient contents due to compost and biochar additions may have been beneficial to plant performance, as compost and willow biochar contains significant amounts of essential elements. In contrast, despite low N content of biochar, positive (Berglund et al., 2004; DeLuca et al., 2009), negligible, or negative effects (Dempster et al., 2012) on nitrification rates have been reported, depending on soil pH (Yao et al., 2011). Liu et al. (2012) showed that compost and biochar addition increased total SOC, plant-available Ca, K, P, and Na contents by 2.5, 2.2, 2.5, 1.2 and 2.8 times, respectively, and increased the soil pH value by up to 0.6 and doubled plant-available soil water retention', in comparison to the control.

Soils fertilized with compost or manure have higher SOM content, porosity, hydraulic conductivity and aggregate stability, and are more enriched in P, K, Ca and Mg (Edmeades, 2003), as well as having lower bulk density (Tammeorg et al., 2014) relative to fertilized soils. CEC is a measure of the soils' ability to hold cations, which is associated with clay and SOM content (Troeh and Thompson, 2005). The ECEC of the experimental soil in this study was low, which might have caused higher leaching in mineral fertilized than organic amended soil during the plant growth period, owing to lower retention capacity of applied nutrients. In this study, the ratio of Ca to Mg showed variations for different treatments. The ratio of Ca to Mg had a significant influence on soil chemical properties and nutrient availability (Hazelton and Murphy, 2007). Because the availability of one of them in excessive amount in the soil may induce the deficiency of either of them. For instance, the excessive use of lime causes deficiencies of K, Mg, Fe, Mn, Zn, or Cu (Jones Jr, 2003).

Shoot biomass showed positive significant correlations with plant height, chlorophyll content, root biomass, specific leaf weight, soil water content, plant available nutrients and uptake. The direct effect of available soil nutrients and plant nutrient uptake from F + Com, F + WB, or F + WB + Com treated soil exceeded the direct effect of available nutrients and nutrient uptake from F + AB and fertilizer only treatments in terms of the performance of maize growth and biomass production. Correlation of plant biomass with plant nutrient uptake was the highest, and the compost and biochar amendment facilitated availability of these nutrients in this study. Positive associations between soil available nutrients, denoting that these nutrients were most limiting and corresponding with the soil P and NO<sub>3</sub>-N had a more significant influence on the shoot and root biomass than other nutrients, denoting that these nutrients were most limiting and corresponding with the soil P and NO<sub>3</sub>-N how and uptake by plants. A recent study has also shown positive linear correlations between soil chemical characteristics (K, P, EC, CEC, NO<sub>3</sub>, NH<sub>4</sub> and S), wheat seed

germination, root and shoot growth, as a result of addition of different biochar types, but negative correlation between seed germination and Al content (Solaiman et al., 2012). From this result, it can be inferred that high shoot and root biomass, high chlorophyll content, and taller plant height are the traits associated with the performance of maize.

Principal component analysis (PCA) indicated that the first component (PRIN1) provided a reasonable summary of the data, accounting for about 84% of the total variance. In other words, PRIN1 explained most of the variation in the entire data set, and the other three, most of the remaining variation. It is normally believed that characters with larger absolute values closer to unity within the first principal component influence the clustering more than those with lower absolute values closer to zero (Jolliffe, 2002). In this study, however, almost all characters in the first eigenvector individually contributed similar effects to the total variation of the treatments, suggesting that the first component was primarily a measure of the whole characters. Thus, the differentiation of the treatments into different clusters was rather dictated by the cumulative effects of several characters. Similarly, Sena et al. (2002) compared conventionally managed plots that intensively utilized pesticides and chemical fertilizers, with non-disturbed forest areas and alternatively managed plots, using PCA to visualize the effects of alternative soil amendment.

#### 2.3.3. Nutrient leaching

Differences in cumulative water percolation and extent of nutrient leaching among treatments were caused by variations in plant water uptake and efficiency of nutrient retention. Leaching of nutrients was significantly reduced, owing to compost and biochar amendment compared to the control and mineral fertilized soil, supporting our third objective. Fertilizer + WB + Co had more impact in lowering the cumulative leaching of nutrients than F + AB or F + AB + Com, possibly due to more pore spaces and increased sorption capacity of biochars through oxidative reactions on the biochar surfaces over time. Singh et al. (2010b) have shown the reduction of leaching of NH<sub>4</sub>-N by 55-93% from the applications of manure and wood biochars on different soil types. Losses of NO<sub>3</sub>-N through leaching range from 0 to 60% of the applied N fertilizer (Meisinger and Delgado, 2002).

Marked differences were observed among treatments in the magnitude of leaching of nutrients at the end of the experiment. A similar study showed that charcoal application decreased the proportion of leached N and Ca on Ferralsols (Lehmann et al., 2003). While N and K proved very mobile in soil, application of F + WB and F + WB + Com reduced leaching of NO<sub>3</sub>-N by 66 and 73%, and K by 68 and 69%, compared to fertilizer only treated soil, respectively. Thus, the retention of N and K should

be specifically targeted with additions of slow-releasing nutrients and/or soil amendments. Leaching occurs at much slower rate when nearly all the ions are present in an exchangeable form (Troeh and Thompson, 2005), or when uptake by plants increases (Lehmann et al., 2003). Low leaching at high nutrient availability, as found in this study, ensures sustainable soil fertility, which coincides with the findings of Lehmann et al. (2003). Soils which have been strongly weathered and leached often have low levels of exchangeable Ca and Mg, and plant growth may be nutrient limited as a result (Glaser et al., 2002). A recent study has indicated that biochar significantly reduced the leaching of NH<sub>4</sub>-N (by 12 - 86%), NO<sub>3</sub> (by 26 - 95%), basic cations, P, and certain micronutrients (Sika, 2012). In contrast, Novak et al. (2009a) reported higher EC, K and Na concentrations of leachates, but lower concentrations of Ca, P, Mn and Zn. The degree of leaching of cations, such as Ca, Mg and Na, was directly related to the differential nutrient retention capacity of treatments, as the availability of these nutrients for plants was directly dependent on the soil reserve. Although the initial soil K status was sufficient for plant growth, the K uptake to initial soil K was the lowest for the control since K-uptake might be limited by the low availability of other nutrients, such as N and P.

#### 2.4. Conclusions

This study showed that the experimental soil was deficient in plant available nutrients, consistent with the general observation that Ferralsols of the humid tropics are nutrient depleted and suboptimal for plant growth, without additions of organic and inorganic amendments. Applications of F + Com, F + WB, or F + Com + WB were more effective in improving soil carbon, soil moisture, and nutrient retention capacity, relative to mineral fertilizer alone. The use of compost and biochar as soil amendments reduced the loss of some nutrients through leaching, with nutrient leaching of NO<sub>3</sub>-N, P, and exchangeable bases significantly decreased as the growth of plants progressed. Although F + Com + WB, or AB, did not outperform F + Com in terms of biomass yield and nutrient uptake, the combined application of compost and biochar, especially WB may enhance and sustain soil biophysical and chemical characteristics, since most of the compost will disappear over time through decomposition, whereas the biochar is likely to stay in the soil for decades. Root biomass was significantly increased by F + Com, F + WB, and F + WB + Com amendments, compared to other treatments in this study. Further long-term research is required to evaluate and quantify the benefits and spillover effects of these amendments in terms of improving and sustaining soil fertility, crop productivity and economic returns to users. Moreover, despite a number of positive short-term research results, the amount of recalcitrant C supplied by biochar and compost that is sequestered in the soil needs to be determined through long-term field experiments.

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# **Overview of Chapter 3**

This chapter is based on a paper published in the *Science of the Total Environment* with minimal formatting changes. I have investigated the effects of compost, biochar, compost, biochar-compost mix and co-composted biochar-compost on the enhancement of maize growth, yield and nutrient uptake, soil physicochemical properties, and greenhouse gas emissions in an important maize-growing climate and soil type. The trial site was located at Tolga, North Queensland, Australia on a Ferralsol.

The study was a field trial, designed to determine whether soil quality and plant productivity could be enhanced, and greenhouse gas emissions reduced by biochar and compost, applied singly or together and with inorganic fertilizer. I used willow biochar (WB; Earth Systems Pty Ltd, Melbourne, Vic.) produced at a temperature of 500°C as soil amendment. I collected rigorous plant and soil data throughout the study period. I organized, analyzed and interpreted the data, and wrote the manuscript.<sup>3</sup>

<sup>&</sup>lt;sup>3</sup> Agegnehu, G., Bass, A. M., Nelson, P. N., and Bird, M. I. (2016). Benefits of biochar, compost and biochar–compost for soil quality, maize yield and greenhouse gas emissions in a tropical agricultural soil. *Science of the Total Environment* **543**, 295-306.

# Chapter 3

# Benefits of Biochar, Compost and Biochar-Compost for Soil Quality, Maize Yield and Greenhouse Gas Emissions in a Tropical Agricultural Soil

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#### Abstract

Soil quality decline represents a significant constraint on the productivity and sustainability of agriculture in the tropics. In this study, the influence of biochar, compost, and mixtures of the two on soil fertility, maize yield and greenhouse gas (GHG) emissions was investigated in a tropical Ferralsol. The treatments were: 1) control with business as usual fertilizer (F); 2) 10 t ha<sup>-1</sup> biochar (B) + F; 3) 25 t ha<sup>-1</sup> compost (Com) + F; 4) 2.5 t ha<sup>-1</sup> B + 25 t ha<sup>-1</sup> Com mixed on site + F; and 5) 25 t ha<sup>-1</sup> <sup>1</sup> co-composted biochar-compost (COMBI) + F. Total aboveground biomass and maize yield were significantly improved relative to the control for all organic amendments, with increases in grain yield between 10-29%. Some plant parameters, such as leaf chlorophyll, were significantly increased by the organic treatments. Significant differences were observed among treatments for the  $\delta^{15}N$  and  $\delta^{13}C$ contents of kernels. Soil physicochemical properties, including soil water content (SWC), total soil organic carbon (SOC), total nitrogen (N), available phosphorus (P), nitrate-nitrogen (NO<sub>3</sub><sup>-</sup>-N), ammonium-nitrogen ( $NH_4^+$ -N), exchangeable cations, and cation exchange capacity (CEC) were significantly increased by the organic amendments. Maize grain yield was correlated positively with total biomass, leaf chlorophyll, foliar N and P content, SOC, and SWC. Emissions of CO2 and N2O were higher from the organic-amended soils than from the fertilizer-only control. However, N<sub>2</sub>O emissions generally decreased over time for all treatments and emission from the biochar was lower compared to other treatments. Our study concludes that the biochar and biochar-compost-based soil management approaches can improve SOC, soil nutrient status and SWC, and maize yield and may help mitigate greenhouse gas emissions in certain systems.

**Keywords**: Biochar, co-composted biochar-compost, compost, Ferralsol, greenhouse gas fluxes, soil quality

## 3.1. Introduction

Soil nutrient depletion, declining agricultural productivity and anthropogenic climate change are threatening the sustainability of agricultural production in the tropics (Gruhn et al., 2000b; Parry, 2007; Pender, 2009). The benefits of inorganic fertilizers have been widely demonstrated since the 'green revolution' (Vanlauwe et al. 2010), and have played a significant role in increasing agricultural production and productivity over the last half century (Gruhn et al., 2000b). Shrinking land area per capita and declining soil quality have led to steady increases in fertilizer use. However, the application of inorganic fertilizer alone is not a sustainable solution for improving soil fertility and maintaining yields. Rather, it has been widely realized that application of excessive inorganic fertilizer, especially

nitrogen, may cause soil deterioration and other environmental problems owing to more rapid organic matter mineralization (Liu et al., 2010; Palm et al., 2001).

In most tropical environments, sustainable agriculture faces constraints due to low nutrient status and rapid mineralization of soil organic matter (Jenkinson et al., 1991; Zech et al., 1997). As a result, the cation exchange capacity (CEC) of the soils, i.e., the mineralogy of which is dominated by low-CEC clays, is further decreased. Under such conditions, the efficiency of mineral fertilizers is very low as mobile nutrients, such as nitrate-nitrogen (NO<sub>3</sub><sup>-</sup>-N) or potassium (K<sup>+</sup>), are readily leached from the topsoil during periods of high rainfall (Socolow, 1999). Additionally, costs of inorganic fertilizers can be prohibitive in developing countries (Sanchez, 2002). Consequently, nutrient deficiency is prevalent in many crop production systems of the tropics. Although the green revolution over the last four decades has fed the growing global population, some production practices have become increasingly unsustainable, environmentally damaging, and unlikely to keep up with demand (Barrow, 2012). Thus, there is a need for a new approach to increased yields, reduce negative impacts, and enhance sustainability, with new approaches being accessible to subsistence farmers as well as commercial producers (Glaser, 2007; Lal, 2008; Sohi et al., 2010).

In recent years, application of biochar to soil has emerged as a potential strategy for sequestering carbon, reducing greenhouse gas (GHG) emissions and improving soil quality (Lehmann et al., 2006; Liang et al., 2014; Vaccari et al., 2011). Evidence shows that the application of biochar can play a significant role in improving SOC (Glaser et al., 2002), soil water holding and plant-available water capacity (Abel et al., 2013; Atkinson et al., 2010; Hansen et al., 2016), soil aeration, increased soil base saturation, nutrient retention and availability, decreasing fertilizer needs and nutrient leaching (Laird, 2008; Lehmann et al., 2003; Steiner et al., 2007), stimulation of soil microbes, increased microbial biomass and activity (Thies and Rillig, 2009), enhancing crop growth and yield, reducing anthropogenic GHG fluxes, and increasing carbon sequestration (Lal, 2011a; Lehmann et al., 2006). Carbon sequestration in soil is favored for the additional reasons of improving soil quality and achieving sustainable use of natural resources (Lal, 2008, 2011a).

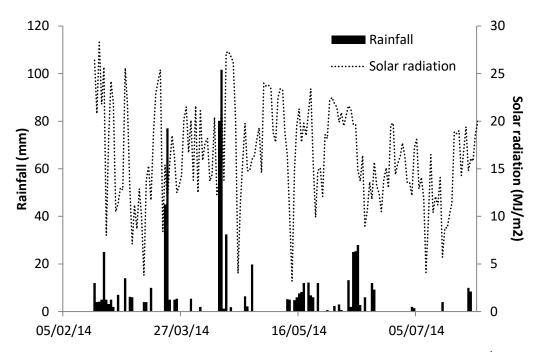
Maize (*Zea mays* L.) is an important crop globally, being grown on a wide variety of soil types and in a wide range of climates. In Australia, 62,200 ha were planted with maize in 2010 - 2011 at an average yield of 6.0 t ha<sup>-1</sup> (ABS, 2012). Under irrigation, and with intensive fertilizer inputs, yields of 15 t ha<sup>-1</sup> or more are possible. The area that could potentially be planted with maize in northern Australia is considerably larger than currently utilized, although water availability is a major constraint (Chauhan et al., 2013). The yield and quality of maize are affected by soil type, water availability and nutrition (Bossio et al., 2010; Chauhan et al., 2013). Maize responds positively to N, P and K inputs, with yield also increased by liming of acid soils (Aitken et al., 1998). In north Queensland, inputs of up to 80 kg ha<sup>-1</sup> N, 35 kg ha<sup>-1</sup> P and 50 kg ha<sup>-1</sup> K may be required for optimal yield, along with zinc (Zn), sulfur (S) and molybdenum (Mo) in some cases. Moisture deficit during growth can reduce leaf size and number, consequently leading to low grain yield through reduced capacity for photosynthesis. Lobell et al. (2013) recently noted the vulnerability of maize production globally to climate change and linked observed yield declines in the last two decades to more frequent exposure of crops to temperature extremes, which in turn imposes additional water stress on crops. On the other hand, using the same data, Basso and Ritchie (2014) concluded that soil moisture deficit has been directly responsible for observed yield declines.

There has been little work on the impact of organic amendments in combination with biochar on maize growth and yield. Lashari et al. (2015) found significant improvement in soil properties, plant performance and maize yield due to the use of manure composts with biochar and pyroligneous solution on a saline soil in China, over a two year period, with beneficial impacts of increasing over time. Nur et al. (2014) demonstrated that maize biomass and yield were more than doubled over two crop cycles using compost and biochar in combination on a calcareous soil in Indonesia. Similarly, promising results have been obtained from the use of compost-biochar combinations on low fertility soils in Laos (Mekuria et al., 2014). In contrast to the above studies on tropical soils, Lentz et al. (2014) found only small impacts of biochar-compost applications on maize in a temperate climate. However, in all the above cases, the use of biochar and compost, alone and in combination, generally improved soil characteristics. The emerging picture for biochar and compost amendment use under maize is one of potentially significant benefits for degraded soils in the tropics, both in terms of the soils' properties and maize yield, but lower benefits to yield in temperate environments. This general assessment is dependent in detail on interactions between biochar, soil, crop, climate and time. The aims of this study were, therefore, to test the hypotheses that application of biochar, compost and biochar-compost mixes: 1) enhances maize growth, yield and nutrient uptake; 2) enhances soil physicochemical properties; and 3) mitigates greenhouse gas emissions in an important maizegrowing climate and soil type.

## 3.2. Materials and methods

## 3.2.1. Trial site description and soil analysis

The trial site was located at Tolga, north Queensland, Australia (17.2191°S 145.4713°E; 778 m above sea level). The soil is a dark reddish brown Ferralsol (IUSS 2007), or Red Ferrosol (Isbell (1996), of the Tolga series (Malcolm et al. 1999) developed on Quaternary basalt, grading from light clay at 0-0.2 m depth to medium clay at 0.5 - 1.0 m depth. Daily weather data for this station over the trial period are shown in Figure 3.2. The long-term mean annual precipitation is 1,032 mm, and mean annual minimum and maximum temperatures are 17.1 and 27.3°C, respectively (Walkamin Research Station: 17.1347 °S; 145.4281°E; 594m above sea level). Pre-planting soil samples were collected in December 2013, from depths of 0 - 30 cm and 30 - 100 cm from nine locations across the trial site. The locations were selected by dividing the trial area into a 3 m  $\times$  3 m grid and randomly selecting three sampling points within each of the nine grid cells. One core was taken from each point, to a depth of 0 - 30 cm, and these samples were combined into one composite sample for each grid cell. One core of 30 - 100 cm depth was also taken from each grid cell using a vehicle mounted hydraulic corer. Soil samples returned from the field were weighed, dried at 60°C and then reweighed to determine water content, which were used to calculate soil bulk density based on corer dimensions and core length. After manual homogenization, approximately 1kg of dry soil was then passed through a soil mill with 2mm screen. Material >2mm was weighed and discarded. The <2 mm fraction 'fine earth' fraction, was sealed in a labelled Ziploc plastic bag. A  $\sim 10$  g sub-sample of this material was weighed, dried at 105°C and then ball milled to a powder. Soil samples were analyzed by SGS Pty Ltd, Cairns, Australia (0 – 30 cm), or Analytical Research Laboratories (ARL) Pty Ltd, Awatoto, New Zealand (30 - 100 cm). Soil water content (SWC) was measured at 0 - 12 cm depth using a Campbell Scientific Hydrosense II soil moisture probe at each sampling location. The gravimetric SWC of each sample was also measured. Soil profiles were described and classified from 50 mm diameter cores to approximately 1 m depth (one core at each trial site), according to NCST (2009).



**Figure 3.1.** Rainfall and solar radiation recorded at daily intervals between the 18<sup>th</sup> of February 2014 and the 30<sup>th</sup> of July 2014.

Pre-planting physicochemical properties of the trial soil are shown in Table 3.1. This moderately acidic clay soil (0 - 30 cm) had a comparatively high organic matter content and moderate soil pH, exchangeable K and Cu, but low electrical conductivity (EC) and cation exchange capacity (CEC). Soil cores from 0 - 30 cm were taken in the row at the mid-season growth stage and after harvesting and analyzed as described above. SOC and total soil N contents were determined as they were for plants. Soil pH, exchangeable cations, CEC, EC, NO<sub>3</sub><sup>-</sup>N and NH<sub>4</sub><sup>+</sup>-N contents were determined by the ARL Pty Ltd. Soil pH was measured in H<sub>2</sub>O and 0.01 M CaCl<sub>2</sub>, using pH meter and a 1:2.5 soil weight to extractant-volume ratio. The EC was determined by a conductivity meter on a 1:2.5 soil: water suspension (Rayment and Higginson, 1992). Colwell P was measured in 1:50 soil solution extracts in 0.5 M sodium bicarbonate after end-over-end shaking for 16 hrs. The extracted P was determined colorimetrically on centrifuged and filtered extracts using a SEAL AQ2 + Discreet Analyzer (Seal Analytical Ltd, Fareham, Hampshire, UK), and the ammonium molybdate/ascorbic acid color reaction with potassium antimonyl tartrate was added to control the reaction rate (Rayment and Lyons, 2011). Exchangeable K, Na, Ca and Mg were determined using 1M ammonium acetate extraction buffered at pH 7, using mechanical shaking, at a soil: solution ratio of 1:20 (Rayment and Higginson, 1992) and atomic absorption analysis. CEC was calculated as the sum of exchangeable K, Na, Ca and Mg. Soil NO<sub>3</sub>-N and NH<sub>4</sub><sup>+</sup>-N were determined colorimetrically by an automated photometer using 1M KCl extraction method (Rayment and Lyons, 2011).

| Limit of                |                          |           |         |      |           |      |  |  |  |  |  |  |  |
|-------------------------|--------------------------|-----------|---------|------|-----------|------|--|--|--|--|--|--|--|
| Parameters              | Unit                     | detection | 0-30 cm | SE   | 30-100 cm | SE   |  |  |  |  |  |  |  |
| Water content           | %w/w                     |           | 19      | 0.28 | 21.7      | 0.50 |  |  |  |  |  |  |  |
| pH (H <sub>2</sub> O)   | Units                    |           | 5.6     | 0.09 | 6.3       | 0.03 |  |  |  |  |  |  |  |
| pH (CaCl <sub>2</sub> ) | Units                    |           | 5.1     | 0.08 | 6.3       | 0.04 |  |  |  |  |  |  |  |
| Total nitrogen          | %w/w                     |           | 0.14    | 0.01 | 0.04      | 0.01 |  |  |  |  |  |  |  |
| Total carbon            | %w/w                     |           | 2.01    | 0.07 | 0.61      | 0.15 |  |  |  |  |  |  |  |
| C: N ratio              |                          |           | 15.5    | 0.3  | 15.4      | 1.4  |  |  |  |  |  |  |  |
| Carbon stock            | t ha <sup>-1</sup>       |           | 37.6    | 2.5  | 30.7      | 7.0  |  |  |  |  |  |  |  |
| Colwell P               | mg kg <sup>-1</sup>      | 1.00      | 17.3    | 2.1  | 9.6       | 1.40 |  |  |  |  |  |  |  |
| NH4-N                   | mg kg <sup>-1</sup>      | n/d       | n/d     | n/d  | 7.7       | 1.10 |  |  |  |  |  |  |  |
| NO <sub>3</sub> -N      | mg kg⁻1                  | n/d       | n/d     | n/d  | 20.9      | 2.12 |  |  |  |  |  |  |  |
| Conductivity            | uS cm <sup>-1</sup>      | 1.00      | 0.13    | 0.01 | 0.05      | 0.00 |  |  |  |  |  |  |  |
| Exchangeable Na         | cmol(+) kg <sup>-1</sup> | 0.01      | 0.06    | 0.0  | 0.03      | 0.00 |  |  |  |  |  |  |  |
| Exchangeable K          | cmol(+) kg <sup>-1</sup> | 0.01      | 0.58    | 0.11 | 0.12      | 0.01 |  |  |  |  |  |  |  |
| Exchangeable Ca         | cmol(+) kg <sup>-1</sup> | 0.01      | 3.9     | 0.45 | 2.5       | 0.23 |  |  |  |  |  |  |  |
| Exchangeable Mg         | cmol(+) kg <sup>-1</sup> | 0.01      | 0.87    | 0.07 | 0.60      | 0.05 |  |  |  |  |  |  |  |
| Exchangeable Al         | cmol(+) kg <sup>-1</sup> | 0.02      | 0.07    | 0.0  |           |      |  |  |  |  |  |  |  |
| CEC                     | cmol(+) kg <sup>-1</sup> | 0.02      | 5.4     | 0.47 | n/d       | n/d  |  |  |  |  |  |  |  |
| Total S                 | mg kg <sup>-1</sup>      | 1.00      | 37.4    | 6.3  | n/d       | n/d  |  |  |  |  |  |  |  |
| DTPA Cu                 | mg kg <sup>-1</sup>      | 0.05      | 1.7     | 0.14 | n/d       | n/d  |  |  |  |  |  |  |  |
| DTPA Zn                 | mg kg <sup>-1</sup>      | 0.05      | < 0.05  | 0.0  | n/d       | n/d  |  |  |  |  |  |  |  |
| DTPA Mn                 | mg kg <sup>-1</sup>      | 0.50      | 290     | 37   | n/d       | n/d  |  |  |  |  |  |  |  |
| DTPA Fe                 | mg kg <sup>-1</sup>      | 0.50      | 9.3     | 0.33 | n/d       | n/d  |  |  |  |  |  |  |  |
| Total B                 | mg kg <sup>-1</sup>      | 0.05      | 0.48    | 0.03 | n/d       | n/d  |  |  |  |  |  |  |  |

**Table 3.1.** Pre-planting soil properties from samples collected at depths of 0 - 0.30 cm and 30 - 100 cm in December, 2013 (n = 9;  $\pm$  SE).

n/d: Not determined

## 3.2.2. Trial set-up

The feedstock for biochar production was waste willow wood (*Salix spp*.), derived from removal and restoration activities. The biochar (B; Earth Systems Pty. Ltd.) was produced using a containerized automated batch pyrolysis plant (Charmaker MPP20). Processing of whole logs at up to 5 t per load

required over 5 - 7 hours with the highest heating temperatures of 550°C. The biochar was ground to < 10 mm prior to delivery in a Keenan spreader. Two paired compost windrows (each 60 m long, 1.5 m high and 4 m wide) were produced at the King Brown Technologies compost production facility: one containing compost and biochar (COMBI) and another one compost only. The biochar (equivalent to 18 m<sup>3</sup> by volume) was added to 80 m<sup>3</sup> each of green waste and bagasse, 12.5 m<sup>3</sup> of chicken manure and 12 m<sup>3</sup> of compost. This windrow was paired with an adjacent windrow comprising the same volumes of green waste (43%), bagasse (43%), chicken manure (7%) and compost (7%), but without biochar. In both cases, bagasse was laid down first, green-waste mulch added on top, and the windrow turned once. Then, chicken manure was added and the windrows watered and turned six times. In the case of the COMBI treatment, the biochar was then added on top of the windrow and the pile turned a further two times. Both windrows were then covered with black plastic film. They were turned and watered weekly, and the matured product was screened at 25 mm. After composting, the COMBI amendment contained ~20% biochar. Biochar, compost and COMBI samples were collected randomly from all materials before application in the field for nutrient analysis (Table 3.2).

The experiment comprised five treatments in triplicate, where each replicate occupied 0.13 ha (240 m long by 6 rows with a row spacing of 0.91 m), planted following a peanut crop. The treatments were: 1) fertilizer as a control (F); 2) willow biochar (B) applied at 10 t ha<sup>-1</sup>; 3) compost (Com) applied at 25 t ha<sup>-1</sup>; 4) 2.5 t ha<sup>-1</sup> B + 25 t ha<sup>-1</sup> Com mixed on site (B + Com); and 5) co-composted biochar-compost (COMBI) applied at 25 t ha<sup>-1</sup>. The experimental field was strip plowed and all amendments were applied before planting and being mechanically mixed into the upper 10 cm. Maize, variety ATW1 treated with vitavax fungicide, was planted at 6,000 seeds ha<sup>-1</sup> on 13 February 2014. Fertilizer was applied in the row as DAP (di-ammonium phosphate; 18% N, 20% P, 1.6%S) at 186 kg ha<sup>-1</sup>, as urea and muriate of potash across all treatments followed by Nipro FlowPhos 13Z (9% N, 13.5% P, 1%K, 0.9% Zn) at 30 L ha<sup>-1</sup>. In addition, Rutec Zn 7,000 (70%w/v Zn; 6.9%w/v N) was applied across all treatments at 0.3 L ha<sup>-1</sup> 21 days post emergence. This equated to a total fertilizer application of 150 kg ha<sup>-1</sup> N, 41 kg ha<sup>-1</sup> P, 120 kg ha<sup>-1</sup> K and 3 kg ha<sup>-1</sup> S. Other agronomic practices were applied during the crop growth period, as per usual farm practice. The total rainfall during the crop growing period was 723.2 mm.

| Element                        | Unit                | В     | Com    | COMBI  |
|--------------------------------|---------------------|-------|--------|--------|
| рН (H <sub>2</sub> O)          | Units               | 8.3   | 7.5    | 7.5    |
| pH (CaCl <sub>2</sub> )        | Units               | 9.5   | n/d    | nd     |
| Organic carbon (OC)            | %                   | 78    | 30.6   | 34.7   |
| Total nitrogen (N)             | %                   | 0.38  | 1.19   | 0.95   |
| $\delta^{15}N$                 | ‰                   | n/d   | +7.5   | +7.8   |
| $\delta^{13}C$                 | ‰                   | nd    | -24.3  | -21.3  |
| Sulfur (S)                     | %                   | 0.019 | 0.014  | 0.012  |
| Colwell phosphorus (P)         | mg kg <sup>-1</sup> | 79.5  | 917    | 1104   |
| Acid neutralizing capacity     | %CaCO3              | 2.5   | n/d    | nd     |
| Exchangeable potassium (K)     | cmol(+)/kg          | 7.25  | 1.62   | 1.74   |
| Exchangeable calcium (Ca)      | cmol(+)/kg          | 2.20  | 4.15   | 4.15   |
| Exchangeable magnesium (Mg)    | cmol(+)/kg          | 1.45  | 2.38   | 2.30   |
| Exchangeable sodium (Na)       | cmol(+)/kg          | 0.24  | 0.52   | 0.52   |
| Exchangeable aluminum (Al)     | cmol(+)/kg          | < 0.1 | < 0.1  | < 0.1  |
| Acidity                        | cmol(+)/kg          | n/d   | 0.3    | 0.11   |
| Cation exchange capacity (CEC) | cmol(+)/kg          | 11.2  | 8.77   | 8.81   |
| Electrical conductivity (EC)   | dS/m                | 0.71  | 2.3    | 2.0    |
| DTPA Copper (Cu)               | mg kg <sup>-1</sup> | 2.55  | 45.0   | 44.0   |
| DTPA Zinc (Zn)                 | mg kg <sup>-1</sup> | 83.5  | 133    | 133    |
| DTPA Manganese (Mn)            | mg kg <sup>-1</sup> | 110   | 49.6   | 54.6   |
| DTPA Iron (Fe)                 | mg kg <sup>-1</sup> | 0.045 | 246    | 218    |
| Boron (B)                      | mg kg <sup>-1</sup> | 9.25  | 4.8    | 4.3    |
| Molybdenum (Mo)                | mg kg <sup>-1</sup> | < 0.3 | < 0.2  | < 0.2  |
| Cobalt (Co)                    | mg kg <sup>-1</sup> | <0.4  | < 0.05 | < 0.05 |

**Table 3.2.** Characterization of Earth System willow biochar (B), compost (Com) and co-composted biochar-compost (COMBI).

CEC: Cation exchange capacity; EC: Electrical conductivity; n/d: Not determined.

## 3.2.3. Measurements during trial

After planting, periodic sampling and measurements of soil parameters, leaf chlorophyll, specific leaf weight, and emission of carbon dioxide ( $CO_2$ ) and nitrous oxide ( $N_2O$ ) were undertaken from all three

replicates of each treatment over all sampling dates. The measurements of CO<sub>2</sub> and N<sub>2</sub>O were undertaken on days 5, 14, 21, 43, 75, 98 and 145 after planting (18 February – 8 July 2014), in conjunction with SWC. SWC at 0 - 12 cm depth was taken at planting using a HydroSense II probe (Campbell Scientific, Inc.). CO<sub>2</sub> and N<sub>2</sub>O fluxes were measured with a standard closed chamber methodology, using an INNOVA 1412i field portable photoacoustic gas analyzer (LumaSense Technologies, Ballerup, Denmark). Build-up of gases in the headspace was measured over 4 - 8 minutes, depending on flux, at 45 second intervals. Duplicate soil collars (15 cm diameter) remained in place in each plot for the trial duration (n = 6 per treatment). The average GHG fluxes were calculated from all three replicates over all sampling dates of each treatment. Those values, therefore, encompassed the temporal and spatial variability. For total GHG flux over the trial period for each treatment was calculated from the area under each temporal curve via trapezoidal integration. Gas fluxes (F) in mg m<sup>-2</sup> h<sup>-1</sup> were calculated using the following equation:

$$F = \left(c \times \left(\frac{P}{1013}\right) \times \left(\frac{273}{T+273}\right) \times M \times \left(\frac{V}{A}\right) / 1000 \times 3600.$$
 (1)

Where c = rate of concentration increase (ppm) at t = 0, using a polynomial curve fit, P is the air pressure in the chamber, T is the ambient temperature within the chamber in  $^{\circ}C$ , M is the molar weight of the subject gas, V is the volume of the headspace and A is the area of the sample chamber. Total gas emitted over the trial was calculated using a simple trapezoidal integration method, assuming linear changes in flux rates between sample points.

The amount of GHG emission per unit yield was calculated as follows:

$$E = \frac{M}{Y}.$$
 (2)

Where M is the cumulative emissions of CO<sub>2</sub>-C and N<sub>2</sub>O-N; Y is the grain yield (kg ha<sup>-1</sup>).

Relative leaf chlorophyll content was measured using a SPAD-502 (Konica Minolta, Tokyo) based on transmittance at wavelength regions 650 nm (for chlorophyll) and 940 nm (as a control). The measurements were undertaken on days 38, 46, 58, 70, 88 and 102 after planting, in conjunction with the collection of leaf punch samples for the determination of specific leaf weight and leaf water content. For each measurement, duplicate readings were made on the second fully expanded leaf from the top of the main plant stem, approximately half way along the leaf, taking care to avoid veins and mid-rib. This procedure was repeated for six randomly selected plants from all three replicates of each treatment. A hole-punch with a diameter of 8 mm was used to take a leaf disc from the middle of the leaf lamella from leaves of the same age and position from each replicate. Leaf discs were transferred immediately to individual, sealed plastic bags that were kept in an insulated box above ice packs until processed. Leaf discs were weighed before being dried at  $60^{\circ}$ C and re-weighed. The number of plants and cobs per plant, per 5 m or row, were counted at physiological maturity. The crop was harvested on 17 June 2014. After harvest, above-ground biomass, grain yield and yield components of maize were determined. Total biomass and grain yield of maize recorded on plot basis was converted to kg ha<sup>-1</sup> for statistical analysis.

Ten leaf samples were clipped at their base prior to tasseling, and were stored for the determination of C, N, P, K and NO<sub>3</sub><sup>-</sup>-N concentrations. Total plant N and C concentrations were determined using an elemental analyzer (ECS 4010 CHNSO Analyzer; Costech Analytical Technologies INC, Valencia, CA, USA) fitted with a Zero Blank Auto-sampler (Costech Analytical Technologies, INC) at James Cook University's Advanced Analytical Centre, Cairns, Australia. Plant P, K and NO<sub>3</sub>concentrations were quantified at ARL. NO<sub>3</sub>-N in plant tissue was determined using 2% acetic acid as the extractant (Miller, 1998). Plant K concentration was determined after wet digestion with sulfuric acid, by atomic absorption spectrometry (Watson et al., 1990). Plant P concentration was determined photometrically in the same digest with the molybdenum blue method (Mills and Jones, 1996). Total N and C concentrations and isotopic composition of oven-dried maize grain samples were also determined using the ECS 4010 CHNSO Analyzer and a ThermoFinnigan DeltaVPLUS Continuous-Flow Isotope Ratio Mass Spectrometer (EA-IRMS) at James Cook University. Stable isotope results are reported as per mil (‰) deviations from the VPDB reference standard scale for  $\delta^{13}$ C and from the international air standard for  $\delta^{15}$ N. Precisions on internal standards were better than ±0.2‰ for both isotope determinations.

## 3.2.4. Data analysis

The data were subjected to analysis of variance using the general linear model procedure (PROC GLM) of SAS statistical package version 9.2 (SAS Institute 2008, Cary, NC). The total variability for each trait was quantified using the following model:

$$Y_{ij} = \mu + R_i + T_j + RT_{ij} + e_{ij}.....(3)$$

Where  $Y_{ij}$  is the measured value,  $\mu$  = grand mean,  $R_i$  is effect of the *i*<sup>th</sup> replication,  $T_j$  is effect of the *j*<sup>th</sup> treatment,  $RT_{ij}$  is the interaction, and  $e_{ij}$  is the variation due to random error. Means for the treatments (n = 5) were compared using the MEANS statement with the least significant difference (LSD) test at the 5% probability level. Correlation analyses were performed between plant parameters, nutrient uptake and soil nutrient contents using the SAS CORR procedure.

## 3.3. Results

## 3.3.1. Plant growth and yield

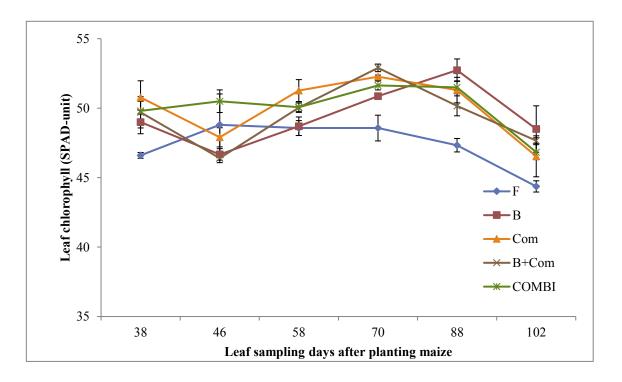
The application of biochar and compost individually, or in combination to fertilized soil, had positive effects on most of the measured plant parameters. The treatments yielded significantly (p < 0.01) higher total biomass and grain yield than the yields obtained from F only amended soils (Table 3.3). The addition of B + F resulted in the highest maize total biomass and grain yield (Table 3.3). In contrast, the maximum number of plants, 4.5 m<sup>-2</sup>, was recorded from the F only treatment (data not shown), in which the total biomass and grain yield were the lowest of all treatments. Compost and COMBI + F amended plots had higher 100-grain weights than other treatments, indicating these organic soil amendments had positive effects on grain filling. Compared to F alone, grain yield and total biomass were higher by 29, 10, 20 and 13% and 17.7, 14, 7.6 and 12%, respectively for B + F, Com + F, B + Com + F, and COMBI + F (Table 3.3). There was no significant effect of treatments on harvest index.

The average leaf chlorophyll, measured during the crop growth period, was significantly (p < 0.01) increased by the organic treatments until late in the trial period, with a decrease following grain filling and leaf senescence (Figure 3.3). Leaf chlorophyll was broadly similar across all plots amended by organic treatments. Addition of organic amendments increased the average leaf chlorophyll by 4.2 to 5.7% over F alone, which indicates healthier plants, explaining the higher biomass and grain yields. Analysis of variance over the sampling time revealed that soil amendments by chlorophyll sampling date interaction also significantly (p < 0.01) influenced leaf chlorophyll content (Figure 3.3). Average specific leaf weight (SLW) and leaf water content (LWC) did not differ significantly between any treatments at any time. However, SLW increased with plant age (Table 3.3). Percentage LWC was inversely correlated with SLW, so the LWC decreased as the growth of plants progressed (Table 3.3). Despite numerical variations, significant differences were not observed amongst organic amendments, including compost for most measurements of agronomic and soil parameters.

| Treatments                | GY                    | TBY                   | HI    | HGW    | Av. CHLC | Av. SLW                | Av. LWC |
|---------------------------|-----------------------|-----------------------|-------|--------|----------|------------------------|---------|
|                           | (t ha <sup>-1</sup> ) | (t ha <sup>-1</sup> ) | (%)   | (g)    | (SPAD-   | (mg mm <sup>-2</sup> ) | (%)     |
|                           |                       |                       |       |        | unit)    |                        |         |
| Control (F)               | 7.14 c                | 17.87 b               | 39.9  | 34.7 b | 47.4 b   | 0.049                  | 74.4    |
| $\mathbf{B} + \mathbf{F}$ | 9.21 a                | 21.04 a               | 43.8  | 33.5 b | 49.4 a   | 0.049                  | 75.3    |
| Com + F                   | 7.82 bc               | 20.81 a               | 37.6  | 38.1 a | 50.0 a   | 0.050                  | 74.7    |
| B + Com + F               | 8.57 ab               | 19.46 ab              | 44.1  | 35.0 b | 49.5 a   | 0.049                  | 75.2    |
| COMBI + F                 | 8.08 bc               | 20.21 a               | 40.0  | 39.1 a | 50.1 a   | 0.051                  | 74.7    |
| <i>p</i> value            | 0.016                 | 0.017                 | 0.152 | 0.011  | 0.001    | 0.174                  | 0.825   |
| LSD (0.05)                | 1.05                  | 1.73                  | 5.9   | 3.0    | 1.30     | 0.002                  | 2.1     |
| CV (%)                    | 6.8                   | 4.6                   | 7.6   | 4.4    | 1.4      | 2.5                    | 1.5     |

Table 3.3. Effects of biochar, compost and their mixture on growth and yield of maize.

Within each column, means with different letters are significantly different at p < 0.05. Average CHLC: average chlorophyll content (average of six temporal measurements); GY: grain yield; TBY: total biomass yield; HI: harvest index; HGW: hundred grain weight; LSD: least significant difference; CV: coefficient of variation.



**Figure 3.2.** Mean leaf chlorophyll content as influenced by the interaction of treatments and time of measurements after planting maize (n = 90). Error bars represent ±1SE.

## 3.3.2. Plant nutrient status and grain quality

The organic amendments significantly (p < 0.05 and p < 0.01) increased foliar N and P concentrations for samples taken prior to tasseling (Table 4). Leaf N and P concentrations ranged from 3.2 to 3.6% and 0.28 to 0.40%, respectively, with the lowest being from the control (F). The leaf N and P concentrations in the B + F, Com + F, B + Com + F, and COMBI + F treatments were higher than that of F only treatment, by 4.7-13.4% and 10.6-41.3%, respectively. The ratio of leaf N: P was 10.5 to 11.0 for all treatments.

|                | ]     | Mid-seasor | n leaf nutr | ient conce | entrations |        | Kernel is         | sotope         | C and M |       |       |       |
|----------------|-------|------------|-------------|------------|------------|--------|-------------------|----------------|---------|-------|-------|-------|
|                | С     | N          | C: N        | K          | Р          | N: P   | δ <sup>13</sup> C | $\delta^{15}N$ | С       | N     | C: N  | GPC   |
| Treatment      | (%)   | (%)        | ratio       | (%)        | (%)        | ratio  | (‰)               | (‰)            | (%)     | (%)   | ratio | (%)   |
| F              | 43.0  | 3.21 b     | 13.4        | 3.62       | 0.28 b     | 11.5 a | -10.7 ab          | 0.63 d         | 43.7    | 1.75  | 25.1  | 10.9  |
| B + F          | 44.7  | 3.53 a     | 12.7        | 3.17       | 0.31 b     | 11.3 a | -10.6 a           | 1.36 a         | 44.3    | 1.67  | 26.5  | 10.4  |
| Com + F        | 45.0  | 3.36 ab    | 13.4        | 3.66       | 0.38 a     | 9.2 b  | -10.9 bc          | 0.96 c         | 44.6    | 1.75  | 25.5  | 11.0  |
| B + Com + F    | 44.1  | 3.48 ab    | 12.7        | 3.77       | 0.39 a     | 9.1 b  | -11.0 c           | 0.84 c         | 43.6    | 1.75  | 25.0  | 11.0  |
| COMBI + F      | 45.4  | 3.64 a     | 12.5        | 4.21       | 0.40a      | 8.9 b  | -10.7 ab          | 1.09 b         | 43.2    | 1.69  | 25.7  | 10.5  |
| <i>p</i> level | 0.152 | 0.050      | 0.220       | 0.074      | 0.001      | 0.026  | 0.032             | 0.018          | 0.194   | 0.446 | 0.386 | 0.413 |
| LSD (0.05)     | 1.97  | 0.29       | 1.07        | 0.67       | 0.04       | 1.9    | 0.28              | 0.38           | 1.23    | 0.18  | 1.77  | 0.79  |
| CV (%)         | 2.4   | 4.3        | 4.4         | 9.7        | 6.6        | 10.1   | -1.4              | 20.5           | 1.52    | 5.5   | 3.7   | 3.9   |

**Table 3.4.** Effects of biochar, compost and their mixture on leaf nutrient concentration and kernel C and N content, isotopic composition and protein content.

Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation.

Grain  $\delta^{15}$ N and  $\delta^{13}$ C values significantly differed among treatments, with the highest for both being from B + F, and the lowest from the F only treatment. Applications of B + F, Com + F, B + Com + F and COMBI + F increased grain  $\delta^{15}$ N by 2.2, 1.5, 1.3 and 1.7 times, respectively, relative to the F alone treatment (Table 3.4). Com + F and B + Com + F soil amendments resulted in lower  $\delta^{13}$ C values of -10.9 and -11.0 ‰, respectively, compared to the other treatments. Grain contents of C and N and C: N ratios were not significantly different between any treatments (Table 3.4). Grain protein content did not differ significantly amongst the treatments (Table 3.4), ranging from 10.4% to 11.0 %, which is within the optimum range (Gupta et al., 2009).

## 3.3.3. Soil physicochemical properties

Soil physical and chemical properties at the mid-season growth stage and after harvest, under the different treatments, are presented in Tables 3.5 and 3.6. Soil water content (SWC) was significantly (p < 0.05) increased by all organic treatments on average throughout the trial, with the difference between organic amendments and conventional practice being greatest in the dry season as the crop matured (Table 3.5). The average maximum SWC was higher by 9, 23.6, 24.5 and 14.6% as a result of the additions of B + F, Com + F, B + Com + F and COMBI + F, respectively, as compared to the F only treatment (Table 3.5). The treatments had no significant effect on soil bulk density.

SOC content was significantly (p < 0.01) increased by all organic treatments at the mid-season sampling, but not by the end of the trial period. However, soils from treatments that included biochar will have had their carbon stock increased by the amount of biochar added, with a long-term sequestration potential of 0.62 t C per ton of biochar added. This is not evident in the data presented, as much of the biochar is > 2 mm and hence does not contribute to the < 2 mm fraction of soil analyzed. The organic amendments significantly ( $p \le 0.05$ ) increased some soil chemical parameters, including total N, available P, NO<sub>3</sub>-N and NH<sub>4</sub><sup>+</sup>-N, and C: N ratio at the mid-season trial, but a systematic difference was not apparent by the trial end-season (Table 3.5). Although numerically higher values were observed in soils treated with organic amendments, soil pH was not consistently nor significantly influenced by any treatment at any point (Table 3.5).

At the mid-season sampling, the highest SOC content and C: N ratio were obtained from B + F addition, and soil N contents from Com + F and COMBI + F, but there were no statistically significant differences amongst Com + F, B + Com + F and COMBI + F treatments for SOC (Table 3.5). The highest plant available P was obtained from COMBI + F, and soil NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N from B + Com + F soil amendments. In comparison to the F only treatment, applications of B + F, Com + F, B +

Com + F and COMBI + F increased soil C 1.7, 1.4, 1.5 and 1.5 times, and N contents 1.1, 1.2, 1.1 and 1.2 times, respectively (Table 3.5). Biochar, Com + F, B + Com + F, and COMBI + F increased soil available P 1.1, 1.4, 1.4 and 1.5 times, respectively, compared to the F only treatment. Soil NH<sub>4</sub><sup>+-</sup> N content was increased by a factor of 1.3, 1.4, 1.9 and 1.4, and soil NO<sub>3</sub><sup>--N</sup> by a factor of 1.2, 1.1, 1.7 and 1.4 with the respective application of B + F, Com + F, B + Com + F and COMBI + F, compared to F (Table 3.5). Moreover, despite the lower magnitude of response in comparison to the mid-season growth stage, C: N ratio, soil available P and NH<sub>4</sub><sup>+-N</sup> were increased by the organic amendments at the trial end-season. Soil NO<sub>3</sub><sup>--N</sup> showed a significant response to the treatments at harvest, but with a lower magnitude and inconsistent trends compared to the mid-season growth stage. Soil C decreased in the order B + F > B + Com + F > COMBI + F > Com + F > F (Table 3.5).

|                | SBD                   | Av. SWC  | pН                 | SOC    | Ν       | C: N   | Nutrient concentration (mg kg <sup>-1</sup> ) |                     |                                 |
|----------------|-----------------------|----------|--------------------|--------|---------|--------|---|---------------------|---------------------------------|
| Treatments     | $(g \text{ cm}^{-3})$ | (%)      | (H <sub>2</sub> O) | (%)    | (%)     | Ratio  | Colwell P                                     | NH4 <sup>+</sup> -N | NO <sub>3</sub> <sup>-</sup> -N |
| Mid-season     |                       |          |                    |        |         |        |   |                     |                                 |
| F              | 0.75                  | 23.3 c   | 5.41               | 2.15 c | 0.15 c  | 14.1 c | 24.0 b  | 30.0 c              | 29.6 c                          |
| B + F          | 0.72                  | 25.4 bc  | 5.67               | 3.73 a | 0.16 bc | 23.3 a | 27.0 b  | 38.8 bc             | 36.5 bc                         |
| Com + F        | 0.79                  | 28.8 ab  | 5.82               | 3.11 b | 0.18 a  | 17.8 b | 34.3 a  | 41.3 b              | 32.6 bc                         |
| B + Com + F    | 0.81                  | 29.0 a   | 5.65               | 3.32 b | 0.17 ab | 20.0 b | 33.0 a  | 57.3 a              | 51.1 a                          |
| COMBI + F      | 0.76                  | 26.7 abc | 5.84               | 3.19 b | 0.18 a  | 18.2 b | 36.7 a  | 41.8 b              | 41.9 ab                         |
| <i>p</i> level | 0.549                 | 0.016    | 0.268              | 0.001  | 0.034   | 0.002  | 0.003   | 0.003               | 0.011                           |
| LSD (0.05)     | 0.12                  | 3.2      | 0.32               | 0.36   | 0.02    | 3.1    | 5.5   | 10.1                | 10.7                            |
| CV (%)         | 8.6                   | 6.3      | 3.0                | 6.1    | 5.4     | 8.7    | 9.4   | 12.8                | 14.9                            |
| End-season     |                       |          |                    |        |         |        |   |                     |                                 |
| F              | 0.76                  | 24.7     | 5.76               | 3.09   | 0.18    | 17.2   | 20.8 b  | 27.7                | 18.1 bc                         |
| B + F          | 0.78                  | 24.5     | 5.92               | 3.34   | 0.17    | 19.6   | 19.5 b  | 29.6                | 14.1 c                          |
| Com + F        | 0.76                  | 23.0     | 6.00               | 3.22   | 0.18    | 17.9   | 25.7 b  | 33.7                | 8.5 d                           |
| B + Com + F    | 0.78                  | 21.8     | 5.86               | 3.34   | 0.18    | 18.6   | 21.2 b  | 31.1                | 23.3 a                          |
| COMBI + F      | 0.76                  | 22.8     | 6.00               | 3.24   | 0.17    | 19.1   | 36.7 a  | 30.4                | 21.7 ab                         |
| <i>p</i> level | 0.949                 | 0.246    | 0.182              | 0.143  | 0.115   | 0.004  | 0.005   | 0.074               | 0.001                           |
| LSD (0.05)     | 0.09                  | 3.07     | 0.23               | 0.22   | 0.009   | 0.71   | 7.76  | 3.98                | 4.70                            |
| CV (%)         | 5.96                  | 7.00     | 2.04               | 3.55   | 2.62    | 2.05   | 16.65   | 6.94                | 14.56                           |

Table 3.5. Treatment effects on soil physicochemical properties (0-30 cm) at the mid-season (grain filling) stage and trial end-season.

Within each column, means with different letters are significantly different at p < 0.05. SBD: Soil bulk density; Av. SWC: Average soil water content; LSD: Least significant difference: CV: Coefficient of variation.

|                           | Exchangeable cations (cmol(+) kg <sup>-1</sup> ) |         |         |          |        |         |                          |  |  |  |  |  |  |
|---------------------------|--|---------|---------|----------|--------|---------|--------------------------|--|--|--|--|--|--|
| Treatments                | Ca   | Mg      | K       | Na       | Al     | CEC     | EC (dS m <sup>-1</sup> ) |  |  |  |  |  |  |
| Mid-season                |  |         |         |          |        |         |                          |  |  |  |  |  |  |
| F                         | 4.1 b  | 0.59 b  | 0.49 c  | 0.023 c  | 0.22 a | 5.2 b   | 0.110                    |  |  |  |  |  |  |
| B + F                     | 5.1 ab   | 0.67 b  | 0.57 bc | 0.023 c  | 0.16 b | 6.5 a   | 0.110                    |  |  |  |  |  |  |
| Com + F                   | 5.2 ab   | 0.90 a  | 0.72 a  | 0.030 ab | 0.08 c | 6.9 a   | 0.093                    |  |  |  |  |  |  |
| B + Com + F               | 5.1 ab   | 0.83 a  | 0.66 ab | 0.027 bc | 0.07 c | 6.8 a   | 0.130                    |  |  |  |  |  |  |
| COMBI + F                 | 6.0 a  | 0.82 a  | 0.78 a  | 0.033 a  | 0.05 c | 7.6 a   | 0.090                    |  |  |  |  |  |  |
| <i>p</i> level            | 0.037  | 0.005   | 0.011   | 0.028    | 0.001  | 0.010   | 0.198                    |  |  |  |  |  |  |
| LSD (0.05)                | 1.20   | 0.10    | 0.14    | 0.006    | 0.037  | 1.1     | 0.037                    |  |  |  |  |  |  |
| CV (%)                    | 10.7   | 7       | 12      | 12.5     | 17.1   | 8.6     | 18.7                     |  |  |  |  |  |  |
| End-season                |  |         |         |          |        |         |                          |  |  |  |  |  |  |
| F                         | 4.03 b   | 0.50 c  | 0.49    | 0.01 d   |        | 5.23 b  | 0.04                     |  |  |  |  |  |  |
| $\mathbf{B} + \mathbf{F}$ | 4.93 ab  | 0.61 bc | 0.54    | 0.02 c   |        | 6.20 ab | 0.04                     |  |  |  |  |  |  |
| Com + F                   | 5.53 a   | 0.74 ab | 0.54    | 0.02 c   |        | 6.93 a  | 0.04                     |  |  |  |  |  |  |
| B + Com + F               | 4.93 ab  | 0.76 a  | 0.61    | 0.03 b   |        | 6.50 ab | 0.04                     |  |  |  |  |  |  |
| COMBI + F                 | 6.13 a   | 0.74 ab | 0.53    | 0.04 a   |        | 7.47 a  | 0.05                     |  |  |  |  |  |  |
| <i>p</i> level            | 0.050  | 0.008   | 0.285   | 0.001    |        | 0.035   | 0.648                    |  |  |  |  |  |  |
| LSD (0.05)                | 1.42   | 0.13    | 0.11    | 0.01     |        | 1.30    | 0.012                    |  |  |  |  |  |  |
| CV (%)                    | 12.71  | 10.41   | 11.18   | 22.18    |        | 10.68   | 15.0                     |  |  |  |  |  |  |

Table 3.6. Treatment effects on soil chemical properties at the grain filling stage and trial end-season.

Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation

Pre-planting available soil P, Ca, Mg, Na, Zn and B were low, and manganese (Mn) high (Hazelton and Murphy, 2007). Soil exchangeable Ca, Mg, Na and CEC were significantly (p < 0.05) enhanced by all organic amendments throughout the trial period. Exchangeable K and Al concentrations were significantly changed by the treatments only at the mid-season growth stage, but a systematic difference was not observed at the trial end-season (Table 3.6). In contrast, the soil EC did not significantly vary throughout the trial period. Compared to the F only treatment, application of B + F, Com + F, B + Com + F and COMBI + F increased soil K 1.2, 1.5, 1.3 and 1.6 times, and Mg 1.1, 1.5, 1.4 and 1.4 times, respectively. Application of B + F, B + Com + F and COMBI + F esulted in significantly higher soil Mg content than B + F, and F alone, treatments. In contrast, COMBI + F-amended soil achieved the highest soil K and Na contents and CEC, but significantly reduced soil Al

content compared to B + F, and F only treatments. In B + F, Com + F, B + Com + F and COMBI + F treated soils, CEC was higher by a factor of 1.2, 1.3, 1.2 and 1.5, and Ca by a factor of 1.2, 1.3, 1.3 and 1.5, respectively, compared to the control. Use of B + F, Com + F, B + Com + F, and COMBI + F reduced Al content by 27, 64, 68 and 77%, respectively, compared to the F only treatment (Table 3.6).

Grain yield was correlated significantly and positively with total biomass, leaf chlorophyll, leaf N and P concentration, SOC and SWC (r = 0.81, 0.65, 0.74, 0.72, 0.63 and 0.64, respectively), but not significantly correlated with SLW, grain  $\delta^{15}$ N or soil NO<sub>3</sub><sup>-</sup>-N. Leaf chlorophyll was correlated significantly and positively with total soil N, leaf N and P concentration and CEC of the soil (r = 0.78, 0.74, 0.69 and 0.75, respectively). Soil available Colwell P was strongly correlated with soil CEC and exchangeable Mg and Ca (r = 0.85, 0.77 and 0.79, respectively; Table 3.7). Exchangeable Ca was strongly correlated with the CEC of the soil (r = 0.96; Table 3.7).

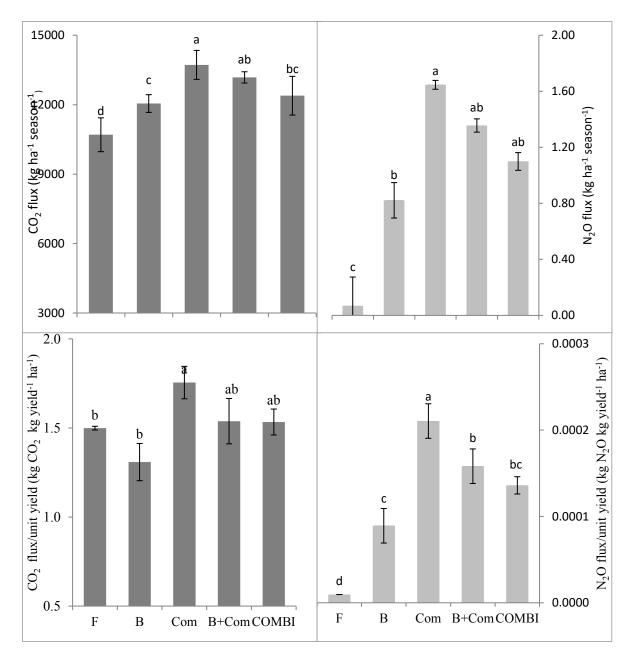
## 3.3.4. Greenhouse gas (GHG) emissions

The highest average emissions of CO<sub>2</sub> and N<sub>2</sub>O over the entire growing season were recorded from Com + F and the lowest from the F only treatment (Figure 4). In this study, the average CO<sub>2</sub> and N<sub>2</sub>O fluxes were in the order Com + F > B + Com + F > COMBI + F > B + F > F (Figure 4). In terms of GHG emissions relative to maize yield, the CO<sub>2</sub> and N<sub>2</sub>O emissions from the Com + F treatment were higher than other treatments (Figure 4). CO<sub>2</sub> emissions varied erratically over time for all treatments, and in general, CO<sub>2</sub> emissions from the organic amended treatments were higher than those from the fertilizer only treatment (Figure 5). Emission of N<sub>2</sub>O generally decreased over time for all treatments, and at the end of the trial period, emission for B + F was lower than other treatments (Figure 5). In general, while N<sub>2</sub>O fluxes were higher from organic amended treatments for the first two months, after this period, fluxes were significantly lower than the F only treatment.

| Character             | SWC                 | Soil                  | Soil                | CEC        | Exch.              | Exch.              | Exch.              | Av.                | Total              | SOC                | pН                 | Grain              | Leaf               | Leaf        | Leaf       | SLW                 | CHL         | TBY    |
|-----------------------|---------------------|-----------------------|---------------------|------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|-------------|------------|---------------------|-------------|--------|
|                       |                     | $\mathrm{NH_{4}^{+}}$ | NO <sub>3</sub> -   |            | Κ                  | Mg                 | Ca                 | Р                  | Ν                  |                    | (H <sub>2</sub> O) | $\delta^{15}N$     | Р                  | Ν           | С          |                     | С           |        |
| GY                    | 0.63*               | 0.53*                 | 0.18 <sup>ns</sup>  | $0.57^{*}$ | 0.62*              | 0.53*              | 0.56*              | 0.52*              | 0.59*              | 0.64*              | $0.58^{*}$         | 0.41 <sup>ns</sup> | 0.72**             | 0.74**      | 0.55*      | -0.35 <sup>ns</sup> | 0.65**      | 0.81** |
| TBY                   | 0.65**              | 0.63*                 | 0.21 <sup>ns</sup>  | 0.54*      | 0.56*              | 0.46 <sup>ns</sup> | $0.52^{*}$         | $0.50^{*}$         | 0.56*              | $0.57^{*}$         | 0.65**             | 0.15 <sup>ns</sup> | 0.71**             | 0.75**      | $0.55^{*}$ | -0.33 <sup>ns</sup> | $0.74^{**}$ |        |
| CHLC                  | 0.59*               | $0.52^{*}$            | 0.28 <sup>ns</sup>  | 0.75**     | $0.62^{*}$         | 0.71**             | 0.63**             | 0.65**             | $0.78^{**}$        | $0.60^{*}$         | 0.72**             | 0.47 <sup>ns</sup> | 0.69**             | $0.74^{**}$ | 0.63*      | 0.54*               |             |        |
| SLW                   | 0.14 <sup>ns</sup>  | 0.67**                | $0.54^{*}$          | 0.56*      | 0.31 <sup>ns</sup> | 0.35 <sup>ns</sup> | 0.51*              | $0.60^{*}$         | 0.63*              | 0.10 <sup>ns</sup> | 0.23 <sup>ns</sup> | 0.48 <sup>ns</sup> | 0.37 <sup>ns</sup> | 0.51*       | $0.56^{*}$ |                     |             |        |
| Leaf C                | 0.17 <sup>ns</sup>  | 0.28 <sup>ns</sup>    | 0.19 <sup>ns</sup>  | 0.74**     | $0.56^{*}$         | 0.53*              | 0.67**             | 0.59*              | $0.62^{*}$         | 0.52*              | 0.44 <sup>ns</sup> | 0.32 <sup>ns</sup> | $0.54^{*}$         | $0.60^{*}$  |            |                     |             |        |
| Leaf N                | 0.28 <sup>ns</sup>  | 0.16 <sup>ns</sup>    | $0.58^{*}$          | 0.64*      | $0.52^{*}$         | 0.43 <sup>ns</sup> | $0.52^{*}$         | 0.42 <sup>ns</sup> | 0.53*              | $0.60^{*}$         | 0.25 <sup>ns</sup> | 0.54*              | 0.44 <sup>ns</sup> |             |            |                     |             |        |
| Leaf P                | -0.60*              | 0.71**                | 0.20 <sup>ns</sup>  | 0.75**     | 0.74**             | 0.83**             | 0.67**             | $0.80^{**}$        | 0.53*              | 0.55**             | $0.58^{*}$         | 0.79**             |                    |             |            |                     |             |        |
| Grain $\delta^{15}N$  | -0.55*              | $0.57^{*}$            | 0.36 <sup>ns</sup>  | 0.66**     | $0.60^{*}$         | 0.67**             | $0.60^{*}$         | 0.73**             | 0.68**             | 0.35 <sup>ns</sup> | 0.44 <sup>ns</sup> |                    |                    |             |            |                     |             |        |
| pH-H <sub>2</sub> O   | 0.36 <sup>ns</sup>  | 0.17 <sup>ns</sup>    | -0.27 <sup>ns</sup> | 0.56*      | 0.55*              | 0.38 <sup>ns</sup> | 0.67**             | 0.42 <sup>ns</sup> | 0.33 <sup>ns</sup> | 0.22 <sup>ns</sup> |                    |                    |                    |             |            |                     |             |        |
| SOC                   | -0.33 <sup>ns</sup> | 0.23 <sup>ns</sup>    | 0.54*               | 0.56*      | 0.39 <sup>ns</sup> | 0.42 <sup>ns</sup> | 0.45 <sup>ns</sup> | 0.47 <sup>ns</sup> | $0.52^{*}$         |                    |                    |                    |                    |             |            |                     |             |        |
| Total soil N          | -0.16 <sup>ns</sup> | 0.59*                 | 0.15 <sup>ns</sup>  | 0.63*      | 0.44 <sup>ns</sup> | 0.75**             | 0.51*              | 0.75**             |                    |                    |                    |                    |                    |             |            |                     |             |        |
| Av. P                 | -0.32 <sup>ns</sup> | $0.58^{*}$            | 0.19 <sup>ns</sup>  | 0.85***    | 0.66**             | 0.77**             | 0.79*              |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |
| Exch. Ca              | -0.26 <sup>ns</sup> | 0.37 <sup>ns</sup>    | $0.07^{ns}$         | 0.96***    | 0.71**             | 0.53*              |                    |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |
| Exch. Mg              | -0.51*              | 0.75**                | 0.34 <sup>ns</sup>  | 0.71**     | 0.75**             |                    |                    |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |
| Exch. K               | -0.63*              | $0.55^{*}$            | 0.12 <sup>ns</sup>  | 0.77**     |                    |                    |                    |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |
| CEC                   | -0.33 <sup>ns</sup> | 0.53*                 | 0.26 <sup>ns</sup>  |            |                    |                    |                    |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |
| Soil NO3 <sup>-</sup> | -0.15 <sup>ns</sup> | $0.60^{*}$            |                     |            |                    |                    |                    |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |
| Soil NH4 <sup>+</sup> | -0.43 <sup>ns</sup> |                       |                     |            |                    |                    |                    |                    |                    |                    |                    |                    |                    |             |            |                     |             |        |

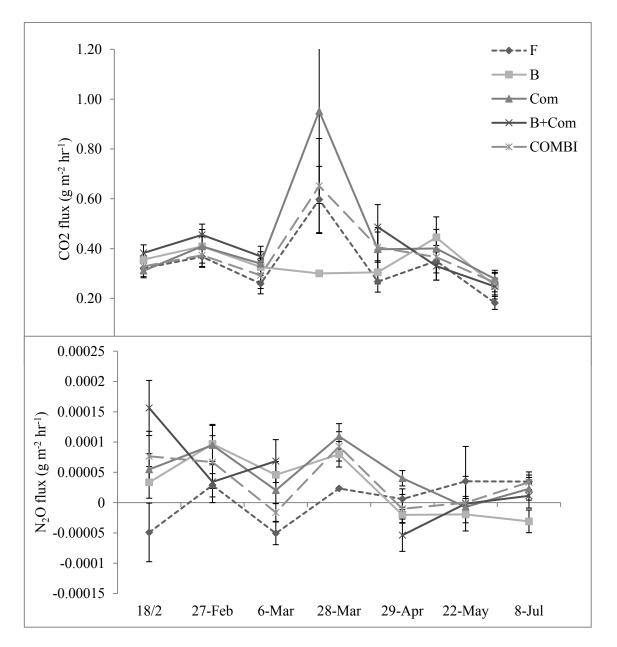
**Table 3.7.** Correlation coefficients among plant parameters, soil water content, contents of soil and plant nutrients tested at five soil fertility treatments in 2014 in north Queensland, Australia.

GY: grain yield; TBY: total biomass yield: CHLC: chlorophyll content; SLW: specific leaf weigh; CEC: Cation exchange capacity; SWC: Soil water content; SOC: Soil organic carbon: Av. P: Available soil phosphorus. \*p < 0.05, \*\*p < 0.01, \*\*\*p < 0.00; ns: Not significant



**Figure 3.3.** The upper half shows the average seasonal emissions of  $CO_2$  and  $N_2O$  and the lower half shows units of  $CO_2$ -C and  $N_2O$ -N produced per unit of maize grain yield (kg  $CO_2$ -C and  $N_2O$ -N ha<sup>-1</sup> kg yield<sup>-1</sup> ha<sup>-1</sup>) over the whole growing season for each treatment.

Note: LSD (p = 0.05) = 963 and 0.35, and CV = 4.1 and 27.3 for CO<sub>2</sub> and N<sub>2</sub>O, respectively) (upper). LSD (p = 0.05) = 0.24 and 0.00005, and CV = 8.2 and 20.1 for CO<sub>2</sub> and N<sub>2</sub>O, respectively) (lower). Columns with the same letter are not significantly different at p = 0.05. Error bars represent ±1 SE.



**Figure 3.4.** The absolute increase/decrease in CO<sub>2</sub>-C and N<sub>2</sub>O-N flux from the four amendments compared to fertilizer only. Where positive values indicate amounts above the concurrent F value and negative values indicate amounts below the concurrent F values. Measurements were carried out on days 5, 14, 21, 43, 75, 98 and 145 after planting (18 February - 8 July 2014). Error bars represent  $\pm$  1 SE.

## 3.4. Discussion

## 3.4.1. Plant growth, yield and nutrient uptake

Our findings indicate that application of all organic amendments promoted growth and productivity of maize. Total biomass and yield were significantly increased by the organic amendments, with grain yield and total biomass increments of 10-29% and 9-18%, respectively, relative to the control. These improvements in crop performance are consistent with other studies (Major et al., 2010; Mekuria et al., 2014; Uzoma et al., 2011; Zhang et al., 2016) and may be attributed to improved availability of nutrients and soil moisture. Previous studies reported that maize yield increased by 98 – 150% and water use efficiency by 91–139% in response to manure biochar addition (Uzoma et al., 2011); 114 - 444% with the application of wood and maize stalk biochars (Cornelissen et al., 2013); and 43% increase in maize total biomass due to the application of charred bark of *Acacia mangium* (Yamato et al., 2006). Major et al. (2010) found no difference in yield for 8 and 20 t ha<sup>-1</sup> biochar application after one year of cropping on a savanna Oxisol in Colombia. However, maize yield in biochar amended treatments increased up to 140% relative to the control over the following three years, implying a longer-term beneficial impact of biochar on yield and soil fertility.

There have been several studies of the impact of biochar application on cereals. For instance, Solaiman et al. (2012) have found that wheat seed germination increased from 93 to 98% with the addition of biochar from different feedstock at 10 t ha<sup>-1</sup>, but the same rate had no significant effect on maize seed germination in New Zealand (Free et al., 2010). The positive effect of biochar on germination could be due to the change in the physical condition of soils, in that the dark color of biochar alters thermal dynamics, possibly water availability and hormonal-type effects. Consequently, this accelerates germination, and improves maize growth over the first month following seedling emergence, allowing more time for growth.

The impact of biochar on maize yield has been studied in both temperate and tropical cropping systems, with a range of results dependent on biochar type, soil type, climate and time. Negative effects on yield have been attributed to a combination of biochar type and soil conditions (Butnan et al., 2015; Rajkovich et al., 2012). Gaskin et al. (2010) reported that biochar addition to a sandy loam soil, in a temperate climate, induced both positive and negative effects on yield, depending on biochar type. In most cases, biochar addition in tropical soils has led to increases of variable magnitude, and in some cases, to considerable yield increases of 20-200% (Kimetu et al., 2008; Martinsen et al., 2014; Yamato et al., 2006). Zhu et al. (2015) also reported that biochar + NPK amendment of a red soil increased maize total biomass by 2.7 - 3.5 and 1.5 - 1.6 times, compared to that of NPK only and biochar only amendments. The effect was attributed to biochar nutrient content (21 - 36% of effect) and indirect increases in fertility (35 - 42% of effect). This implies that biochar amendment is

very beneficial on red Ferralsols, which are low in pH and have low availability of major plant nutrients, such as P and some exchangeable cations.

Chlorophyll content, an indicator of photosynthetic activity, is related to the N content in green plants and serves as a measure of the response of crops to N fertilizer application and soil nutrient status (Minotta and Pinzauti, 1996). Studies have shown that application of biochar and compost with fertilizer significantly increased the leaf chlorophyll content of crops compared to fertilizer alone (Adekayode and Olojugba, 2010; Agegnehu et al., 2015b). The increase in leaf chlorophyll with plant age suggests increased availability of nutrients and water over time, due to the organic amendments. In this study, while Com + F, B + Com + F and COMBI + F all improved the overall plant growth and yield, B + F provided the greatest yield benefit; confirming its importance for improving longterm soil fertility and crop yield, in agreement with the findings of other studies (Cornelissen et al., 2013; Doan et al., 2015; Zhang et al., 2016).

Without amendment, the soil used in this study had low plant-available contents of some nutrients. Among macronutrients, N, K and P are required in the greatest quantities by most cereals. The separate or combined application of biochar and compost significantly increased soil nutrient status during the crop growth period, indicating that their usage may prove beneficial for crop nutrition and yield. Although differences were observed among treatments, leaf N, P and K concentrations were in the reported sufficiency range for all treatments, with 2 - 5%, 0.2 - 0.5% and 1 - 5%, respectively, were considered sufficient (Motsara and Roy, 2008). However, differences between organic and inorganic amendments in leaf N and P concentrations were obvious. Other studies have shown that use of biochar stimulates plant growth and increases fertilizer use efficiency, especially when biochar is combined with fertilizer (Alburquerque et al., 2013; Schulz and Glaser, 2012; Steiner et al., 2008) and compost (Doan et al., 2015; Schulz et al., 2013). According to Steiner et al. (2008), the total N recovery in soil, crop residues and grains was considerably higher with compost (16.5%), biochar (18.1%), and biochar + compost treatments (17.4%) than with mineral fertilizer alone (10.9%).

The availability of essential nutrients in the correct proportion is a key factor for balanced nutrient uptake, healthy plant growth and optimum yield. For example, a 10:1 ratio of N: P is considered optimum for many crops, which was the ratio found in this study. The nutrient concentrations of plants vary with nutrient availability, plant species, growing conditions, time of sampling and plant parts sampled. The soil P supply is a limiting factor in plant growth, both in alkaline (Wandruszka, 2006) and acidic soils of the humid tropics (Fageria and Baligar, 2008). In this study, leaf P content was higher in organic-amended soil and P was more available to the plants than in mineral-fertilized

soil, implying that compost and biochar supplied P to the soil and also improved its availability by reducing sorption and leaching. Previous studies have indicated that amending Ferralsols with biochar and compost lowers leaching, improves the root-fertilizer contact and, thus, optimizes the availability of P to plants (Agegnehu et al., 2015b; Lehmann et al., 2003). Mau and Utami (2014) further demonstrated that cow-dung manure biochar and mycorrhizal amendments increased the availability of P and its uptake by maize plants. Processed poultry manure and its biochar application increased the concentrations of N, P, K, Zn, Cu and Mn in maize plants, but decreased the Ca and Mg concentrations (Inal et al., 2015). Higher nutrient uptake by plants was accompanied by increased plant growth and yield. There was no difference between the N contents of kernels in any treatment. However, organic soil amendments significantly increased grain  $\delta^{15}$ N compared to the fertilizer alone treatment, implying greater utilization of organic-derived N (which has higher  $\delta^{15}$ N than fertilizer N) in the B + F and B + Com + F treatments than in the other treatments.

### 3.4.2. Soil physicochemical properties

Application of organic amendments in this study showed substantial benefits for SWC and, presumably, water uptake by the plants. Although the organic amendments out-performed fertilizeronly treatments, the trends in SWC within treatments were not consistent throughout the crop growth period, which may be due to a combined effect of rainfall pattern during the growth period and differences in water uptake by plants. Soil water content tended to decline over time, as the crop grew and the dry season progressed. Overall, the effects of the amendments on SWC became more evident as the crop grew and the demand for water markedly increased. Abel et al. (2013) has shown that application of maize husk feedstock biochar increased total pore volume, as well as water content, by up to 16.3%. Abel et al. (2013) further demonstrated that the mass of water at permanent wilting point (PWP) of the maize feedstock biochar and maize silage biochar was 93.4% and 43.5%, respectively. This indicates the correlation between the specific surface area (SSA) of biochars and water volume at permanent wilting point (PWP), resulting from a significant higher SSA for maize feedstock biochar (217 m<sup>2</sup> g<sup>-1</sup>) and assumed corresponding higher microporosity than for maize silage biochar (6.3 m<sup>2</sup> g<sup>-1</sup>). The treatments had positive, significant effects on SWC during the mid-season growth stage, but not after harvest, suggesting that soil-plant interactions may have a critical role in influencing the effects of soil amendments. A likely reason for the observed effects of the treatments on SWC was that greater soil water uptake by growing plants with additions of soil amendments may have reduced deep drainage beyond the root zone and evaporation. It could also be that by the time SWC was measured at end of the season it had been drier for a while and so less likely to see any effects.

Biochar addition, regardless of influences on yield, generally leads to improvements in soil properties, particularly total SOC content, water holding capacity, CEC, and availability of nutrients (Butnan et al., 2015; Gaskin et al., 2010; Martinsen et al., 2014; Zhang et al., 2016). Increases in maize yield may have been variously linked to increased nutrient availability and SWC on the one hand, and decreased toxicity of elements such as Al on the other hand; this, in turn, is linked to changes in soil pH. Optimal soil pH is directly associated with the availability of P. Qayyum et al. (2015) found that soil pH and extractable P were significantly increased with application of low temperature coal (LTC) synthesized from sewage sludge and compost + LTC and compost + lime.

Biochar and biochar-compost addition resulted in significant improvements to soil nutrient content. Their application noticeably increased SOC, total N, available P, exchangeable Ca, and CEC, by 43 - 73%, 14 - 29%, 59 - 117%, 31 - 54% and 20 - 41%, compared to the respective initial nutrient content of the soil. However, changes in exchangeable Mg, K and Na contents were negligible relative to their initial values. Previous studies have indicated that total SOC was significantly increased due to the applications of various biochar types (Angst et al., 2014; Kimetu and Lehmann, 2010; Xie et al., 2013). Xie et al. (2013) reported that biochar addition increased SOC and N contents, and decreased N use efficiency. Significantly higher soil available P was obtained from Com + F, B + Com + F, and COMBI + F, than B + F and F alone treatments. In contrast, B + Com + F addition resulted in the maximum soil NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N concentration, implying that B + Com reduced the rates of nitrification and N immobilization relative to ammonification. Lentz et al. (2014) showed that use of wood biochar produced 33% less cumulative net N mineralization and increased the soil NH<sub>4</sub><sup>+</sup>-N:NO<sub>3</sub><sup>-</sup>-N ratio 1.8-fold compared to manure. The NO<sub>3</sub><sup>-</sup>-N content in compost-amended soil was most notably the lowest, possibly due to immobilization during decomposition of the compost, which had higher C: N ratio than the soil.

Soils with low CEC are often low in fertility and vulnerable to soil acidification. The buffering capacity of a soil increases with the increase in CEC and SOC content. Values of exchangeable Ca and CEC in the organic-amended soils were similar at both sampling times, indicating that the additional CEC was stable. Such stable increases in CEC have been noted in previous studies of biochar application to soil. CEC per unit soil C was up to 1.9 times higher in Anthrosols with high black carbon than in the adjacent soils (Liang et al., 2006). Xu et al. (2012b) reported that the enhanced CEC increased soil fertility through greater nutrient availability, as nutrients are retained in the soil against leaching. The application of 20 t  $ha^{-1}$  biochar to a low-fertility, acidic soil of Colombia

led to significant increases in concentrations of several nutrients in soil solution (Major et al., 2012). In the current study, concentrations of most plant nutrients responded positively to organic amendments, an observation consistent with several previous studies in acidic and highly weathered tropical soils (Agegnehu et al., 2015b; Lehmann et al., 2003; Slavich et al., 2013; Van Zwieten et al., 2010). In this study, soil pH-H<sub>2</sub>O increased from the initial value of 5.6 to the final value of 6.0 across all four organic amendments, but none of the treatments differed significantly from each other, which agrees with the findings of Suddick and Six (2013).

#### 3.4.3. Greenhouse gas emissions

This study compared the influence of different organic amendments on CO<sub>2</sub> and N<sub>2</sub>O emissions. On most measurement dates, the  $CO_2$  and  $N_2O$  emissions from organic amended soils were higher than from the fertilizer only treatment. Positive and negative responses of GHG emissions have both been reported as a result of biochar application to soils (Angst et al., 2013; Scheer et al., 2011; Spokas and Reicosky, 2009; Taghizadeh-Toosi et al., 2011; Zhang et al., 2012b). For example, out of sixteen biochar types evaluated for their impact on GHG emissions, five biochars increased, three biochars reduced and eight had no significant effect on CO<sub>2</sub> emission from agricultural soils; all biochar and soil combinations resulted in decreased or unaltered rates of methane (CH<sub>4</sub>) oxidation, and suppressed N<sub>2</sub>O emission (Spokas and Reicosky, 2009). According to Zhang et al. (2012b), biochar soil amendment decreased N<sub>2</sub>O emission, but increased CH<sub>4</sub> emission. The incorporation of 30 t  $ha^{-1}$ biochar also reduced the emission of  $N_2O$  from ruminant urine patches by > 50% (Taghizadeh-Toosi et al., 2011). Biochars tend to be stable in soil, resulting in lower  $CO_2$  loss (between 0.5% and 5.8%) of total added C) than with other bioenergy by-products applied to soil (Cayuela et al., 2010). In contrast, biochar produced from pine waste material, at a temperature of 550°C, did not show potential to curb GHG emission, where the biochar was co-applied with manure to an alkaline soil (Angst et al., 2013).

The average emission of  $N_2O$  and  $CO_2$  from soils containing organic amendments in this study was generally greater than from the conventional fertilizer, which is in accordance with results of some previous studies (Lentz et al., 2014; Schimmelpfennig et al., 2014; Shen et al., 2014). The fluxes of both gases generally showed a declining trend over time, and both  $CO_2$  and  $N_2O$  emissions per unit of yield were similar to, or higher than, the control. Nitrogen fertilizer is the main source of  $N_2O$ emission in agricultural soils. Hence, reduction of N fertilizer application rate is an efficient approach to abate  $N_2O$  emissions. Previous studies have indicated that wheat straw biochar soil amendment at 20 and 40 t ha<sup>-1</sup> resulted in a reduction in  $N_2O$  emission by 10.7% and 71.8%, respectively (Zhang et al., 2012b), and by more than 31% (Zhang et al., 2016) compared to N fertilizer. Lentz et al. (2014) reported that application of wood biochar resulted in 20% less CO<sub>2</sub> and 50% less N<sub>2</sub>O emissions compared to manure. A review and meta-analysis by Cayuela et al. (2014), using published data from 2007-2013, has also shown that biochars produced from different feedstock reduced soil N<sub>2</sub>O emissions by an average of 54% in laboratory and field studies. In contrast, Shen et al. (2014) reported that biochar amendment of a rice field increased N<sub>2</sub>O emission compared to the NPK only treatment. The effect was ascribed to the increase either in soil NO<sub>3</sub><sup>-</sup>-N content (late in the rice season), or in soil NH<sub>4</sub><sup>+</sup>-N content (early in the rice season). Therefore, because of the complexity of the interactions between organic amendments and soil, additional studies involving long-term field experiments need to be conducted to further strengthen our understanding of the mechanisms of CO<sub>2</sub> and N<sub>2</sub>O emissions from biochar-, compost- and biochar + compost-treated soils.

## 3.5. Conclusions

The results from this field trial, on a fine-textured, basalt-derived Ferralsol, indicated a strong and positive response of maize growth and yield to the application of the organic amendments, with smaller differences evident between the effects of different organic amendments. Total biomass and grain yield were significantly increased, relative to the control for all organic amendments, with increases in grain yield of 10-29% (29% in biochar treatment and 10% in the compost only treatment). SOC content was significantly increased by all organic treatments, compared to the fertilizer only treatment, at the mid-season sampling, but this difference had disappeared by the end of the trial. However, soils from those treatments that included biochar will have had their carbon stock increased by the amount of biochar added, with a long-term sequestration potential of 0.78 t C t<sup>-1</sup> of biochar added. This is not evident in the data presented, as much of the biochar is > 2 mm and, hence, does not contribute to the < 2 mm fraction of soil analyzed. Soil water content was significantly increased by all organic treatment was significantly increased by all organic treatments with a conventional practice, with the effect increasing as the crop grew and the dry season progressed. Soil available P, exchangeable Ca, and CEC were significantly increased by all organic amendments throughout the trial.

Emissions of  $CO_2$  varied erratically over time in all treatments, although, in general,  $CO_2$  emissions from the organic treatments were higher than the fertilizer only treatment. N<sub>2</sub>O emissions generally decreased over time for all treatments and emission from the biochar plus fertilizer was lower than other treatments. Emissions of  $CO_2$  and N<sub>2</sub>O, relative to yield from all organic amendments, were similar to or higher than the control. Overall, improved soil water retention, nutrient status and nutrient concentrations of plant shoots, due to the addition of organic treatments, were accompanied with increased plant growth and yield. Since biochar and compost soil amendments add both macroand micro-nutrients, they may help achieve balanced fertilization. Based on the results from this and previous studies, feedstock type and production procedures may lead to biochar with different physicochemical properties and, hence, different effects on soil quality, crop performance and GHG emissions. Thus, to understand the potential significance of carbon in soil in the form of biochar, biochar + compost, or co-composted biochar-compost, their characteristics and dynamics should be investigated on different soil types and different agro-ecosystems, to evaluate their effects in soilcrop systems and GHG fluxes at least over three years.

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## **Overview of Chapter 4**

This chapter is based on a paper published in the *Agriculture, Ecosystem and Environment* with minimal formatting changes. I have investigated the effects of compost, biochar, compost, biochar-compost mix and co-composted biochar-compost on enhances maize growth, yield and nutrient uptake, soil physicochemical properties, and greenhouse gas emissions in an important peanut growing climate and soil type. The trial site was located at the Atherton Tablelands, North Queensland, Australia on a Ferralsol.

The study was a field trial, designed to determine whether soil quality and crop productivity could be enhanced, and greenhouse gas emissions reduced by biochar and compost, applied singly or together. The feedstock for biochar production was waste willow wood (*Salix spp.*), derived from the removal and restoration activities along watercourses in Victoria, Australia. The biochar (WB; Earth Systems Pty Ltd, Melbourne, Vic.) was produced at a temperature of 500°C as a soil amendment. I collected rigorous plant and soil data throughout the study period. I organized, analyzed and interpreted the data, and wrote the manuscript.<sup>4</sup>

<sup>&</sup>lt;sup>4</sup>Agegnehu, G., Bass, A. M., Nelson, P. N., Muirhead, B., Wright, G., and Bird, M. I. (2015). Biochar and biochar-compost as soil amendments: Effects on peanut yield, soil properties and greenhouse gas emissions in tropical North Queensland, Australia. *Agriculture Ecosystems and Environment* **213**, 72-85.

## **Chapter 4**

# Biochar and Biochar-Compost as Soil Amendments: Effects on Peanut Yield, Soil Properties and Greenhouse Gas Emissions in Tropical North Queensland, Australia

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## Abstract

This study investigated the effects of biochar and compost, applied individually or together, on soil fertility, peanut yield and greenhouse gas (GHG) emissions on a Ferralsol in north Queensland, Australia. The treatments were: 1) inorganic fertilizer only (F) as a control; 2) 10t ha<sup>-1</sup> biochar + F (B+F); 3) 25t compost + F (Com + F) ha<sup>-1</sup>; 4) 2.5t B ha<sup>-1</sup> + 25t Com ha<sup>-1</sup> mixed on site + F; and 5) 25t ha<sup>-1</sup> co-composted biochar-compost + F (COMBI+F). Application of B and COMBI increased seed yield by 23% and 24%, respectively. Biochar, compost, and their mixtures significantly improved plant nutrient availability and use, which appeared critical in improving peanut performance. Soil organic carbon (SOC) increased from 0.93% (F only) to 1.25% (B amended), soil water content (SWC) from 18% (F only) to over 23% (B amended), and CEC from 8.9 cmol(+)/kg (F only) to over 10.3 cmol(+)/kg (organic amended). Peanut yield was significantly, positively correlated with leaf chlorophyll content, nodulation number (NN), leaf nutrient concentration, SOC, and SWC for the organic amendments. Fluxes of CO<sub>2</sub> were highest for the F treatment and lowest for the COMBI treatment, whereas N<sub>2</sub>O flux was highest for the F treatment and all organic amended plots reduced N<sub>2</sub>O flux relative to the control. Principal component analysis indicated that 24 out of 30 characters in the first principal component (PRIN1) individually contributed substantial effects to the total variation between the treatments. Our study concluded that applications of B, Com, B + Com or COMBI have strong potential to, over time, improve SOC, SWC, soil nutrient status, peanut yield and abate GHG fluxes on tropical Ferralsols.

**Keywords:** Biochar, carbon sequestration, co-composted biochar-compost, CO<sub>2</sub> and N<sub>2</sub>O fluxes, Ferralsol, soil fertility

## 4.1. Introduction

Intensive agricultural development and environmental change have led to severe land degradation in many parts of the world. As populations increase, the challenge is to boost agricultural production while coping with environmental change in ways that avoid further land degradation. When fertilizer, manure, or compost is applied to soils, it is often rapidly lost. This results in financial costs to the farmer and leaching of major plant nutrients, such as phosphorus (P), potassium (K) and nitrate nitrogen (NO<sub>3</sub>-N), potentially leading to environmental pollution (Barrow, 2012).

Peanut (*Arachis hypogaea* L.) is an annual legume crop that provides food and helps maintain soil fertility through nitrogen fixation (Bogino et al., 2006). The special ability of leguminous crops to work symbiotically with rhizobia to produce protein is becoming increasingly important in world

agriculture, as this potentially leads to more sustainable agricultural systems, reducing requirements for chemical fertilizer, enhancing residual benefits to subsequent crops and increasing crop yields (Giller, 2001). Global consumption of peanuts is increasing at a rate of around 3% per annum. In 2011/12, peanut production in the world was ~35 million tons, to which Australia contributed less than 0.2% (USDA, 2012). China, India and the USA are the main producers, growing 16.0, 5.5 and 1.7 million tons, respectively, accounting for 45%, 16% and 5% of the world's total (USDA, 2012).

The required mean annual temperatures for peanuts generally exceed 20°C at planting depth and the crop also requires 500-600 mm of water during the growing season. Peanuts are N<sub>2</sub>-fixing, so P, K, calcium (Ca) and sulphur (S) are the most common nutrients applied to peanuts, with magnesium (Mg), zinc (Zn), boron (B), copper (Cu), manganese (Mn) and molybdenum (Mo) also applied, where deficiencies are identified (PCA, 2012). The effects of continuous cultivation on the yield of peanuts and cereals on Ferrosols in subtropical southeast Queensland have been studied previously. Yield reductions and low grain protein concentrations were observed in peanuts grown on continuously cropped soil, due to nutrient deficiencies in surface and subsurface layers (Bell et al., 1995).

In northern Australia, peanuts are planted in rotation with cereal crops, pasture or sugarcane. The crop is generally planted from September to January, to take advantage of summer rain, and harvested after 18-24 weeks. Peanuts are mostly planted on light-colored, light textured and friable soils, with good drainage, a relatively high water-holding capacity and an optimal pH of 6-7; though other soils can support the crop, generally with irrigation. Well-drained soils provide proper aeration for the roots and for the nitrifying bacteria that are necessary for proper mineral nutrition of the plant. A soil organic matter content of between 1 and 2% is preferred, both to improve the water-holding capacity of the soil and to supply plant nutrients (Putnam et al., 1991).

In Australia in 2010/11, 7,300 ha were planted to peanuts, producing 18,400 t of peanuts in shell, at an average yield of 2.5 t ha<sup>-1</sup>, with maximum yields under irrigation of 8 t ha<sup>-1</sup> (PCA, 2012). About 97.5% of national area planted to peanuts was north of the latitude equivalent to the Queensland-New South Wales border and 97% of the total production occurred north of this latitude. Songsri et al. (2009) identified water stress as the major abiotic constraint affecting peanut productivity globally. The peanut industry in Australia is, likewise, considered to be significantly exposed to the impacts of climate change, with production in northern Australia reduced by 30% in the last 25 years as a result of temperature increases and lower than average rainfall (Marshall et al., 2014). Meinke et al. (1996) found that 27% yield reductions from the median yield accompanied dry El Nino periods in northern Australia, while 23% increases over the median yield accompanied wet La Niña conditions.

Biochar is the carbon rich product obtained when biomass is thermally decomposed (pyrolyzed) in restricted, or absence of oxygen. The potential of biochar to improve soil fertility and sequester carbon on centennial timescales (thereby mitigating climate change) has been widely recognized (reviewed in Lehmann and Joseph, 2015). Biochar is an effective carbon (C) sink in the soil because of its high proportion of recalcitrant C with hundreds to thousands of years of stability (Atkinson et al., 2010). While there has been significant work on the production and use of peanut shell biochar as both a soil amendment (e.g., Gaskin et al., 2010) and for decontamination of water (Ahmad et al., 2012), there has been little previous work on the impact of biochar or compost on peanut crops themselves. McClintock and Diop (2005) reported significant increases in above- and below-ground peanut biomass in crops grown by subsistence farmers in Senegal, along with improvements in soil effective cation exchange capacity (ECEC) and soil nutrient concentrations (K and Mg) in soils amended with compost. Yamato et al. (2006) reported a significantly increased peanut yield following biochar amendment of an infertile soil in Sumatra, with no significant change in yield for fertile soil, along with general increases in soil pH, N, available P, and ECEC.

Climate variability in southeast Queensland over the past 20 years, in particular, has led to a major reduction in the dry land peanut crop area. Now, ~33% of the total area is rain fed only, accompanied by a major swing to irrigated production (PCA, 2013). Given that water stress has a major impact on peanut crops, and that both compost and biochar can increase SWC, there is the potential to both increase peanut yield in northern Australia, and to provide an element of 'drought-proofing' to peanut farming operations, through the addition of compost and/or biochar amendments to soils under peanut cultivation. However, there has been no research into the effect of these amendments on crop growth and soil properties in the Australian context. Therefore, based on several studies reporting the positive effects of biochar and compost on soil fertility and productivity of a range of crops elsewhere (e.g., Fischer and Glaser, 2012; Lehmann et al., 2003; Liang-Feng et al., 2014; Liu et al., 2013), we hypothesized that the addition of biochar and compost amendments to soils under peanut cult: (1) enhance soil organic carbon, plant available nutrients and soil water retention and; and (2) improve plant growth and crop yield.

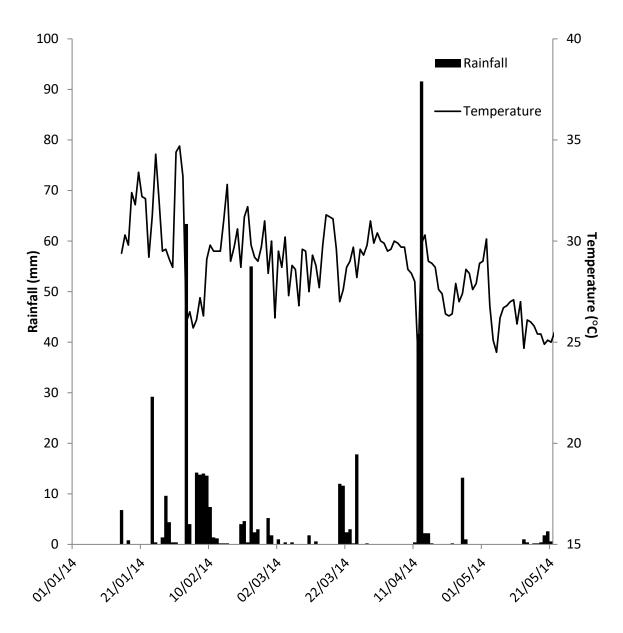
## 4.2. Materials and methods

## 4.2.1. Experimental site

The soil at the site is a Ferralsol (IUSS 2007), developed on Quaternary basalt near Mareeba on the Atherton Tablelands, North Queensland (17.0232°S 145.4027°E; 433 m above sea level). Ferralsols

represent the most highly weathered soils in the classification system (Brady and Weil, 2014). Particle size analysis of the 0 - 30cm interval indicated the soil comprised 21.9% coarse sand (0.2 - 2.0 mm), 40.1% fine sand (0.02 - 0.2 mm), 6.0% silt (0.002 - 0.02 mm) and 32.0% clay (< 0.0002 mm). Daily climate data for this station over the trial period are shown in Figure 4.1. The long-term average annual total precipitation is 880 mm with mean annual maximum and minimum air temperatures of 28.8°C and 17.8°C, respectively. A total of 116 rows were made available for the trial, with plantings arranged in high-density single rows. Crop rows were 0.90 m wide and 360 m in length. Treatment replicates incorporated four rows each to coincide with currently utilized farm implements and practices, with a four-row buffer zone between each treatment. Treatment sequencing was randomized and the total plot area was 0.13 ha per replicate.

One month prior to initiation of the field trial, composite soil samples from a depth of 0 - 30 cm and 30 - 100 cm were randomly collected from nine locations across the trial site. The sites were selected by dividing the trial area into a  $3 \times 3$  grid, and within each of the nine grid cells a sampling point was randomly chosen. At each sampling point, three 0 - 30 cm cores were taken using a vehicle mounted hydraulic corer. One core of 30 - 100 cm per sampling point was also taken. Soil moisture was taken at 0 - 12 cm, using the Campbell Scientific Hydrosense II soil moisture probe, at each sampling location. The soil moisture content was also measured in each sample after oven-drying. Table 4.1 shows the pre-planting physicochemical characteristics of the trial soil.



**Figure 4.1.** Daily rainfall and maximum air temperature recorded between the 1<sup>st</sup> of January 2014 and the 21<sup>st</sup> of May 2014.

| Item                                   | Unit                   | Limit | Average | SE   | Average   | SE   |
|--|------------------------|-------|---------|------|-----------|------|
|  |                        |       | 0-30 cm |      | 30-100 cm |      |
| Soil water content                     | %w/w                   |       | 13.2    | 0.49 | 17.9      | 1.3  |
| Bulk density                           | g/cm3                  |       | 0.55    | 0.02 | 0.89      | 0.03 |
| pH (H <sub>2</sub> O)                  |                        |       | 6.2     | 0.12 | 6.7       | 0.07 |
| pH (CaCl <sub>2</sub> )                |                        |       | 5.6     | 0.12 | 6.4       | 0.06 |
| Electrical conductivity (EC)           | dS/m                   | 1.00  | 0.08    | 0.01 | 0.03      | 0.0  |
| Total nitrogen (N)                     | %                      |       | 0.06    | 0.00 | 0.01      | 0.00 |
| Soil carbon (C)                        | %                      |       | 0.89    | 0.06 | 0.22      | 0.02 |
| C:N ratio                              |                        |       | 14.8    | 0.3  | 22.0      | 0.8  |
| Organic matter (OM)                    | %                      | 0.10  | 1.5     | 0.13 | n/d       | n/d  |
| Colwell phosphorus (P)                 | (mg kg <sup>-1</sup> ) | 1.00  | 58      | 6    | 10.3      | 1.6  |
| Exchangeable sodium (Na)               | cmol(+)/kg             | 0.01  | 0.08    | 0.0  | 0.05      | 0.0  |
| Exchangeable potassium (K)             | cmol(+)/kg             | 0.01  | 0.68    | 0.11 | 0.26      | 0.03 |
| Exchangeable calcium (Ca)              | cmol(+)/kg             | 0.01  | 6.7     | 0.54 | 3.6       | 0.32 |
| Exchangeable magnesium (Mg)            | cmol(+)/kg             | 0.01  | 1.6     | 0.14 | 1.4       | 0.21 |
| Exchangeable aluminium (Al)            | cmol(+)/kg             | 0.01  | 0.05    | 0.0  | 0.02      | 0.0  |
| Cation exchangeable capacity (CEC)     | cmol(+)/kg             | 0.02  | 9.1     | 0.75 | 5.4       | 0.45 |
| Ca: Mg Ratio                           |                        | 0.1   | 4.3     | 0.36 | n/d       | n/d  |
| Sulfur (S)                             | mg kg <sup>-1</sup>    | 1.00  | 15.9    | 1.8  | n/d       | n/d  |
| Chlorine (Cl)                          | mg kg <sup>-1</sup>    | 5     | 10.5    | 1.0  | n/d       | n/d  |
| Copper (Cu)                            | mg kg <sup>-1</sup>    | 0.05  | 1.5     | 0.11 | n/d       | n/d  |
| Zinc (Zn)                              | mg kg <sup>-1</sup>    | 0.05  | < 0.05  | 0.0  | n/d       | n/d  |
| Manganese (Mn)                         | mg kg <sup>-1</sup>    | 0.50  | 119     | 13.3 | n/d       | n/d  |
| Iron (Fe)                              | mg kg <sup>-1</sup>    | 0.50  | 14.4    | 1.1  | n/d       | n/d  |
| Boron (B)                              | mg kg <sup>-1</sup>    | 0.05  | 0.54    | 0.04 | n/d       | n/d  |
| Ammonium nitrogen (NH <sub>4</sub> -N) | mg kg <sup>-1</sup>    | n/d   | n/d     | n/d  | 3.3       | 0.23 |
| Nitrate nitrogen (NO <sub>3</sub> -N)  | mg kg <sup>-1</sup>    | n/d   | n/d     | n/d  | 6.1       | 0.42 |

**Table 4.1.** Physicochemical properties of pre-planting soil samples at 0 - 30 cm and 30 - 100 cm soil depth.

Nitrogen concentrations in soils fell sharply with depth, with most of the N being in the top 0 - 30 cm layer of soils. SE: Standard error; n/d: Not determined.

# 4.2.2. Experimental set-up

The feedstock for biochar production was waste willow wood (*Salix spp.*), derived from removal and restoration activities along watercourses in Victoria, Australia. The biochar (B; Earth Systems Pty Ltd) was produced using a containerized automated batch pyrolysis plant (Charmaker MPP20). Processing of whole logs at up to 5 t per load required over 5-7 hours with highest heating temperatures of over 550°C. The low-density willow feedstock produced biochar with low bulk density (0.17 - 0.21 g cm<sup>-3</sup>), porosity (28 - 37%), apparent skeletal density (0.28 g cm<sup>-3</sup>), Brunauer Emmett Teller (BET) surface area (332 m<sup>2</sup> g<sup>-1</sup>), total pore volume (0.20 m<sup>3</sup> g<sup>-1</sup>), and ash yield (2.7%). The analyses were conducted by PSS Pty Ltd (Gosford, N.S.W.) by mercury porosimetry and surface characteristics by nitrogen adsorption. The biochar was ground to < 10 mm prior to field application.

Two paired compost windrows (each 60 m long, 1.5 m high and 4 m wide) were produced at the King Brown Technologies compost production Facility: one containing compost and biochar (COMBI), and another one containing only compost (Com). The biochar (equivalent to 18 m<sup>3</sup>, or 9% by volume) was added to 80 m<sup>3</sup> (40%) each of green waste and bagasse, 12.5 m<sup>3</sup> (6%) of chicken manure, and 12 m<sup>3</sup> (6%) of compost. This windrow was paired with an adjacent windrow comprising the same volumes of green waste (43%), bagasse (43%), chicken manure (7%) and compost (7%), but without biochar. In both cases, bagasse was laid down first, green-waste mulch added on top, and the windrow turned once. Then chicken manure was added, and the windrows watered and turned six times. In the case of the COMBI, the biochar was then added on top of the windrow and the pile turned a further two times. Both windrows were then covered with black plastic film. They were turned and watered weekly, and the matured product was screened at 25 mm. Biochar, compost and COMBI samples were collected randomly from around and within each pile of material before application in the field, to determine physicochemical characteristics. Nutrient contents of the organic amendments were analyzed in the laboratory following similar methods for soil analysis before the start of the trial (Table 4.2).

| Element                        | Unit                | В     | Com    | COMBI  |
|--------------------------------|---------------------|-------|--------|--------|
| pH (H <sub>2</sub> O)          | Units               | 8.3   | 7.5    | 7.5    |
| pH (CaCl <sub>2</sub> )        | Units               | 9.5   | n/d    |        |
| Carbon (C)                     | %                   | 78    | 30.6   | 34.7   |
| Nitrogen (N)                   | %                   | 0.38  | 1.19   | 0.95   |
| δ15N                           | %0                  | n/d   | +7.5   | +7.8   |
| δ13C                           | ‰                   | n/d   | -24.3  | -21.3  |
| ANC                            | %CaCO <sub>3</sub>  | 2.5   |        |        |
| Sulfur (S)                     | %                   | 0.019 | 0.014  | 0.012  |
| Colwell phosphorus (P)         | mg kg <sup>-1</sup> | 79.5  | 917    | 1104   |
| Acid neutralizing capacity     | %CaCO <sub>3</sub>  | 2.5   | n/d    | n/d    |
| Exchangeable potassium (K)     | cmol(+)/kg          | 7.25  | 1.62   | 1.74   |
| Exchangeable calcium (Ca)      | cmol(+)/kg          | 2.20  | 4.15   | 4.15   |
| Exchangeable magnesium (Mg)    | cmol(+)/kg          | 1.45  | 2.38   | 2.30   |
| Exchangeable sodium (Na)       | cmol(+)/kg          | 0.24  | 0.52   | 0.52   |
| Exchangeable aluminum (Al)     | cmol(+)/kg          | < 0.1 | < 0.1  | < 0.1  |
| Hydrogen (H <sup>+</sup> )     | cmol(+)/kg          | n/d   | 0.3    | 0.11   |
| Cation exchange capacity (CEC) | cmol(+)/kg          | 11.2  | 8.77   | 8.81   |
| Electrical conductivity (EC)   | dS/m                | 0.71  | 2.3    | 2.0    |
| Copper (Cu)                    | mg kg <sup>-1</sup> | 2.55  | 45.0   | 44.0   |
| Zinc (Zn)                      | mg kg <sup>-1</sup> | 83.5  | 133    | 133    |
| Manganese (Mn)                 | mg kg <sup>-1</sup> | 110   | 49.6   | 54.6   |
| Iron (Fe)                      | mg kg <sup>-1</sup> | 0.045 | 246    | 218    |
| Boron (B)                      | mg kg <sup>-1</sup> | 9.25  | 4.8    | 4.3    |
| Molybdenum (Mo)                | mg kg <sup>-1</sup> | < 0.3 | < 0.2  | < 0.2  |
| Cobalt (Co)                    | mg kg <sup>-1</sup> | < 0.4 | < 0.05 | < 0.05 |

**Table 4.2.** Characterization of earth system willow biochar (B), compost (Com) and co-composted biochar-compost (COMBI).

ANC: Acid neutralizing capacity; CEC: Cation exchange capacity; n/d: Not determined.

The experiment comprised five treatments in triplicate, where each replicate occupied 0.13 ha, planted following a maize crop. The treatments were: 1) recommended inorganic fertilizer (F) as a

control, against which all the other treatments were compared; 2) biochar (B) applied at 10 t ha<sup>-1</sup> + F; 3) compost (Com) applied at 25 t ha<sup>-1</sup> + F; 4) 25 t Com ha<sup>-1</sup> + 2.5 t B ha<sup>-1</sup> (B + Com) mixed on site + F; and 5) co-composted biochar-compost (COMBI) applied at 25 t ha<sup>-1</sup> + F. All amendments were applied by broadcast spreading with truck-mounted, computer-controlled distribution bins, and rotary blade spreaders. Spreading occurred after primary deep ripping and disc harrowing. Amendments were then incorporated by rotary hoe, prior to planting, using GPS navigation control. Peanut (cv., *Menzies*) was planted on 13<sup>th</sup> January 2014 with a 4 row 'pneumatic precision planter', with a seed placement accuracy of 1mm. Fertilizer was applied in rows at a rate of 26.6 kg N ha<sup>-1</sup>, 31.6 kg P ha<sup>-1</sup> as ammonium phosphate, 66.2 kg K ha<sup>-1</sup> as muriate of potash, 2.6 kg S ha<sup>-1</sup>, and 0.54 kg Zn ha<sup>-1</sup> as zinc sulfate. Other agronomic practices were applied equally for all treatments during the crop growth period, as per usual on-farm practice. The total rainfall during the crop growing period was 541 mm. The crop was harvested on 17<sup>th</sup> June 2014.

#### 4.2.3. Sampling and measurements

After planting, periodic sampling and measurement of soil parameters, leaf chlorophyll content, specific leaf weight, and emissions of carbon dioxide (CO<sub>2</sub>) and nitrous oxide (N<sub>2</sub>O) were undertaken. The measurements of CO<sub>2</sub> and N<sub>2</sub>O were conducted on days 3, 10, 24, 37, 75 and 124 after planting, in conjunction with soil water content (SWC), between 15<sup>th</sup> January 2014 and the 15<sup>th</sup> May 2014. For gas emission measurements, a standard closed chamber methodology was used, in conjunction with an INNOVA-1412 field portable photoacoustic gas analyzer (LumaSense Technologies, Denmark), with two chambers emplaced per replicate. The total average GHG emissions were calculated from all three replicates over all sample dates of each treatment. This method, therefore, encompassed temporal and spatial variability. For total GHG flux over the trial period, the area under each treatment curve was calculated via standard integration. The intensity of gas emission per unit grain yield was calculated as the ratio of the cumulative emission of CO<sub>2</sub>-C or N<sub>2</sub>O-N (kg ha<sup>-1</sup>) and the seed yield (kg ha<sup>-1</sup>). Soil water content (SWC) at 12 cm depth was taken using a HydroSense II probe (Campbell Scientific, Inc.). Gas fluxes (F) in mg m<sup>-2</sup> h<sup>-1</sup> were calculated using the following equation:

$$F = \left(c \times \left(\frac{P}{1013}\right) \times \left(\frac{273}{T+273}\right) \times M \times \left(\frac{V}{A}\right) / 1000 \times 3600.$$
 (1)

Where c = rate of concentration increase (ppm) at t is 0 using a polynomial curve fit, P is the air pressure in the chamber, T is the ambient temperature within the chamber in  $^{\circ}C$ , M is the molar weight of the subject gas, V is the volume of the headspace and A is the area of the sample chamber. Total

gas emitted over the trial was calculated using a simple trapezoidal integration method assuming linear changes in flux rates between sample points.

The amount of GHG emission per unit yield was calculated as follows:

$$E = \frac{M}{Y}.$$
 (2)

Where *M* is the cumulative emissions of CO<sub>2</sub>-C and N<sub>2</sub>O-N and *Y* is the grain yield (kg ha<sup>-1</sup>).

Leaf chlorophyll measurements were undertaken on days 23, 36, 52, 59, 74, 101 and 134 after planting, in conjunction with specific leaf weight between 5<sup>th</sup> February and 27<sup>th</sup> May 2014, using a chlorophyll meter (SPAD 502, Konica Minolta, Tokyo). For each measurement, duplicate readings were made on the second fully expanded leaf, from the top of the main plant stem, approximately half way along the leaf, taking care to avoid veins and mid-rib. This procedure was repeated for six randomly selected plants. A hole-punch with a diameter of 8 mm was used to take a leaf disc, from the middle of the leaf lamella from 18 leaves of the same age and position, from each replicate at three weeks intervals. Leaf discs were transferred immediately to individual, sealed plastic bags that were kept in an insulated box above ice packs until all leaf discs were taken. Fresh leaf discs were weighed before being placed in individual aluminum foil cups and placed in an oven for drying. Leaf discs were dried at 70°C for 48 h before reweighing them. SLW was calculated as dry weight of leaf disc per area of hole-punch, and the LWC was calculated as follows:

LWC (%) = 
$$\frac{\text{Leaf fresh weight-leaf dry weight}}{\text{Leaf fresh weight}} \times 100.....(3)$$

The number of plants was counted after complete emergence. At harvest, above-ground biomass, seed yield and mass of yield components were recorded, and kernel samples were taken for analysis. The number and weight of nodules were measured on five randomly selected plants after 50% flowering. Total peanut pod and seed yields recorded on plot basis were converted to kg ha<sup>-1</sup> for statistical analysis.

# 4.2.4. Plant and soil analysis

Fifteen leaf samples, including leaves measured for leaf chlorophyll content, around each station, were clipped at their base and stored for the determination of C, N, P, K and NO<sub>3</sub>-N. Phosphorus, K and NO<sub>3</sub>-N concentrations in plants were quantified at the Analytical Research Laboratories (ARL) in New Zealand. Nitrate-N in plant tissue was determined using 2% acetic acid as the extractant (Miller, 1998). Plant K content was determined after wet digestion with sulfuric acid by atomic

absorption spectrometry (Watson et al., 1990). Plant P content was determined photometrically in the same extract with the molybdenum blue method (Mills and Jones, 1996). Total plant N and C concentrations were determined using an elemental analyzer (ECS 4010 CHNSO Analyzer; Costech Analytical Technologies INC, Valencia, CA, USA) fitted with a Zero Blank Auto-sampler (Costech Analytical Technologies, INC). Stable isotope compositions of oven-dried seed samples were also determined using a ThermoFinnigan DeltaV<sup>PLUS</sup> Continuous-Flow Isotope Ratio Mass Spectrometer (EA-IRMS) at James Cook University's Cairns Analytical Unit. Stable isotope results are reported as per mil (‰) deviations from the VPDB reference standard scale for  $\delta^{13}$ C, and from the international air standard for  $\delta^{15}$ N. Precisions (S.D.) on internal standards were better than ±0.2 ‰ for both isotope determinations.

Soil cores from 0 - 30 cm were taken in row at the mid-point of the growing season and after harvesting. The roots were separated, soil dried in an oven to constant weight, and then ground using Split Phase Motor Grinding Mill to pass through a 2-mm sieve. SOC and total soil N contents were determined as for plants. Soil pH, exchangeable cations, cation exchange capacity (CEC), electrical conductivity (EC), NO<sub>3</sub>-N and NH<sub>4</sub>-N contents were determined by the ARL Pty. Ltd. in New Zealand. Soil pH was measured in H<sub>2</sub>O and 0.01 M CaCl<sub>2</sub> using pH meter and a 1:2.5 soil weight to extractant-volume ratio. The EC was determined by a conductivity meter on a 1:2.5 soil to water suspension ratio (Rayment and Higginson, 1992). Colwell P was measured on 1:50 soil solution extracts in 0.5 M sodium bicarbonate after mixing for 16 hrs. The extracted P was determined colorimetrically on centrifuged and filtered extracts, using a SEAL AQ2+ Discreet Analyzer (Seal Analytical Ltd, Fareham, Hampshire, UK) and the ammonium molybdate/ascorbic acid color reaction with potassium antimonyl tartrate was added to control the reaction rate (Rayment and Lyons, 2011). Exchangeable K, Na, Ca and Mg were determined using 1M ammonium acetate extraction buffered at pH 7, using mechanical shaking at a soil to solution ratio of 1:20 (Rayment and Higginson, 1992) and atomic absorption analysis. CEC was calculated as the sum of exchangeable K, Na, Ca and Mg. Soil NH<sub>4</sub>-N and NO<sub>3</sub>-N were determined colorimetrically by an automated photometer using 1M KCl extraction method (Rayment and Lyons, 2011).

#### 4.2.5. Statistical analysis

The data were subjected to analysis of variance using the general linear model procedure (PROC GLM) of SAS statistical package version 9.1 (SAS Institute 2003, Cary, NC). The total variability for each trait was quantified using the following model (Gomez and Gomez, 1984).

$$Y_{ij} = \mu + R_i + T_j + RT_{ij} + e_{ij}.....(4)$$

Where  $Y_{ij}$  is the measured value,  $\mu$  = grand mean,  $R_i$  is effect of the i<sup>th</sup> replication,  $T_j$  is effect of the j<sup>th</sup> treatment,  $RT_{ij}$  is the interaction, and  $e_{ij}$  is the variation due to random error. Means for the treatments (n = 5) were compared using the MEANS statement with the least significant difference (LSD) test at the 5% probability level. Linear regression analysis was performed using replicated data and treatment means between plant parameters, nutrient uptake, and soil nutrient contents following the SAS REG procedure. Principal component analysis (PCA) was performed after standardizing the data using the SAS PRINCOMP procedure to distinguish the treatments as a function of the soil management and determine the most important parameters to characterize them.

# 4.3. Results

# 4.3.1. Yield and plant growth

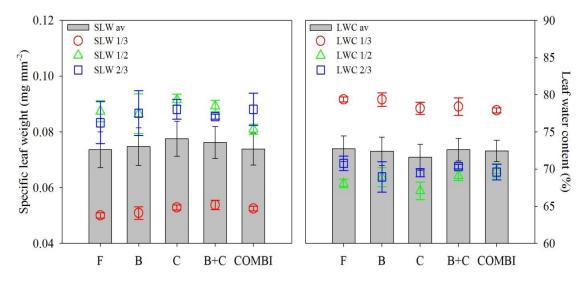
All 'organic' amendments significantly ( $p \le 0.05$ ) increased the peanut seed and pod yields and leaf chlorophyll contents of plants during the crop growth period, relative to the F control (Table 4.3). Neither plant number, nor specific leaf weight (SLW) at any growth stage of the plants, was significantly different between treatments. Applications of B, Com, B + Com, and COMBI increased the seed yield by 21, 18, 21 and 22%, and pod yield by 23, 17, 21 and 24%, compared to the F only treatment. The effect of treatments on leaf chlorophyll content became more pronounced as the crop grew. Compared to the F treatment, average leaf chlorophyll content increased by 7, 10, 10 and 9% in B, Com, B + Com, and COMBI amended soil, respectively. A significant difference was not observed between organic amendments for seed yield, total biomass or chlorophyll content. SLW increased as the crop growth progressed, being the highest near physiological maturity (Figure 4.3). Leaf water content was inversely correlated with specific leaf weight.

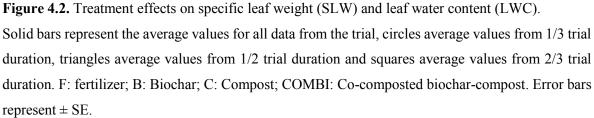
Root nodulation significantly improved crop performance, as total peanut pod yield was substantially higher in B, Com, B + Com and COMBI amended soil than in F only treated soil. Nodulation number (NN) and nodulation dry weights (NDW) differed significantly (p < 0.01) among treatments (Table 4.3). The highest average NN (112 plant<sup>-1</sup>) and NDW (107 mg plant<sup>-1</sup>) were recorded from B followed by the COMBI treatments, and the lowest scores were from the F treatment (Table 4.3). B, Com, B + Com and COMBI additions increased NN plant<sup>-1</sup> by 25, 9, 17 and 20%, and NDW by 25, 9, 15 and 18%, respectively, compared to the F treatment.

| Treatment   | SY                     | PY                     | Av. CHLC    | NN                  | NDW                       |
|-------------|------------------------|------------------------|-------------|---------------------|---------------------------|
|             | (kg ha <sup>-1</sup> ) | (kg ha <sup>-1</sup> ) | (SPAD-unit) | plant <sup>-1</sup> | (mg plant <sup>-1</sup> ) |
| Control (F) | 4167 b                 | 5617 b                 | 45.7 b      | 91 c                | 87.4 c                    |
| B + F       | 5048 a                 | 6910 a                 | 47.7 a      | 112 a               | 108.4 a                   |
| Com + F     | 4921 a                 | 6552 ab                | 48.4 a      | 101 b               | 95.3 bc                   |
| B + Com + F | 5023 a                 | 6786 a                 | 48.8 a      | 106 ab              | 100.5 ab                  |
| COMBI +F    | 5096 a                 | 6946 a                 | 48.0 a      | 109 ab              | 102.8 ab                  |
| p level     | 0.050                  | 0.025                  | 0.024       | 0.003               | 0.001                     |
| LSD (0.05)  | 646.1                  | 797.3                  | 1.7         | 8.3                 | 7.1                       |
| CV (%)      | 7.1                    | 6.5                    | 1.9         | 4.2                 | 3.8                       |

**Table 4.3.** Effects of soil amendments on peanut seed yield (SY), total pod yield (PY), leaf chlorophyll (SPAD unit), nodulation number (NN) and nodulation dry weight (NDW) per plant.

Within each column, means with different letters are significantly different at p < 0.05. Av. CHLC: Average chlorophyll content (average of six temporal measurements); LSD: Least significant difference; CV: Coefficient of variation.





### 4.3.2. Plant nutrient uptake

All organic amendments significantly (p < 0.05 and p < 0.01) improved leaf nutrient concentrations of C, N, P, K and NO<sub>3</sub>-N at the mid-growth stage. While, at the late growth stage, the treatments only significantly ( $p \le 0.05$ ) affected NO<sub>3</sub>-N uptake, and not C, N, P and K (Table 4.4). The highest plant C and total N concentrations of (44.1% and 4.1%, respectively) were obtained from B + Com, and the lowest from the F treatment (Table 4.4). Applications of B + Com and COMBI increased plant C concentrations by 20.5 and 16.4%, and plant N uptake by 28.1 and 21.9%, respectively, compared to F only treatment. Leaf P, K and NO<sub>3</sub>-N concentrations at the mid-growth stage ranged from 0.24 -0.42%, 2.8 - 4.2% and 371 - 877 mg kg<sup>-1</sup>, respectively, with the highest values being from B + Com for P and NO<sub>3</sub>-N, and COMBI for K uptake. The highest values for leaf N and P concentrations were obtained from the B + Com treatment, and the lowest K concentration from F treatment (Table 4.4). However, there was no statistically significant difference between B + Com and COMBI for these parameters. The respective N and P contents of plants in the B + Com and COMBI treatments were 1.3 and 1.2, and 1.8 and 1.7 times that of the F treatment. Foliar NO<sub>3</sub>-N, P and K concentrations ranged from 10.2 - 75 mg kg<sup>-1</sup>, 0.12 - 0.14% and 1.32 -1.60%, respectively. The variability among replications for NO<sub>3</sub>-N were large both at mid- and late-plant growth stages.

|             | Mid-growth stage leaf nutrient concentration |        |                        |                       |       | Late growth stage leaf nutrient concentration |      |                        |      |      |  |
|-------------|--|--------|------------------------|-----------------------|-------|---|------|------------------------|------|------|--|
|             | С  | N      | NO <sub>3</sub> -N     | Р                     | K     | С   | N    | NO3-N                  | Р    | K    |  |
| Treatment   | (%)  | (%)    | (mg kg <sup>-1</sup> ) | (g kg <sup>-1</sup> ) | (%)   | (%)   | (%)  | (mg kg <sup>-1</sup> ) | (%)  | (%)  |  |
| Control (F) | 36.6 c                                       | 3.2 b  | 623 b                  | 2.06 b                | 2.48  | 42.2  | 2.43 | 75.3 a                 | 0.12 | 1.40 |  |
| B + F       | 39.0 bc                                      | 3.7 ab | 371 c                  | 2.60 a                | 2.49  | 42.9  | 2.28 | 20.0 b                 | 0.13 | 1.46 |  |
| Com + F     | 40.7 abc                                     | 3.8 a  | 448 c                  | 2.64 a                | 2.49  | 42.3  | 2.23 | 12.2c                  | 0.14 | 1.60 |  |
| B + Com+ F  | 44.1 a                                       | 4.1 a  | 877 a                  | 2.66 a                | 2.51  | 43.0  | 2.36 | 10.2 c                 | 0.14 | 1.33 |  |
| COMBI + F   | 42.6 ab                                      | 3.9 a  | 872 a                  | 2.61 a                | 2.52  | 42.5  | 2.24 | 10.5 c                 | 0.14 | 1.32 |  |
| p level     | 0.045  | 0.038  | 0.001                  | 0.003                 | 0.965 | 0.65  | 0.63 | 0.001                  | 0.44 | 0.30 |  |
| LSD (0.05)  | 4.8  | 0.50   | 145                    | 0.18                  | 0.15  | 1.3   | 0.33 | 17.2                   | 0.03 | 0.31 |  |
| CV (%)      | 6.3  | 7.1    | 12.1                   | 3.9                   | 3.2   | 1.6   | 7.7  | 35.6                   | 10.5 | 11.7 |  |

**Table 4.4.** Effects of biochar, compost and their mixture on leaf nutrient concentration of peanut.

Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation.

The organic treatments had a significant effect on peanut seed  $\delta^{15}N$  composition, but not on  $\delta^{13}C$ , C, or total N contents (Table 4.5). The highest seed  $\delta^{15}N$  was obtained from the F only treatment, followed by B. The treatments showed similar effects on  $\delta^{13}C$  and C concentrations of peanut. The maximum N concentration was achieved from B + F and COMBI +F, and the lowest from B + Com + F treatment. Seed C: N ratios were similar for all treatments, although the highest value was obtained from B + Com + F treatment, which had the lowest N concentration.

| Treatments  | <u> </u> | N (%)   | C: N ratio  | δ <sup>13</sup> C | δ <sup>15</sup> N |
|-------------|----------|---------|-------------|-------------------|-------------------|
| Treatments  | C (70)   | IN (70) | C. IN Tatio | 0 °C              | 0 ' IN            |
| Control (F) | 61.7     | 5.09    | 12.2        | -27.3             | 1.64 a            |
| B + F       | 61.7     | 5.29    | 11.7        | -27.6             | 1.49 ab           |
| Com +F      | 61.7     | 5.02    | 12.3        | -27.3             | 1.18 bc           |
| B + Com + F | 61.8     | 4.79    | 12.9        | -27.2             | 1.00 c            |
| COMBI + F   | 61.6     | 5.27    | 11.7        | -27.3             | 1.04 bc           |
| p level     | 0.99     | 0.30    | 0.45        | 0.82              | 0.04              |
| LSD (0.05)  | 1.83     | 0.55    | 1.66        | 0.69              | 0.46              |
| CV (%)      | 1.57     | 5.74    | 7.27        | -1.35             | 19.2              |
|             |          |         |             |                   |                   |

**Table 4.5.** Changes of peanut seed C, total N, C: N ratio and C and N isotope composition ( $\delta^{13}$ C and  $\delta^{15}$ N) to biochar and compost applications.

Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation

# 4.3.3. Soil physicochemical characteristics

Soil analysis results at the mid-plant growth stage and after harvesting, indicated that application of all organic amendments significantly ( $p \le 0.05$  and  $p \le 0.01$ ) increased SOC, total N, available P, NO<sub>3</sub>-N and NH<sub>4</sub>-N concentrations relative to soil that only received inorganic fertilizer, but not soil pH (Table 4.6). At harvest, soil C: N ratio significantly responded to the organic treatments, but not at the mid-growth stage of the plants. Significant differences were observed between the treatments for SOC at the mid-growth stage. COMBI addition resulted in the highest SOC, N and NO<sub>3</sub>-N in the soil (Table 4.6). At the mid-growth stage, in B, Com, B + Com and COMBI amended soil, SOC content increased by a factor of 1.3-1.4, and the total N content increased by a factor of 1.1-1.3, compared to the control. The mean SOC decreased in the order COMBI > B > B + Com > Com > F at the mid-plant growth stage. Although, after B and COMBI additions, NO<sub>3</sub>-N concentration is highly variable in soils, the soil NO<sub>3</sub>-N and NH<sub>4</sub>-N concentrations significantly increased by a factor

of 1.7 and 1.9, and 2.0 and 1.9, respectively, relative to F at mid plant growth stage. Colwell soil P was higher by a factor of 1.1, 1.2 and 1.2, owing to B, B + Com and COMBI additions, respectively, in comparison with the F and Com treatments. While the Colwell P in the compost was considerably higher than that in biochar, the available P in the soil was lower in the compost treatments than the biochar treatment. In general, lower total soil N and soil C contents were obtained after harvesting than at mid-plant growth stage, whereas the soil NO<sub>3</sub>-N content was lower at mid-plant growth stage than after harvesting. This suggests that the demand and uptake for NO<sub>3</sub>-N during peak plant growth stage was higher than late the plant growth stage. In contrast, higher extractable Colwell P and NH<sub>4</sub>-N concentrations were recorded at mid-plant growth stage than after harvesting.

The average volumetric soil water content (SWC) was significantly (p < 0.01) improved by the organic amendments during the plant growth stage. The maximum SWC was obtained from B treated soil followed by the COMBI, and the lowest was from the F treatment (Table 4.6). At the mid-growth stage of plants, the average SWC was higher by 30, 14, 18 and 24% for the soils amended with B, Com, B + Com and COMBI, respectively, than the F treatment. The average SWC decreased in the order B > COMBI > B + Com > Com > F. However, by harvest the treatment effects were no longer statistically significant. Soil amendments had significant ( $p \le 0.05$ ) effects on exchangeable K, Mg, Al and CEC at mid-plant growth stage, but not on exchangeable Na, Ca and EC. Exchangeable Na was the only element significantly affected by the treatments at harvest. At the mid-growth stage, exchangeable K, Mg and CEC increased by a factor of 1.2, 1.4 and 1.2, respectively, by adding biochar, compared with the control. At the mid-growth stage, contents of plant-available nutrients increased in the order K > Mg > CEC > Al with organic treatments (Table 4.7). Soil bulk density was not significantly affected by the treatments (Table 4.6).

|                           | SBD                   | SWC (%) | pН                 | Soil C  | Ν       | C: N    | Nutrient cor | ncentration (m     | g kg <sup>-1</sup> ) |
|---------------------------|-----------------------|---------|--------------------|---------|---------|---------|--------------|--------------------|----------------------|
| Treatments                | $(g \text{ cm}^{-3})$ |         | (H <sub>2</sub> O) | (%)     | (%)     | ratio   | Colwell P    | NH <sub>4</sub> -N | NO <sub>3</sub> -N   |
| Mid-point                 |                       |         |                    |         |         |         |              |                    |                      |
| Control (F)               | 0.79                  | 17.9 c  | 7.0                | 0.93 c  | 0.07 b  | 13.3    | 52.8 b       | 8.4 c              | 3.1 b                |
| $\mathbf{B} + \mathbf{F}$ | 0.64                  | 23.3 a  | 6.9                | 1.25 a  | 0.08 ab | 15.6    | 58.7 ab      | 16.7 a             | 5.3 a                |
| Com + F                   | 0.74                  | 20.4 b  | 6.9                | 1.15 b  | 0.08 ab | 14.4    | 52.7 b       | 13.2 b             | 5.6 a                |
| B + Com + F               | 0.58                  | 21.1 b  | 6.9                | 1.19 ab | 0.08 ab | 14.9    | 63.0 a       | 14.1 ab            | 5.5 a                |
| COMBI + F                 | 0.82                  | 22.2 b  | 6.9                | 1.23 a  | 0.09 a  | 13.7    | 60.8 ab      | 16.0 ab            | 6.0 a                |
| <i>p</i> level            | 0.070                 | 0.002   | 0.982              | 0.001   | 0.03    | 0.134   | 0.050        | 0.002              | 0.011                |
| LSD (0.05)                | 0.18                  | 1.9     | 0.48               | 0.06    | 0.01    | 1.6     | 8.6          | 3.0                | 1.4                  |
| CV (%)                    | 8.9                   | 4.8     | 3.6                | 2.8     | 6.1     | 5.8     | 7.8          | 11.7               | 14.8                 |
| End-point                 |                       |         |                    |         |         |         |              |                    |                      |
| Control (F)               | 0.97                  | 26.9    | 6.6                | 1.00b c | 0.08 a  | 13.4 c  | 42.3 b       | 7.07 bc            | 7.97 c               |
| B + F                     | 0.91                  | 28.3    | 6.7                | 1.08 ab | 0.08 a  | 12.4 c  | 41.0 b       | 11.73 a            | 10.77 ab             |
| Com + F                   | 0.93                  | 27.2    | 6.8                | 1.08 ab | 0.07 ab | 15.0 b  | 43.3 b       | 5.87 c             | 9.27 bc              |
| B + Com + F               | 0.96                  | 27.4    | 6.9                | 1.01 bc | 0.06 b  | 16.1 ab | 55.3 a       | 6.77 c             | 9.30 bc              |
| COMBI + F                 | 0.92                  | 28.8    | 6.8                | 1.12 a  | 0.07 ab | 16.7 a  | 44.7 b       | 9.47 ab            | 12.13 a              |
| <i>p</i> level            | 0.623                 | 0.281   | 0.599              | 0.019   | 0.045   | 0.001   | 0.024        | 0.005              | 0.050                |
| LSD (0.05)                | 0.09                  | 2.11    | 0.39               | 0.019   | 0.01    | 1.35    | 8.26         | 2.65               | 2.67                 |
| CV (%)                    | 5.3                   | 4.03    | 3.1                | 4.57    | 9.06    | 4.88    | 9.68         | 17.22              | 14.33                |

Table 4.6. Effects of soil amendments on soil physicochemical properties at the grain filling stage and trial end-point.

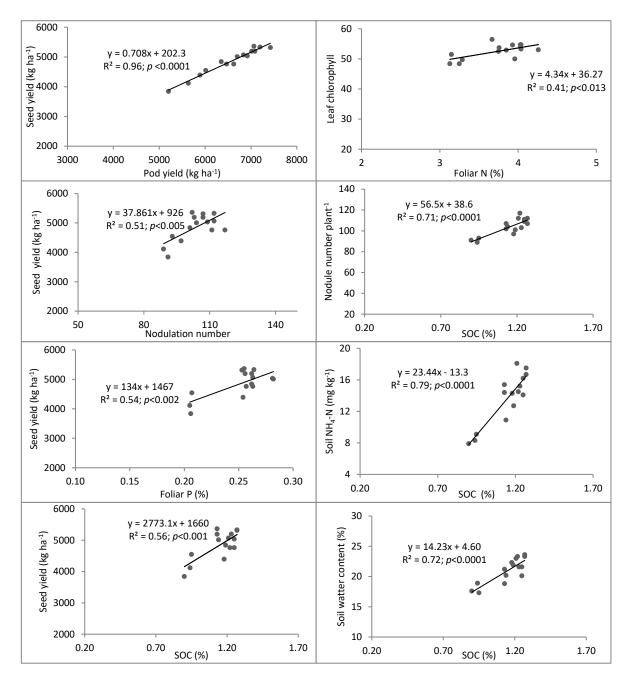
Within each column, means with different letters are significantly different at p < 0.05. SBD: Soil bulk density; SWC: Soil water content; LSD: Least significant difference; CV: Coefficient of variation.

| Exchangeable cations (cmol(+)/kg) |       |        |         |         |        |         |                          |  |  |  |
|-----------------------------------|-------|--------|---------|---------|--------|---------|--------------------------|--|--|--|
| Treatments                        | Ca    | Mg     | K       | Na      | CEC    | Al      | EC (dS m <sup>-1</sup> ) |  |  |  |
| Mid-point                         |       |        |         |         |        |         |                          |  |  |  |
| Control (F)                       | 6.8   | 1.30 b | 0.72 c  | 0.04    | 8.9 b  | 0.01 b  | 0.03                     |  |  |  |
| $\mathbf{B} + \mathbf{F}$         | 8.0   | 1.49 a | 0.81 bc | 0.05    | 10.3 a | 0.03 a  | 0.04                     |  |  |  |
| Com + F                           | 8.1   | 1.51 a | 0.82 bc | 0.04    | 10.5 a | 0.02 ab | 0.04                     |  |  |  |
| B + Com + F                       | 7.9   | 1.51 a | 0.93 ab | 0.04    | 10.4 a | 0.02 ab | 0.04                     |  |  |  |
| COMBI + F                         | 7.7   | 1.57 a | 1.00 a  | 0.04    | 10.3 a | 0.02 ab | 0.04                     |  |  |  |
| <i>p</i> level                    | 0.093 | 0.025  | 0.05    | 0.165   | 0.025  | 0.020   | 0.540                    |  |  |  |
| LSD (0.05)                        | 1.0   | 0.15   | 0.18    | 0.01    | 0.93   | 0.01    | 0.01                     |  |  |  |
| CV (%)                            | 7.0   | 5.4    | 11.4    | 15.7    | 4.9    | 18.9    | 19.4                     |  |  |  |
| End-point                         |       |        |         |         |        |         |                          |  |  |  |
| Control (F)                       | 8.0   | 1.47   | 0.70    | 0.02 bc | 10.3   |         | 0.04                     |  |  |  |
| $\mathbf{B} + \mathbf{F}$         | 8.0   | 1.41   | 0.77    | 0.03 ab | 10.2   |         | 0.04                     |  |  |  |
| Com + F                           | 8.2   | 1.47   | 0.80    | 0.02 bc | 10.5   |         | 0.04                     |  |  |  |
| B + Com + F                       | 8.5   | 1.52   | 0.91    | 0.02 c  | 11.0   |         | 0.04                     |  |  |  |
| COMBI + F                         | 7.9   | 1.48   | 0.81    | 0.04 a  | 10.2   |         | 0.05                     |  |  |  |
| <i>p</i> level                    | 0.961 | 0.955  | 0.247   | 0.026   | 0.911  |         | 0.648                    |  |  |  |
| LSD (0.05)                        | 2.28  | 0.32   | 0.19    | 0.01    | 2.14   |         | 0.012                    |  |  |  |
| CV (%)                            | 14.97 | 11.44  | 12.53   | 20.59   | 10.91  |         | 15.0                     |  |  |  |

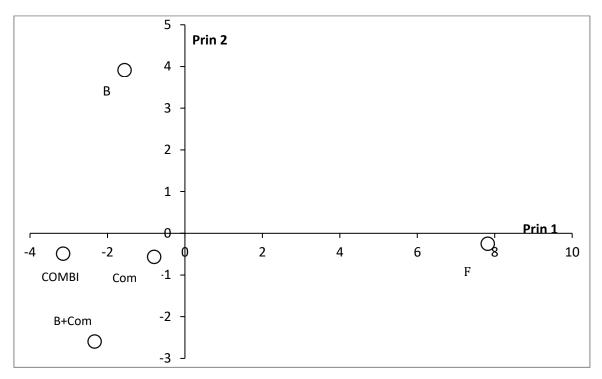
**Table 4.7.** Effects of soil amendments on soil chemical properties at the grain filling stage and trial end-point.

Within each column, means with different letters are significantly different at p < 0.05. LSD: Least significant difference; CV: Coefficient of variation.

Seed yield was significantly ( $p \le 0.01$ ) positively correlated with pod yield, NN, mid-growth stage leaf P concentration, and SOC ( $R^2 = 0.96$ , 0.52, 0.54 and 0.56, respectively), under the various treatments (Figure 4.4). The improvement in SOC, due to the addition of organic amendments, showed significant positive correlations with SWC, NH<sub>4</sub>-N and NN ( $R^2 = 0.72$ , 0.79 and 0.86, 0.71, respectively; Figure 4.4). The results of principal component analysis (PCA) revealed that the first three principal components (Prin1-Prin3) accounted for ~94% of the total variation between the treatments, of which ~85% was contributed by the Prin1 and Prin2 (Table 4.8). The bi-plot of Prin1 and Prin2 showed the clustering of treatments F, B, Com, B + Com, and COMBI (Figure 4.5). Characters with larger absolute values within the first principal component influenced the clustering more than those with lower absolute values. The differentiation of the treatments into different clusters was dictated by the cumulative effects of several characters. Thus, 24 out of 30 characters in the PRIN1 individually contributed substantial effects (-0.156-0.223) to the total variation of the treatments (data not shown).



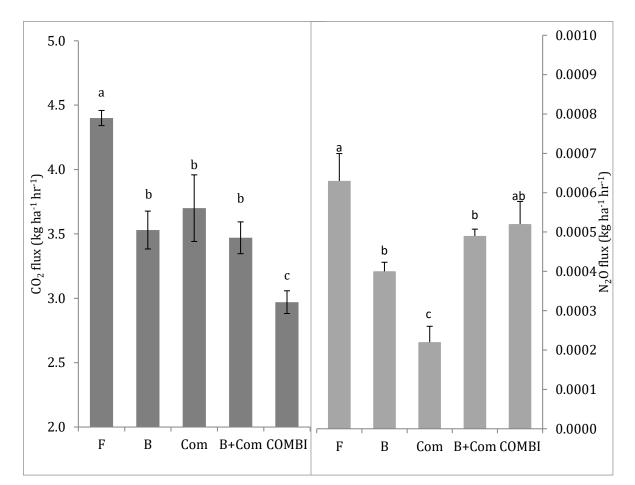
**Figure 4.3.** Correlations of plant parameters, plant nutrient concentration and soil physicochemical properties (n = 15) tested at five soil fertility treatments. Note: SOC: Soil organic carbon; NN: Nodulation number.



**Figure 4. 4.** Plot of principal component one and principal component two (Prin1 and Prin2) in five treatments. Note: F: Fertilizer; B: Biochar; Com: Compost; COMBI: Co-composted biochar-compost.

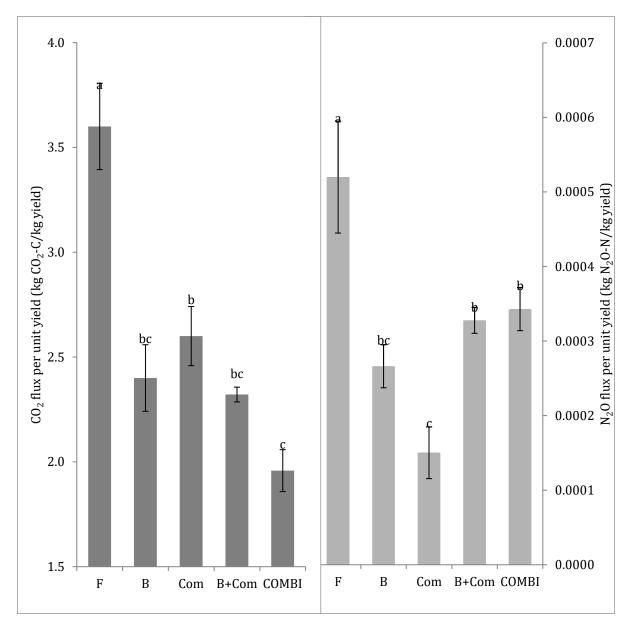
# 4.3.4. Greenhouse gas fluxes

Measurement of soil greenhouse gas fluxes indicated that the average emissions of  $CO_2$  were highest in the F treatment and lowest in the COMBI treatment. Organic amended soil reduced the emission of  $CO_2$  by 16 - 33% compared to the F treatment (Figure 4.6). As a consequence of higher total  $CO_2$ flux over the trial, coupled to lower crop yield, the amount of  $CO_2$  produced per unit of crop yield was significantly higher for the F treatment compared to organic amendments (Figure 4.7). This was approximately 28 - 46% more  $CO_2$ -C produced per unit of peanut produced in the F treatment than the organic amendments. In general,  $CO_2$ -C fluxes from soils containing amendments were lower than the F treatment. A significant exception was measured in March 2014 when  $CO_2$ -C fluxes from the B + Com and Com treatments were approximately 0.2 - 0.3 kg ha<sup>-1</sup> hr<sup>-1</sup> more than F (Figure 4.7).

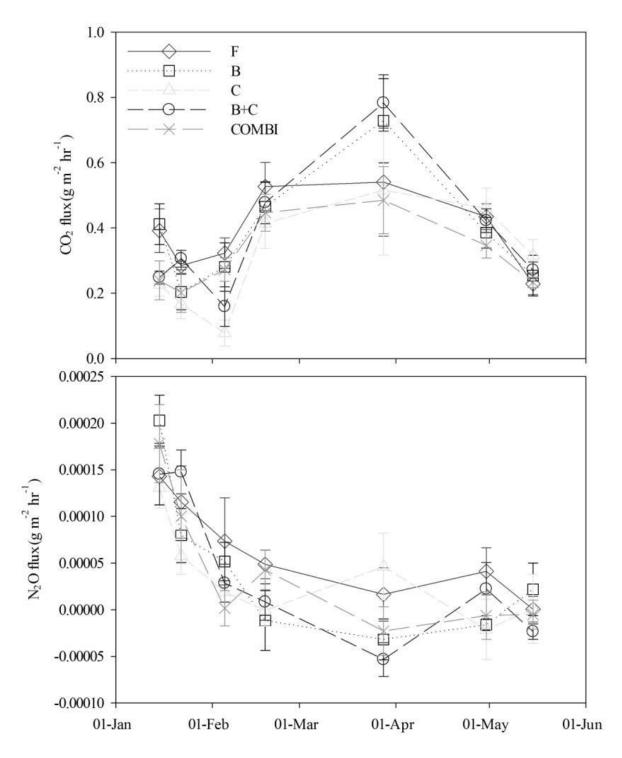


**Figure 4.5.** The average fluxes of CO<sub>2</sub> and N<sub>2</sub>O (kg ha<sup>-1</sup> hr<sup>-1</sup>) from all treatments including all data from all sampling dates. Note: LSD (p = 0.05) = 0.29 and 0.0001, and CV = 4.3 and 15.2 for CO<sub>2</sub> and N<sub>2</sub>O, respectively). Columns with the same letter are not significantly different at p = 0.05. Error bars represent ± 1 SE.

Fluxes of N<sub>2</sub>O were highest in the F treatment and lowest in the Com treatment. Although, all amended plots did reduce N<sub>2</sub>O flux relative to the F, with nearly 17-65% less N<sub>2</sub>O produced per unit of peanut produced in the organic amended than F amended soil (Figure 4.6). N<sub>2</sub>O fluxes, per unit peanut yield, were lower in all organic amended treatments than the F treatment (Figure 4.7). The organic amendments resulted in a 34 - 71% reduction in N<sub>2</sub>O emissions per unit yield, compared to the F only treatment (Figure 5). N<sub>2</sub>O flux in the B, Com and B + Com treatments exceeded that in the F treatment initially, but quickly dropped below F levels. After initially greater fluxes, only the Com treatment, again, exceeded the F treatment N<sub>2</sub>O fluxes in March and May 2014 (Figure 4.8).



**Figure 4.6.** Units of CO<sub>2</sub>-C and N<sub>2</sub>O-N produced per unit of peanut seed yield (kg CO<sub>2</sub>-C and N<sub>2</sub>O-N ha<sup>-1</sup>/kg yield ha<sup>-1</sup>). Note: LSD (p = 0.05) = 0.41 and 0.0001, and CV = 8.6 and 18.9 for CO<sub>2</sub> and N<sub>2</sub>O, respectively). Columns with the same letter are not significantly different at p = 0.05. Error bars represent ± 1SE.



**Figure 4.7.** The absolute increase/decrease in CO<sub>2</sub>-C and N<sub>2</sub>O-N flux from the four amendments compared to fertilizer only. Where positive values indicate amounts above the concurrent F value and negative values indicate amounts below the concurrent F values. Error bars represent  $\pm 1$  SE.

# 4.4. Discussion

# 4.4.1. Response of peanut yield and plant growth to soil amendments

The application of organic amendments in this study had a significantly positive effect on the growth and yield of peanut, when applied together with normal fertilizer rate. This supports our second hypothesis whereby applications of organic amendments improved plant growth and crop yield. Our results, indicating an increase of 15 to 21% after organic amendment, were similar to those obtained in other studies. Yamato et al. (2006) reported a significant increase in peanut yield up to 50% following the application of 10 t ha<sup>-1</sup> charred bark of *Acacia* biochar, with 75 kg ha<sup>-1</sup> of each NPK, for an infertile soil in Indonesia. Peanuts grown on 15 t ha<sup>-1</sup> farmyard manure biochar amended soil only in the first year along with 160, 16 and 42 kg ha<sup>-1</sup> NPK resulted in yield increment of about 7% in the first year and 35% in the second year compared to only NPK fertilized plots (Islami et al., 2011).

Schulz et al. (2013) found that addition of 100 t ha<sup>-1</sup> composted biochar to sandy and loamy soil increased growth of oat plants. Yield increases have also been reported for maize with the applications of 20 t ha<sup>-1</sup> wood biochar plus 159, 30 and 138 kg NPK ha<sup>-1</sup> (Major et al., 2010) and 4 t ha<sup>-1</sup> maize cob biochar plus 154, 56 and 28 kg NPK ha<sup>-1</sup> (Cornelissen et al., 2013). Addition of 30 t ha<sup>-1</sup> biochar plus 122 and 25 NP ha<sup>-1</sup> resulted in a wheat yield increment of 28-39% (Vaccari et al., 2011). In contrast, Sukartono et al. (2011) reported that addition of 15 t ha<sup>-1</sup> cattle manure, cow-dung biochar and coconut shell biochar with 135,33 and 62 kg ha<sup>-1</sup> NPK, on a sandy soil in Indonesia, resulted in 6.0, 5.9 and 5.7 t ha<sup>-1</sup> maize, respectively. A review by Liu et al. (2013) concluded that, in spite of significant variation between individual studies, legumes generally exhibit a better response to biochar application than other crops, with average increases of 40% for seed yield and 25% for total biomass. For instance, the average increase in yield across all studies after biochar application was ~30, 29 and 14%, respectively, for legumes, vegetables and grasses, and only 8, 11 and 7%, respectively, for maize, wheat and rice.

Biochar soil amendment showed distinct crop productivity differences in relation to soil texture where biochar increased yield by about 30, 16, 7 and 7%, respectively, for sand, clay, silt and loam soil (Liu et al., 2013). In this study, significant differences were observed in plant growth and yield between inorganic and organic treatments, but not amongst organic amendments. The yield improvement observed may be attributable to the improved nutrient and water retention capacity of the organic

amendments and associated nutrient input, relative to that in fertilizer-only treated soil. Thus, the increase in yield due to organic amendments was in conformity with the expected improvements.

Chlorophyll content of leaves is a practical indicator of both potential photosynthetic productivity and general plant vigor, which is related to the N concentration in green plants and serves as a measure of the response of crops to N fertilizer application and soil nutrient status (Dana and Juan, 2004). The soil fertility effect on chlorophyll content in this study was significant, in that chlorophyll content was considerably higher than plots amended with mineral fertilizer. All organic treatments with inorganic fertilizer led to a modest, but significant, improvement of leaf chlorophyll content. This is associated with increased soil available N and leaf N and may partly contribute to yield improvement. Other studies reported that the application of 40% (w/w) bamboo biochar increased chlorophyll contents of ryegrass by 20 - 32% (Hua et al., 2012), and maize by 8 - 12% due to 10 t ha<sup>-1</sup> willow biochar plus 140, 35 and 90 kg ha<sup>-1</sup> NPK, and 20 t ha<sup>-1</sup> compost with the same NPK rate (Agegnehu et al., 2015b), compared to fertilizer only. Minotta and Pinzauti (1996) found that total leaf chlorophyll content was almost doubled at high versus low soil fertility status with the incidence of high light environment.

We observed a trend of increasing leaf chlorophyll content with more advanced crop growth stage, which contributes greatly to the performance of the crop. This may be due to improved N concentration in the growing plants supplied by the organic amendments. In spite of being statistically insignificant, the highest SLW was recorded from the B and Com treatments, which is in agreement with the findings of Agegnehu et al. (2015b). SLW increased across the treatments, as the growth of plants progressed, with the difference being highest at plant maturity. SLW is related to the resistance or susceptibility of plant leaves to insect attack, with higher SLW providing higher resistance (Steinbauer, 2001).

# 4.4.2. Soil amendment effects on plant nutrient uptake

Organic amendments significantly improved total foliar N and C contents at the mid-growth stage, but not at the late growth stages of the plants, indicating that N is not limiting; which is not surprising for legumes, as they can fix atmospheric N<sub>2</sub>. Foliar NO<sub>3</sub>-N, P and K concentrations were significantly increased due to the organic amendments during the mid-plant growth stage, with the highest being from B + Com followed by COMBI. Higher NO<sub>3</sub>-N and P concentrations in the crop implies that organic amended soil maintained higher concentrations of these nutrients in the soil solution. The decrease in NO<sub>3</sub>-N, P and K concentrations with the plant age may be associated with progressive assimilation of these nutrients in biomass. Fageria (2014) reported a similar pattern of N, Ca and Mg decrease in bean plants with plant age.

Applications of B and Com, singly or in combination, increased foliar P concentration by 11 - 19% at the mid-growth stage of plants compared to the F only treatment, implying that P is a limiting nutrient for growth at this site, as is commonly the case for most legumes. The use of biochar and compost can supply the soil with P and improve its availability, by reducing sorption and leaching. Other studies have reported that biochar and compost amended Ferralsols resulted in lower leaching and higher P uptake by plants (Agegnehu et al., 2015b; Lehmann et al., 2003). Legume species differ widely in their ability to grow in soils of low P status. Hocking et al. (1997) hypothesized that white lupin, and to a lesser extent pigeon pea, can access soil P from a pool that is relatively inaccessible to other legume species, but this is not the case for peanuts.

The stimulating effect of biochar on crop yield has been attributed to various mechanisms, depending on biochar, crop, soil type and trial conditions (Atkinson et al., 2010; Liu et al., 2013; Spokas et al., 2012). In our study, soil pH did not affect growth and yield of peanut, suggesting that the peanut yield response was due to the effect of biochar on nutrient availability and root nodulation. B, Com and their mixture increased the availability of some essential elements in the soil, including P, K and Mg. Because peanut can acquire N via nodules, P appears to be the major limiting nutrient for peanut yield on Ferralsols. Moreover, P supply is essential for the formation, development and function of nodules (Agegnehu and Tsige, 2006; Tang et al., 2001), thereby stimulating biological N fixation (Rondon et al., 2007). In our study, the organic amendments added a significant amount of available P (6-19 mg kg<sup>-1</sup>), inferring that organic amendments can provide a slow-release P pool, additional to conventional fertilizer, through mineralization reactions (Slavich et al., 2013; Wang, 2012). This input contributes to the increase in soil available P and can be critical to alleviate the P limitation on peanut crops. Another common mechanism of biochar application to increase P availability is the liming effect that decreases P adsorption and facilitates P desorption from Al and Fe oxides (Cui et al., 2011; Glaser et al., 2002; Lehmann and Rondon, 2006; Yuan and Xu, 2011). However, this mechanism did not contribute to the increment of soil available P in our study, due to the limited liming capacity of the biochar used in the present study, as demonstrated by the absence of any change in soil pH associated with biochar addition.

The treatment effect was significant for peanut seed isotope  $\delta^{15}N$  composition, but not for  $\delta^{13}C$ , seed C and N content, and C: N ratio. Significantly higher  $\delta^{15}N$  composition was obtained from F and B additions than other treatments, with the lowest values recorded from compost-containing treatments,

suggesting promotion of atmospheric nitrogen fixation by the compost-containing treatments. The organic amendments had significant effects on root nodulation in that B, B + Com, and COMBI amended soil significantly increased NN and NDW per plant. Mnalku (2011) found NN of 30 - 121 mg per plant and NDW of 36 - 108 mg per plant. Other studies have shown that biochar can promote rhizobia nodulation of, and biological N fixation by, legume species (Biederman and Harpole, 2013; George et al., 2012; Quilliam et al., 2013; Tagoe et al., 2008; Xu et al., 2015).

The main reason for the higher root nodulation accompanying the organic amendments may be greater boron and molybdenum availability, whereas greater K, Ca and P availability, as well as higher pH, and lower N availability and Al saturation, may have contributed to a lesser extent (Rondon et al., 2007). Root nodulation may be associated with other nutrients contributed by biochar and compost. For example, cobalt is an essential nutrient required by root nodule bacteria, boron is essential for legumes, and molybdenum is essential for N fixation because of its specific role in nitrogenase. Nutrient deficiencies in root nodule bacteria can affect a range of physiological functions, such as nutrient uptake, growth regulation and gene function (Sessitsch et al., 2002). Since peanut plants can fix N, leaves in the organic-amended soil displayed higher level of N (N > 3.2%) during peak growth stage (e.g., pegging stage), compared to the F treatment. In this case, increased level of total soil N, including NO<sub>3</sub>-N and NH<sub>4</sub>-N following biochar and compost amendment, did enhance leaf N. We observed a trend of increasing leaf N content from B, Com and B + Com amended soil, which might contribute to crop performance. Overall, enhanced N supply and nodulation, associated with the organic amendments, appears to improve peanut leaf N on a Ferralsol and this, consequently, may improve photosynthesis and yield.

# 4.4.3. Soil amendment effects on soil nutrient dynamics and GHG emissions

Applications of B, Com, and their mixture appeared to address the major nutrient deficiencies of the soil at the trial site, which is consistent with the results of the pre-planting soil analysis results. SOC is an indicator of the soil organic matter content (SOM), which aerates the soil and helps retain water and nutrients. SOM also provides substrate for soil microbial biomass, which in turn, can make nutrients more plant-available. Studies have demonstrated that compost addition increased the quantity as well as the quality of SOM, thus improving soil quality (Fischer and Glaser, 2012; Rivero et al., 2004). Another study indicated that a sandy soil amended by manure and coconut shell biochars increased SOC and persisted after the harvest of the second crop, while SOC in manure treated soil was not significantly different from the fertilizer only treatment after the first and second maize harvest (Sukartono et al., 2011).

In this study, results of soil analysis at the mid-growth stage of plants indicated that B and COMBI amended soil increased SOC content by a factor of 1.4, and total N content by a factor of 1.3, compared with the initial SOC and N contents. Recent studies have reported that SOC was significantly increased due to the applications of different biochars (Angst et al., 2014; Slavich et al., 2013; Stavi and Lal, 2013). However, the SOC and total N contents of the soil in this study (although increased) were still below levels considered sufficient (1.8% for SOC and 0.25% for N), which may be due to high N uptake by plants, and high contents of above- and below-ground plant residues. This research suggests that the Ferralsol of the study area (humid, tropical Queensland) is deficient in SOC and total N. Subsequently, this suggests that successive applications of B and B + Com will be beneficial to, over time, enhance the organic matter content, nutrient status, and nutrient and water retention capacity of the soil.

Nitrate is the form of soil N that is most readily available for plant uptake. Use of COMBI resulted in the highest total soil N and NO<sub>3</sub>-N, implying that co-composted biochar has been more effective in the retention of N during the composting process and in making the N plant available in the soil than compost or biochar alone. Co-composting of poultry manure and farmyard manure with biochar has previously been shown to reduce the losses of N in the mature composts (Dias et al., 2010; Prost et al., 2013). Adding acid biochar and co-composting reduced NH<sub>3</sub>-N loss by 58 to 63% (Doydora et al., 2011), and total N losses by up to 52% (Steiner et al., 2010). A review by Clough et al. (2013) showed that the available N in biochar amended soil was affected by complex processes, including absorption, leaching, N mineralization, nitrification, and N immobilization. Biochar application often immobilizes soil mineral N because of the input of labile C and increased soil C:N ratio (Ippolito et al., 2012), or fixes N through absorption (Reverchon et al., 2014), thereby reducing the fraction of N that is available for plants from plant-based biochar (Gaskin et al., 2010). Studies have also shown that N in plant-based biochars may be less available than that in biochar from animal manures (Chan et al., 2008; Tagoe et al., 2008). Thus, in some cases biochar application can decrease soil available N and plant tissue N concentration (Bargmann et al., 2014; de Sousa et al., 2014). However, Jones et al. (2012) reported that biochar addition had limited effects on the turnover of <sup>14</sup>C-labelled SOC and dissolved organic C and N, and no long-term effect on N mineralization, NH<sub>3</sub> volatilization, denitrification or NH<sub>4</sub> sorption.

Since the N absorption and immobilization effects of biochar were small in our study, reduction of leaching due to biochar addition may have played a critical role in improving N retention (Agegnehu et al., 2015b; Ding et al., 2010; Zheng et al., 2013). Biochar and COMBI addition increased soil N by 14% and 29%, respectively. This may be due to the amount of N added and a low C: N ratio of

the soil, which limits N immobilization. A soil C:N ratio of over 32 promotes N immobilization (Bruun et al., 2012; Novak et al., 2010), by a significantly higher amount than observed in the present study. This study demonstrated positive effects of biochar, compost, and their mixtures on SOC content, nutrients levels and water retention capacity of Ferralsol under field conditions, in agreement with the findings of Liu et al. (2012). Soil water contents recorded throughout the crop growing period were significantly lower in the F than in the organic treatments, suggesting that organic amendments enhanced water retention capacity of the soil, which confirms recent findings (Barrow, 2012; Troy et al., 2014).

The amendment of the soil with biochar and compost significantly improved the CEC of the soil, indicating that the retention of non-acidic cations by the soils increased. CEC is an important parameter in retaining inorganic nutrients, such as  $K^+$  and  $NH_4^+$  in soil (Lee et al., 2013), and biochar has been associated with the enhancement in CEC of some biochar-amended soils (Glaser et al., 2001; Van Zwieten et al., 2010), thereby increasing the availability and retention of plant nutrients in soil and potentially increasing nutrient use efficiency. Biochar is not only a soil conditioner that increases CEC, but may act as a fertilizer itself. Biochar contains ash, and so adds nutrients, such as K, Ca and Mg to the soil solution, increasing the pH of the soil and providing readily available nutrients for plant growth (Glaser et al., 2002). Recent studies suggest that a K fertilization effect associated with biochar is critical for promoting growth, biological N fixation and the competitive ability of legume species (Mia et al., 2014; Oram et al., 2014; Xu et al., 2015). In this study, high K concentrations in the Com and COMBI treatments may also have contributed to the peanut yield response associated with the organic amendments, where a significant increase in soil available K was observed. In addition to P and K, the organic amendments also increased the availability of other plant nutrients, suggesting that a general improvement of soil fertility may contribute to stimulation of the yield of peanut.

Seed yield was positively correlated with pod yield, leaf chlorophyll, SWC, SOC, plant available nutrients and uptake. The direct effect on the performance of the peanut crop of available soil nutrients and plant nutrient uptake from B, B + Com and COMBI amended soil exceeded the direct effect of available nutrients and nutrient uptake from the Com and F treatments. Correlation of seed yield with SOC was the highest, and the B, B + Com and COMBI amended soil improved the retention and availability of nutrients in this study. Available soil Mg, NO<sub>3</sub>-N and NH<sub>4</sub>-N had a more significant influence on the seed yield, chlorophyll content and NN than other nutrients, suggesting that these nutrients were most limiting to yield. Recent studies have also shown positive linear correlations

between soil chemical characteristics, shoot and root growth of maize and wheat as a result of addition of different biochar types (Agegnehu et al., 2015b; Solaiman et al., 2012).

Principal component analysis (PCA) indicated that Prin1 and Prin2 provided a reasonable summary of the data, accounting for about 78% of the total variance. In this study, several characters in the first eigenvector individually contributed similar effects to the total variation of the treatments, suggesting that the first component was, primarily, a measure of most characters. Thus, the differentiation of the treatments into different clusters was dictated by the cumulative effects of several characters. Other studies also compared the effects of different soil amendments using PCA (Agegnehu et al., 2015b; Sena et al., 2002). According to the bi-plot of Prin1 and Prin2, the treatments can be grouped into four classes, that is, F, B and Com, with COMBI and B + Com behaving similarly, suggesting there is no difference between COMBI and B + Com added together at the point of application to the soil.

The fluxes of CO<sub>2</sub> and N<sub>2</sub>O were generally lower in organic-amended plots than the control, but the magnitudes of these differences were not constant across the trial. For example, the B and B + Com, at the beginning of April, exceeded the control CO<sub>2</sub> flux by a significant amount, but this declined and the fluxes from these treatments remained similar for the rest of the trial. This implies that any labile carbon remaining in the biochar was quickly utilized and left only a relatively refractory pool from then on. Similarly, N<sub>2</sub>O flux was highest initially for B treatment, but this was finally significantly reduced. Kammann et al. (2012) reported that wood chip biochar addition significantly reduced CO<sub>2</sub> and N<sub>2</sub>O emissions and improved the GHG-to-yield ratio under field-relevant conditions. A similar study showed that soil N<sub>2</sub>O fluxes were from 26% to 79% lower in biochar treated plots than N<sub>2</sub>O fluxes in control plots (Castaldi et al., 2011). A study by Zhang et al. (2012b) also indicated that use of 40 t ha<sup>-1</sup> wheat straw biochar decreased total global warming potential of CH<sub>4</sub> and N<sub>2</sub>O, by about 42% without N fertilization, and 48% with N fertilization.

# 4.5. Conclusion

Our results indicated that biochar and/or compost in a range of combinations added as soil amendments with standard practice fertilizer can improve soil health and boost productivity of peanut with the additional environmental benefits of global warming mitigation. This approach can, therefore, contribute positively to agricultural and environmental sustainability. Biochar and biochar-compost applications positively impacted soil fertility, for example, through their effect on SOC, CEC, and plant available nutrients. Significant increases in peanut yield and plant available soil nutrients were observed due to biochar and compost addition, in comparison to the fertilizer only

treatment, indicating that application of organic amendments does provide agronomic benefits. The response of peanut to biochar and compost could be due to their effects on plant available nutrients, biological N fixation, soil water, and nutrient retention, although other mechanisms cannot be discounted. There was no additional benefit of co-composting biochar with compost, compared to simply adding them on the field together, although there might be benefits in terms of reducing the time of the composting process. Further research is required to verify the residual effects of biochar and biochar-compost soil amendments on sustainable crop yield, C sequestration, and soil quality on Ferralsols. Moreover, the amount of conventional fertilizer that could be reduced, and the resultant economic benefit because of biochar and compost addition needs to be determined, for longer-term economic and environmental sustainability.

# Acknowledgements

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# **Overview of Chapter 5**

This chapter is based on a paper published in the *Soil and Tillage Research* with minimal formatting changes. I have investigated the interaction effects of organic amendments (compost, biochar, compost, biochar-compost mix and co-composted biochar-compost), and five levels of nitrogen fertilizer on plant growth, yield, nutrient uptake and soil physicochemical properties, in an important barley-growing climate and soil type. The trial sites were located in the central highlands of Ethiopia at two sites (one at Holetta Research Center and the other on a farmer's field) on Nitisols.

The study was a factorial experiment in a split plot design, with organic amendments as main plots and five N fertilizer rates as sub-plots. The feedstock for biochar production was acacia (*Acacia spp.*) derived from stems, bark and branches. The pyrolysis was undertaken in earth kilns. I collected plant and soil data at each growth stage of plants. The trials were visited by researchers and agricultural professionals at different growth stages of plants. I organized, analyzed and interpreted the data, and wrote the manuscript for publication. This study was comprehensive, i.e. involving two factors (organic amendments and nitrogen fertilizer), and well-designed experiment.<sup>5</sup>

<sup>&</sup>lt;sup>5</sup> Agegnehu, G., Nelson, P. N., and Bird, M. I. (2016). Crop yield, plant nutrient uptake and soil physicochemical properties under organic soil amendments and nitrogen fertilization on Nitisols. *Soil and Tillage Research* **160**, 1-13.

# Chapter 5

# **Crop Yield, Plant Nutrient Uptake and Soil Physicochemical Properties under Organic Soil Amendments and Nitrogen Fertilization on Nitisols**

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## Abstract

Sustaining soil fertility and enhancing food production on smallholder farms is a great challenge in sub-Saharan Africa. The effects of organic amendments and nitrogen fertilizer on soil physicochemical properties and barley yield were investigated at two sites (Holetta and Robgebeya) both on Nitisols in the central highlands of Ethiopia in the 2014 cropping season. The treatments were factorial combinations of no organic amendment (control), 10 t ha<sup>-1</sup> biochar only (B), 10 t ha<sup>-1</sup> compost only (Com), 10 t Com ha<sup>-1</sup> + 2 t B ha<sup>-1</sup> and 10 t ha<sup>-1</sup> co-composted biochar-compost (COMBI) as main plots, and five N fertilizer levels (0, 23, 46, 69 and 92 kg  $ha^{-1}$ ) as sub-plots, with three replicates. Application of organic amendments and N fertilizer all significantly improved soil fertility and barley yield. The highest yield, chlorophyll content, number of productive tillers and nutrient uptake were obtained from the Com + B soil amendment at Holetta, and from Com at Robgebeya. Mean grain yield responses of barley to the organic amendments were 30 - 49% at Holetta, and 51 -78% at Robgebeya, compared to the control. Fertilizer N significantly increased grain yield, chlorophyll content and N uptake at both locations. The highest grain yield obtained was at 69 kg N ha<sup>-1</sup> at Holetta and at 92 kg ha<sup>-1</sup> at Robgebeya. The organic amendment by N fertilizer interaction significantly influenced grain yield at both sites. Com + B and 69 kg N ha<sup>-1</sup> addition resulted in the highest grain yield (5,381 kg ha<sup>-1</sup>) at Holetta, whereas Com and 92 kg N ha<sup>-1</sup> resulted in the highest grain yield (4,598 kg ha<sup>-1</sup>) at Robgebeya. Organic amendments significantly improved soil properties through increases in soil water content, soil organic carbon (SOC), cation exchange capacity (CEC), and pH (0-20 cm depth). Addition of B, Com, and B + Com increased SOC and CEC by 23-27% and 20-24% at Holetta and 26 - 34% and 19 - 23% at Robgebeya, compared to their respective initial values. Soil pH increased from the initial value of 5.0 to 5.6 at Holetta and from 4.8 to 5.4 at Robgebeya at harvest, due to biochar soil amendment. Grain yield was significantly correlated with total biomass, number of productive tillers, SOC, and CEC. We conclude that application of organic amendments optimizes soil physicochemical properties and will help sustain barley yields in the Ethiopian highlands. The use of B, Com, or Com + B may substantially reduce the amount of mineral fertilizer required for the sustainable production of barley in the long term.

**Keywords:** Biochar, compost co-composted biochar-compost, Nitisol, nitrogen, soil physicochemical properties

# 5.1. Introduction

Poor soil fertility is a major constraint to agricultural productivity in the highlands of Ethiopia, where population and livestock pressure is high (Agegnehu et al., 2014a; Zelleke et al., 2010). Chemical

fertilizer application has been limited to date, and improvement of agricultural productivity necessitates more than the application of chemical fertilizers alone. Core constraints include: topsoil erosion (rates estimated at 10-13 mm per annum on average); soil acidity covering ~40% of the country; soil salinity; a significant depletion of soil organic matter due to extensive use of biomass and manure as animal feed and fuel; depletion of macro and micro-nutrients; and declining soil physical properties (Agegnehu et al., 2014b; Regassa and Agegnehu, 2011; Zelleke et al., 2010). The problem is further exacerbated by widespread deforestation and a lack of land management strategies appropriate to specific soils, landscape and climate (Shiferaw and Holden, 2000; Zelleke et al., 2010).

Barley (Hordeum vulgare L.) is one of the main cereals cultivated in the highlands of Ethiopia, and yields are now seriously affected by low soil fertility. Barley is the fourth most important crop, after maize, rice and wheat, in terms of total world production, and it is the major grain used for malting and brewing (FAO, 2013). It is also an important food grain and malting crop for subsistence farmers in the highlands of Ethiopia (Agegnehu et al., 2014a). It is predominantly grown from 2,000 to 3,500 m above sea level in Ethiopia (Mulatu and Lakew, 2011), and is nationally the fifth most important crop after tef (*Eragrostis tef*), maize, wheat and sorghum, covering an area of  $\sim 1.018$  million ha, but the national average yield is very low at 1.75 t ha<sup>-1</sup> (CSA, 2013). Although there is a considerable potential for increased barley production, numerous factors limit yields (Mulatu and Lakew, 2011). The most important abiotic stresses include low soil fertility, low soil pH, poor soil drainage, drought, and poor agronomic practices. Fertilizer use for barley production is the lowest among all the cereals, which is only 48.3% of the total area of land covered by barley, compared to tef, wheat and maize receiving fertilizer on 59.7%, 69.1% and 56.3%, respectively (Mulatu and Lakew, 2011). Low barley yields can be attributed mainly to low soil pH (less than 5.5) and deficiency of nutrients, especially N and P, due to continuous cropping of cereals (Agegnehu et al., 2014a) and low levels of fertilizer application (Agegnehu et al., 2011). Plants grown on acidic soils may be limited by: deficiencies of N, P, K, Ca, Mg, or Mo; toxicity of Al or Mn; reduced nutrient cycling; reduced uptake of nutrients by plant roots; and inhibition of root growth (Marschner, 2011). Soil acidity adversely affects morphological, physiological and biochemical processes in plants and, thus, N uptake and use efficiency (Fageria and Baligar, 2005; Marschner, 2011).

The supply of N is one of the main factors influencing barley production. The rate of N fertilizer application depends on the purpose for which barley is grown. When barley is grown for feed or food, it is best to apply fertilizer at a higher rate than when it is grown for malting because grain protein content is not as critical in food barley as it is in malting barley (Agegnehu et al., 2014a; Jankovic and Ikanovic, 2011). In Ethiopia, where pH, SOC and N content of most soils are low, the N fertilizer

rates applied for barley production range from 23-46 kg N ha<sup>-1</sup>. Soils with low SOC contents have low crop yields and low use efficiency of added nutrients. Soil organic matter content and bulk density can be improved upon with the addition of organic wastes. Soil organic matter is vital for sustainable yields as it is able to retain water and nutrients, provide a habitat and energy for soil biota, and improve soil structure (Lal, 2011a; Lorenz et al., 2007). Land use change and farming practices have already led to a marked reduction in SOC, and with the increased temperatures expected, SOC content is likely to fall further (Raich et al., 2002). Loss of SOC reduces soil fertility and, hence, further exacerbates climate change.

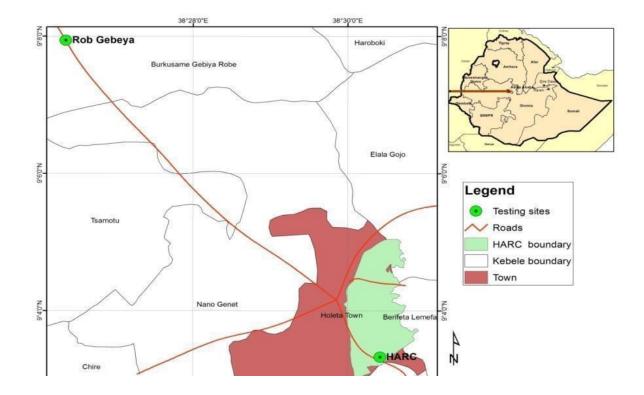
Biochar and compost have been proposed as soil amendments to increase SOC levels and soil fertility. Application of compost can increase soil pH, SOC and N contents, available P, K, Ca, Mg, Na and S, and decreased bulk density (Agegnehu et al., 2016a; Courtney and Mullen, 2008; Fischer and Glaser, 2012). Compost addition to soil has also been shown to increase the yield of barley similarly to, or beyond, the effects of mineral fertilizer application (Kimpinski et al., 2003). However, annual application of high doses of compost had an inhibitory effect on enzyme activity and barley yields (Marcote et al., 2001).

Biochar has two key properties: a high affinity for nutrients and water, reducing nutrient losses; and offsite pollution from nutrient leaching. Unlike compost or manure, biochar has a half-life in soil of up to several centuries (Glaser et al., 2002; Lehmann, 2007). The application of biochar alone to barley has previously not shown any significant effect on yield, but had a significant interaction effect when applied with N fertilizer (Gathorne-Hardy et al., 2009). Other studies have indicated that the combined application of biochar with compost could lead to enhanced soil fertility, improved plant growth, and C sequestration potential (Schulz et al., 2013; Schulz and Glaser, 2012). The combined application of compost and biochar on Dystric Cambisol using maize crop had a synergistic positive effect on SOC content, nutrient content, soil bulk density, and water holding capacity of the soil under field conditions in Germany (Liu et al., 2012). Castellini et al. (2015) reported that small decreases in bulk density (0.014 g cm<sup>-3</sup>) may result in substantial modifications in soil water retention, close to water saturation. Previous studies suggest that an integrated soil fertility management approach may have more sustainable agronomic and economic impact than a focus on chemical fertilizer alone (Agegnehu et al., 2014b; Vanlauwe et al., 2010). Generally, there is no adequate information on the interactive effects of biochar, biochar-compost, and N fertilizer on soil fertility and crop performance in agricultural soils. Therefore, the objectives of this study were to determine the effects of organic amendments, N fertilizer and their interactions on: 1) growth, nutrient uptake and yield of barley; and 2) soil physicochemical properties under rain-fed conditions in the Ethiopian highlands.

#### 5.2. Materials and methods

#### 5.2.1. Characteristics of experimental sites

The experiment was conducted in the main cropping season of 2014 at Holetta Agricultural Research Centre (9°3' 19.43" N, 38° 30' 25.43" E, 30 km west of Addis Ababa at 2,400 m above sea level; Figure 1), and on a farmer's field at Robgebeya in the central highlands of Ethiopia (9°7' 56.55" N, 38°26' 21.41" E, about 47 km west of Addis Ababa, at 2,530 m above sea level; Figure 5.1). The long-term average annual rainfall at Holetta is 1,100 mm, of which about 85% falls from June to September with the remainder from January to May. Average minimum and maximum air temperatures are 6.2 and 22.1°C, respectively (Figure 5.2). The soil type is acidic Eutric Nitisol at both sites. Nitisols are well drained soils with a clayey sub-surface horizon that is deeply stretched and has typical nutty, or polyhedric blocky peds, with shiny faces (IUSS Working Group WRB, 2014). The soil at Holetta had higher SOC, total N, available P, exchangeable K and CEC than that at Robgebeya (Table 5.1). Micronutrients, including Cu, Zn and B, appear to be deficient in most Nitisols of the Ethiopian highlands (Tulema et al., 2007).



**Figure 5.1.** Map of the experimental sites at Holetta Agricultural Research Center (HARC) and Rogebeya in the central highlands of Ethiopia.

Three weeks prior to planting, soil samples were collected from 0 - 20 cm depth at each experimental site. Six sampling locations were selected by dividing the trial area into six cells ( $17 \text{ m} \times 10 \text{ m}$ ). Three sampling points were randomly selected within each of the six cells. At each sampling point, surface organic litter was cleared away, and four samples from a 3-m radius were collected using a manual auger with 20 cm core barrel of 6 cm internal diameter. Roots were removed by hand. The twelve samples were combined, making a bulk sample for each cell, resulting in six composite samples per site for analysis. The samples were weighed, oven-dried at 70°C and then reweighed to determine soil moisture content. These weights were used to calculate soil bulk density, based on auger dimensions and core length. After manual homogenization, the samples were ground to pass a 2-mm sieve. Soil samples were analyzed for: pH using a ratio of 2.5 ml water to 1 g soil (McLean, 1982); available P using the Bray-II method (Bray and Kurtz, 1945); SOC using the Walkley and Black (1934) method; total N content by the Kjeldahl digestion (Nelson and Sommers, 1982); exchangeable cations; and CEC using ammonium acetate method (Black, 1965) at the soil and plant analysis laboratory of Holetta Agricultural Research Center, Ethiopia. Table 5.1 shows the pre-planting physicochemical characteristics of the trial soil.

# 5.2.2. Experimental set-up and procedure

Biochar was produced at Ginchi, roughly 70 km west of Addis Ababa, while compost and cocomposted biochar-compost were produced at Holetta Research center. The feedstock for biochar production was acacia (*Acacia spp.*) derived from stems, bark and branches. The pyrolysis was undertaken in earth kilns. The earth formed the necessary gas-tight insulating barrier behind which carbonization could take place, which would allow the biomass to burn away. The kiln was fired and the biomass heated up to allow pyrolysis for six days. The kiln was mostly sealed, except for a few air pockets initially left open for steam and smoke to escape. After cooling, the kilns were opened and the biochar was removed. The biochar was crushed to particle size below 25 mm prior to field application. The biochar mass was approximately 15 - 20% of the original biomass, which is in accordance with Adam (2009). The efficiency of traditional charcoal production methods is about 10% - 22% (calculated on using oven-dry wood with 0% water content), while the efficiency of improved charcoal production system (ICPS) is 30% - 42%. The ICPS reduces emissions to the atmosphere by up to 75%, compared with traditional carbonization processes (Adam, 2009). Compost was prepared following standard procedures (Tulema et al., 2007). A homogenized mixture of farmyard manure (FYM), feed leftovers from dairy cattle and bedding materials were composted in pits (1.5 m  $\times$  2.4 m wide by 1.0 m deep). The source of the FYM was Holetta Agricultural Research Centre (HARC), where the National Livestock Research Program is coordinated. About 75% of the FYM is manure and the rest is plant materials, such as hay and crop residues. Fresh and decomposing materials were placed in alternating layers, with fresh and straw materials constituting the bottom layer. The compost was turned every two weeks and was mature after 15 weeks. The final compost was attributed by a lower C:N ratio of about 20, and a higher pH value, compared to the starting feed mixture (Fischer and Glaser, 2012). The co-composted biochar-compost was prepared following the same procedure, using a ratio of 1:5 biochar: compost on a dry weight basis. Biochar, mature compost and co-composted biochar-compost samples were collected from all pits for analysis of chemical properties before they were applied in the field (Table 5.1). The samples were analyzed following the methods for soil analysis.

The experimental sites were prepared for sowing using standard cultivation practices. Tractormounted disk plowing and disk harrowing were carried out in May and June at the research station. However, the on-farm trial field was plowed using oxen-drawn implements at the depth of about 10 cm for first plowing and 15 cm for the last pass. The experiment was a factorial split-plot design with five organic amendments as main plots and five N fertilizer levels (0, 23, 46, 69 and 92 kg N ha<sup>-1</sup>) as sub-plots, with three replicates for each treatment. Treatment sequencing was randomized and the main and sub-plot areas were 62.5 m<sup>2</sup> (12.5 m × 5 m each) and 10 m<sup>2</sup> (5 m × 2 m each) per replicate, respectively. The total area of each trail site was ~1300 m<sup>2</sup>. The organic amendments were: 1) control without amendment (Con); 2) 10 t ha<sup>-1</sup> acacia biochar (B); 3) 10 t ha<sup>-1</sup> compost (Com); 4) 10 t ha<sup>-1</sup> Com + 2 t ha<sup>-1</sup> B mixed onsite before application; and 5) co-composted biochar-compost (COMBI) applied at 10 t ha<sup>-1</sup>. Applications of B and Com were made on air dry weight basis.

| Parameter                             | Unit                     | Com   | В     | COMBI | Soil J  | properties |
|---------------------------------------|--------------------------|-------|-------|-------|---------|------------|
|                                       |                          |       |       |       | Holetta | Robgebeya  |
| Physical properties                   |                          |       |       |       |         |            |
| Soil bulk density                     | g cm-3                   |       |       |       | 1.02    | 0.98       |
| Soil water content                    | %                        |       |       |       | 16.5    | 15.4       |
| Clay                                  | %                        |       |       |       | 56.1    | 53.6       |
| Silt                                  | %                        |       |       |       | 28.6    | 31.5       |
| Sand                                  | %                        |       |       |       | 15.4    | 14.9       |
| <b>Chemical properties</b>            |                          |       |       |       |         |            |
| pH (H <sub>2</sub> O)                 |                          | 7.54  | 8.27  | 7.87  | 4.97    | 4.83       |
| Organic carbon (OC)                   | %                        | 27.8  | 51.6  | 32.7  | 2.32    | 1.44       |
| Total nitrogen (N)                    | %                        | 1.36  | 0.56  | 1.25  | 0.23    | 0.15       |
| Bray-2 phosphorus (P)                 | g kg <sup>-1</sup>       | 9.3   | 0.90  | 8.3   | 0.015   | 0.007      |
| Exchangeable potassium (K)            | cmol(+) kg <sup>-1</sup> | 3.62  | 7.17  | 4.86  | 0.98    | 0.71       |
| Exchangeable sodium (Na)              | cmol(+) kg <sup>-1</sup> | 1.15  | 0.59  | 0.78  | 0.41    | 0.37       |
| Exchangeable calcium (Ca)             | cmol(+) kg <sup>-1</sup> | 5.83  | 3.97  | 4.41  | 12.2    | 11.6       |
| Exchangeable magnesium (Mg)           | cmol(+) kg <sup>-1</sup> | 2.65  | 2.42  | 2.28  | 2.67    | 2.85       |
| Cation exchange capacity (CEC)        | cmol(+) kg <sup>-1</sup> | 13.3  | 14.2  | 12.3  | 16.2    | 15.5       |
| Ammonium nitrogen (NH4-N)             | mg kg <sup>-1</sup>      | 147   | trace | 63    | 21.0    | 16.3       |
| Nitrate nitrogen (NO <sub>3</sub> -N) | mg kg <sup>-1</sup>      | trace | 77.0  | trace | 100.3   | 88.7       |

**Table 5.1.** Physicochemical properties of compost (Com), acacia biochar (B), co-composted biocharcompost (COMBI) and pre-planting soil (0 - 20 cm depth) at the two sites.

The organic amendments were applied manually and evenly to main plots, 14 days before sowing, and thoroughly mixed in the upper 5 cm of soil. At the time of planting, the experiential plots were finely delineated manually using rakes and fork diggers and the planting rows were made using iron row markers adjusted in 0.2 m row spacing. The experimental plots at both locations were sown with six-row food barley (variety *HB-1307*) at the rate of 90 kg ha<sup>-1</sup> in 10 rows 5 m long with 0.2 m row spacing. A space of 0.5 m was left between plots and 1.0 m between blocks, to avoid border effects of treatments. Sowing took place at the onset of rainfall at each site, this being on the 24<sup>th</sup> of June 2014 at Holetta and the 30<sup>th</sup> of June 2014 at Robgebeya. The preceding crop was potato at both sites.

Triple superphosphate (TSP) was applied to all plots at planting at the recommended rate (20 kg P ha ha<sup>-1</sup>) in a band in the row. The recommended N fertilizer application rate for food barley production

is 46 kg N ha<sup>-1</sup>. To minimize losses and increase efficiency, all the levels of N were applied in the row as urea in two applications; half at planting and the other half 42 days after planting, during the maximum growth period of the crop at full tillering stage, after the first weeding and during light rainfall to minimize loss of N to the atmosphere. Other relevant field trial management practices were applied with close supervision during the crop growth period. The total rainfall during the crop growing period was 714 mm, while the mean maximum and minimum temperatures were 21.3 and 6.8 °C, respectively. Despite minor incidence of shoot fly at the initial growth stage, insecticides were not applied.

### 5.2.3. Sampling and measurements

After sowing, plant and soil parameters were measured periodically. Soil water content (SWC) was measured at a depth of 12 cm on days 12, 26, 41, 62, 72, 85, 107 and 124 after sowing, between 6<sup>th</sup> July 2014 and 1<sup>st</sup> December 2014, using a soil moisture and pH tester (Takemura Electric Works Ltd., Japan) and gravimetric method. The equipment measures soil moisture content when the probe is fully inserted into the soil. Leaf chlorophyll was measured on days 36, 54, 68, 84 and 102 after sowing, between 30<sup>th</sup> July and 15<sup>th</sup> October 2014, using a chlorophyll meter (SPAD 502, Konica Minolta, Tokyo, Japan). For each chlorophyll measurement, duplicate readings were made on the second fully expanded leaf from the top of the main plant stem, approximately half way along the leaf, taking care to avoid veins and mid-rib. This procedure was repeated for ten randomly selected plants in each plot. Ten plants were sampled at boot stage to measure leaf area using CI-202 Leaf Area Meter (CID, INC, USA).

Harvesting took place on the 4<sup>th</sup> November 2014 at Holetta and on the 1<sup>st</sup> December 2014 at Robgebeya. To measure total above-ground biomass and grain yields, the central six rows of each plot (5 m  $\times$  1.2 m) were harvested at soil level. Plant parameters collected were grain yield, above-ground total biomass (separated into barley and weeds), harvest index, thousand grains mass (TGM), number of tillers at full tillering stage, productive (grain-bearing) tillers per square meter at physiological maturity, spike length and plant height. Mature plant height was measured from the ground level to the tip of the spike, excluding the awns, at physiological maturity (average of ten plants). Spike length was measured from the base of the spike to the top of the spike (average of ten plants), excluding the awns. After threshing, seeds were cleaned and weighed. A sample of 250 grains was weighed from each replicate to derive thousand grains mass (TGM in g). Seed moisture content and hectoliter mass was measured using a Grain Analysis Computer (GAC-2100, Germany). Total

biomass (dry matter basis) and grain yields (adjusted to a moisture content of 12.5%), which were recorded on plot basis, were converted to kg ha<sup>-1</sup> for statistical analysis.

The plant shoots sampled for leaf area were also used to determine the nutrient concentration of the plants. Ten plant shoots were randomly selected from each plot at the booting stage, and clipped at their base for the determination of C, N, P and K contents. Plant samples were dried at 70 °C for 72 hours and then ground using a Karl Kolb-RETSCH (Scientific Technical Supplies, Germany) to pass through a 0.5 mm sieve. Total plant C concentration was determined using the Walkley and Black (1934) method, and total N concentration by Kjeldahl digestion (Nelson and Sommers, 1982). Plant K concentration was determined by atomic absorption spectroscopy after wet digestion with sulfuric acid (Watson et al., 1990). Plant P content was determined photometrically in the same digest using the molybdenum blue method (Mills and Jones, 1996). Soil samples from a depth of 0-20 cm were collected after harvesting. For the post-harvest soil samplings, three samples were taken from each plot, and combined into one composite sample per plot for analysis. Soil water content and bulk density were determined using these samples. Soil samples were analyzed for chemical properties following the procedures described above.

### 5.2.4. Data analysis

The data were subjected to analysis of variance using the general linear model procedure (PROC GLM) of SAS statistical package version 9.1 (SAS Institute Cary, NC). The total variability for each trait was quantified using the following model:

$$T_{ijk} = u + R_i + A_j + R(A)_{ij} + N_k + AN_{(jk)} + e_{ijk}$$

Where  $T_{ijk}$  is total observation,  $\mu$  = grand mean,  $R_i$  is effect of the i<sup>th</sup> replication,  $A_j$  is effect of the j<sup>th</sup> organic amendment,  $N_k$  is effect of the k<sup>th</sup> nitrogen level, AN is the interaction, and  $R(A)_{ij}$  and  $e_{ijk}$  are the variations due to random error for the main and sub-plots, respectively. Significance of the A effect was tested against the  $R(A)_{ij}$  mean square as an error term. All other effects were tested against the residual. Means for the main effects of organic amendments and N fertilizer levels (n = 5) were compared using the MEANS statement with the least significant difference (LSD) test at the 5% level. Single degree of freedom orthogonal contrasts was also performed to determine the nature of the crop response to the rates of applied N fertilizer. Means for the interactions were compared using the PDIFF STDERR option in the LSMEANS statement of the GLM procedure of SAS, in particular, specifying the R(A) as an appropriate error term for separating LSMEANS for the interaction of organic amendment and N fertilizer rate. Linear regression analyses were performed to distinguish

between the effects of the main factors (organic amendments-OA and N fertilizer) and their interaction (OA  $\times$  N), following the SAS REG procedure (SAS Institute Inc., 2008).

### 5.3. Results

### 5.3.1. Yield and yield components of barley

Grain yield, total above-ground biomass, mean leaf chlorophyll, plant height, number of productive tillers, leaf area and spike length were all significantly ( $p \le 0.05$  and  $p \le 0.01$ ) increased by organic soil amendments and N fertilizer at both locations (Table 2). Thousand grains mass significantly ( $p \le 0.05$ ) responded to organic amendments only at Holetta and to N fertilizer application only at Robgebeya. Spikelet number per plant differed significantly ( $p \le 0.05$ ) between organic amendments and N fertilizer rates at both locations ( $p \le 0.001$ ). Weed biomass was significantly ( $p \le 0.01$ ) affected by organic amendments and by the N fertilizer rate at both locations. The organic amendment by N fertilizer rate interaction significantly ( $p \le 0.05$ ) improved grain yield, total barley above-ground biomass, productive tillers, and weed biomass at both locations, and thousand grain mass of barley only at Robgebeya ( $p \le 0.05$ ). Leaf area per plant was significantly ( $p \le 0.01$ ) increased by the interaction of organic amendment and N fertilizer rate only at Holetta (Table 5.2).

|                              |     |     | Holett | a    |       |     |     | Robgebe | eya  |       |
|------------------------------|-----|-----|--------|------|-------|-----|-----|---------|------|-------|
| Parameter                    | OA  | N   | OA×N   | CV   | R-MSE | OA  | N   | OA×N    | CV   | R-MSE |
| Grain yield                  | **  | *** | *      | 8.1  | 318   | **  | *** | **      | 10.9 | 338   |
| Biomass yield                | **  | *** | *      | 9.4  | 817   | **  | *** | *       | 10.1 | 787   |
| Harvest index                | NS  | *   | *      | 7.0  | 3.2   | *   | NS  | NS      | 11.2 | 4.5   |
| Plant height                 | *   | *** | NS     | 2.3  | 2.6   | *   | *** | NS      | 5.8  | 6.1   |
| Chlorophyll                  | **  | *** | *      | 4.1  | 1.5   | *   | *** | NS      | 2.2  | 0.90  |
| Leaf area                    | *   | **  | NS     | 12.1 | 10.2  | **  | *** | **      | 13.4 | 6.5   |
| Prod. tillers                | *   | *** | **     | 12.1 | 35.9  | *   | *** | *       | 11.4 | 32.4  |
| Spikelet plant <sup>-1</sup> | *   | *** | NS     | 5.0  | 0.34  | *   | *** | NS      | 4.8  | 0.29  |
| Spike length                 | NS  | *** | NS     | 6.7  | 3.5   | *   | *** | NS      | 6.5  | 3.0   |
| TGM                          | *   | NS  | NS     | 4.2  | 1.8   | NS  | *** | *       | 3.9  | 1.5   |
| WB (g m <sup>-2</sup> )      | **  | *** | **     | 17.4 | 13.4  | **  | **  | **      | 16.6 | 14.1  |
| Shoot C                      | NS  | NS  | NS     | 1.5  | 0.79  | NS  | NS  | NS      | 4.4  | 2.3   |
| Shoot N                      | *   | *** | NS     | 9.7  | 0.19  | *   | *** | NS      | 8.3  | 0.25  |
| Shoot P                      | *   | NS  | NS     | 7.7  | 0.02  | *   | *** | NS      | 5.2  | 0.015 |
| Shoot K                      | *   | **  | NS     | 8.7  | 0.29  | *   | *** | NS      | 7.1  | 0.16  |
| SWC                          | *** | NS  | NS     | 3.7  | 1.7   | *** | *   | NS      | 5.5  | 2.5   |
| SBD                          | *   | NS  | NS     | 3.2  | 0.03  | **  | NS  | NS      | 2.3  | 0.02  |
| Soil pH                      | *   | NS  | NS     | 3.9  | 0.21  | *   | **  | NS      | 1.8  | 0.10  |
| Soil OC                      | *** | NS  | NS     | 3.8  | 0.11  | **  | NS  | *       | 6.1  | 0.10  |
| Soil N                       | *** | NS  | NS     | 3.0  | 0.01  | **  | NS  | NS      | 4.8  | 0.007 |
| C: N ratio                   | NS  | NS  | NS     | 4.7  | 0.50  | *   | NS  | NS      | 7.7  | 0.89  |
| Bray 2 soil P                | *   | NS  | NS     | 7.5  | 1.3   | *   | NS  | NS      | 27.3 | 2.5   |
| Exch. Soil K                 | *   | NS  | NS     | 9.1  | 0.08  | *   | NS  | NS      | 6.9  | 0.05  |
| Exch. Soil Na                | NS  | NS  | NS     | 4.9  | 0.02  | *** | NS  | NS      | 6.2  | 0.02  |
| Exch. Soil Ca                | *** | NS  | NS     | 4.6  | 0.40  | *   | NS  | NS      | 5.4  | 0.40  |
| Exch. Soil Mg                | **  | *   | NS     | 4.7  | 0.13  | *   | *   | NS      | 6.6  | 0.18  |
| CEC                          | *** | *   | NS     | 4.1  | 0.77  | **  | *   | NS      | 4.8  | 0.87  |

**Table 5.2.** Significance of the effects of organic amendments (OA), N fertilizer rates (N) and their interaction (OA  $\times$  N on plant and soil parameters at the two sites.

Significant at  ${}^{*}p \le 0.05$ ,  ${}^{**}p \le 0.01$ ,  ${}^{***}p \le 0.001$ ; NS: Not significant. WB: weed biomass (oven-dried); TGM: Thousand grain mass; SWC: Soil water content (%); SBD: Soil bulk density (g cm<sup>-3</sup>); Prod.: Productive; CV: Coefficient of variation; R-MSE: Root mean square error.

Yields of barley were greater with all organic soil amendments than in the control at both locations. Barley yield was highest on Com + B amended soil at Holetta (4,425 kg ha<sup>-1</sup>) and Com amended soil at Robgebeya (3,630 kg ha<sup>-1</sup>), but differences in yields between the different organic amendments were not statistically significant (Tables 5.3 and 5.4). Above-ground biomass, leaf chlorophyll, productive tillers, spike length and plant heights were significantly higher in organic-amended plots than non-amended plots at both locations (Tables 5.3 and 5.4). The tallest plant height and largest spike size of barley were recorded from Com and Com + B, but differences among organic amendments were not statistically significant. Grain yields were higher by 30, 41, 49 and 42% at Holetta and 67, 78, 70 and 51% at Robgebeya, respectively for B, Com, Com + B and COMBI soil amendments, compared to the control (Tables 5.3 and 5.4). Biochar, Com, Com + B and COMBI amended soil increased the number of productive tillers by 24, 27, 30 and 25% at Holetta and 26, 38, 21 and 19% at Robgebeya, compared to the control. The lowest weed biomass was recorded from biochar amended plots at both locations. Overall, the application of organic amendments increased plant growth, yield, and yield components of barley, with grain yield (as a ratio of control value) decreasing in the order Com + B (1.5) > Com, and COMBI (1.4) > B (1.3) > Control (1.0) at Holetta, and Com (1.8) > Com + B and B (1.7) > COMBI (1.5) > Control (1.0) at Robgebeya. The effects of organic amendments (Com + B versus control at Holetta) on grain yields were significantly greater than the effects of N fertilizer (92 kg N ha<sup>-1</sup> versus 0 kg ha<sup>-1</sup>, Tables 5.3 and 5.4).

Barley grain yield, total biomass, leaf chlorophyll, spike length, spikelet number per plant, number of productive tillers, and plant height significantly and consistently increased with N fertilizer rate ( $p \le 0.001$ ). Although differences in yields among the highest three N rates were not significant, the maximum grain yield of barley was recorded from 69 kg N ha<sup>-1</sup> at Holetta and 92 kg ha<sup>-1</sup> at Robgebeya (Tables 5.3 and 5.4). The application of N fertilizer at the rates of 23, 46, 69 and 92 kg N ha<sup>-1</sup> resulted in linear and quadratic responses, with mean grain yield advantages of 14, 23, 34 and 33%, respectively, relative to the control (no N fertilizer) at Holetta and 43, 72, 79 and 83% over the control at Robgebeya (Tables 5.3 and 5.4). Leaf chlorophyll and number of productive tillers were increased by 10% and 44% at Holetta, and 12% and 35% at Robgebeya, respectively, in response to the highest two rates of applied fertilizer N relative to the control. Thousand grain mass exhibited significant linear (p < 0.001) and quadratic (p < 0.05) response to N fertilizer rate at Robgebeya, but not at Holetta. Plant height and leaf area per plant increased consistently with N fertilizer rate at both locations, but differences between the highest two N levels were not significant for either parameter (Tables 5.3 and 5.4).

|                          |                | ÷                      |      |      |      |             | ·                  |                       |      |                     |                      |
|--------------------------|----------------|------------------------|------|------|------|-------------|--------------------|-----------------------|------|---------------------|----------------------|
|                          | Grain yield    | Total biomass          | HI   | TGM  | PHT  | CHLC        | PTIL               | LA (cm <sup>2</sup>   | SPL  | SPKL                | WB                   |
| Treatments               | $(kg ha^{-1})$ | (kg ha <sup>-1</sup> ) | (%)  | (g)  | (cm) | (SPAD-unit) | (m <sup>-2</sup> ) | plant <sup>-1</sup> ) | (cm) | plant <sup>-1</sup> | (g m <sup>-2</sup> ) |
| OA<br>O to 1             | 2001           | (275                   | 47.2 | 47.2 | 100  | 20.7        | 250                | 72.7                  | ( (  | 50.7                | 100.1                |
| Control                  | 2981           | 6375                   | 47.3 | 47.3 | 108  | 38.7        | 250                | 72.7                  | 6.6  | 50.7                | 109.1                |
| В                        | 3867           | 8868                   | 44.4 | 43.9 | 110  | 41.1        | 310                | 78.3                  | 6.9  | 51.8                | 54.7                 |
| Com                      | 4213           | 9091                   | 46.6 | 46.1 | 112  | 41.2        | 318                | 88.1                  | 7.1  | 52.8                | 77.3                 |
| Com + B                  | 4425           | 9727                   | 46.3 | 46.1 | 113  | 41.3        | 324                | 90.4                  | 7.0  | 55.2                | 71.1                 |
| COMBI                    | 4234           | 9550                   | 44.2 | 44.2 | 111  | 41.0        | 312                | 90.8                  | 6.9  | 53.4                | 72.4                 |
| LSD (0.05)               | 1013           | 2089                   | 6.1  | 6.0  | 3.9  | 1.2         | 53.2               | 6.1                   | 0.40 | 4.9                 | 13.6                 |
| OA vs. Con               | ***            | ***                    | NS   | **   | NS   | ***         | ***                | ***                   | *    | *                   | ***                  |
| N (kg ha <sup>-1</sup> ) |                |                        |      |      |      |             |                    |                       |      |                     |                      |
| 0                        | 3269           | 7207                   | 46.7 | 45.9 | 106  | 38.0        | 233                | 70.3                  | 6.4  | 46.6                | 62.1                 |
| 23                       | 3723           | 8215                   | 44.2 | 45.9 | 110  | 40.8        | 295                | 85.2                  | 6.9  | 53.5                | 68.3                 |
| 46                       | 4014           | 9360                   | 44.9 | 43.2 | 112  | 41.1        | 311                | 80.8                  | 7.1  | 54.3                | 88.5                 |
| 69                       | 4376           | 9553                   | 46.8 | 45.5 | 113  | 41.8        | 339                | 89.2                  | 7.2  | 54.8                | 83.1                 |
| 92                       | 4338           | 9277                   | 46.1 | 47.0 | 113  | 41.4        | 319                | 94.7                  | 7.1  | 54.8                | 82.5                 |
| LSD (0.05)               | 235            | 603                    | 3.7  | 2.3  | 1.9  | 0.52        | 26.5               | 7.5                   | 0.25 | 2.6                 | 9.9                  |
| N vs. 0                  | ***            | ***                    | NS   | NS   | ***  | ***         | **                 | ***                   | ***  | ***                 | ***                  |
| N linear                 | ***            | ***                    | NS   | NS   | ***  | ***         | ***                | ***                   | ***  | ***                 | ***                  |
| N quadratic              | *              | **                     | NS   | NS   | **   | ***         | ***                | NS                    | **   | ***                 | NS                   |

Table 5.3. Table of means for main effects of organic amendments and N fertilizer on barley agronomic parameters at Holetta.

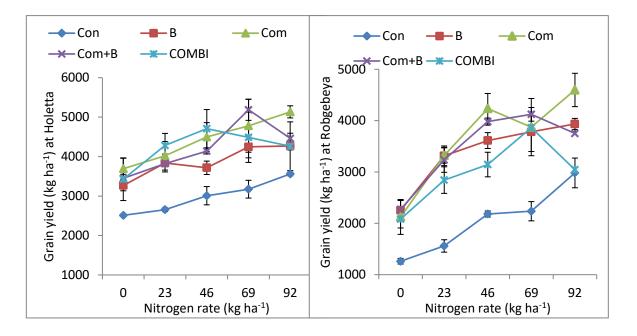
Significant at  ${}^{*}p \le 0.05$ ,  ${}^{**}p \le 0.01$ ,  ${}^{***}p \le 0.001$ ; NS: not significant. B: Biochar; Com: compost; COMBI: Co-composted biochar-compost; HI: Harvest index; TGM: Thousand grain mass; PHT: Plant height; CHLC: Chlorophyll content (SPAD-unit); PTIL: Number of productive tillers; LA: Leaf area; SPL: Spike length; SPKL: Spikelet number; WB: Weed biomass; LSD: Least significant difference at 5% level of significance; N: Nitrogen.

|                          | Grain yield    | Biomass yield  | HI   | TGM  | PHT  | CHLC (SPAD- | PTIL               | LA (cm <sup>2</sup>   | SPL  | SPKL                | WB   |
|--------------------------|----------------|----------------|------|------|------|-------------|--------------------|-----------------------|------|---------------------|------|
| Treatments               | $(kg ha^{-1})$ | $(kg ha^{-1})$ | (%)  | (g)  | (cm) | unit)       | (m <sup>-2</sup> ) | plant <sup>-1</sup> ) | (cm) | Plant <sup>-1</sup> | (g)  |
| OA                       | 2014           | 500.5          | 41.0 | 20.2 | 0.0  | 22.0        |                    | 20.2                  | 5.0  | 10.0                | 110  |
| Control                  | 2044           | 5095           | 41.2 | 39.2 | 92   | 33.9        | 232                | 38.2                  | 5.8  | 40.0                | 119  |
| В                        | 3380           | 8355           | 40.5 | 40.4 | 110  | 38.1        | 293                | 48.9                  | 6.5  | 47.6                | 54   |
| Com                      | 3630           | 8526           | 42.1 | 39.6 | 112  | 37.4        | 321                | 57.9                  | 6.6  | 46.5                | 96   |
| Com + B                  | 3473           | 9059           | 38.7 | 39.6 | 111  | 37.5        | 282                | 49.4                  | 6.6  | 46.5                | 78   |
| COMBI                    | 3096           | 8069           | 38.4 | 39.0 | 109  | 36.3        | 278                | 48.6                  | 6.4  | 45.3                | 80   |
| LSD                      | 637            | 1375           | 2.3  | 2.1  | 17   | 3.7         | 31.0               | 5.9                   | 0.40 | 6.9                 | 25.3 |
| OA vs. Con               | ***            | ***            | NS   | NS   | ***  | ***         | **                 | **                    | **   | **                  | ***  |
| N (kg ha <sup>-1</sup> ) |                |                |      |      |      |             |                    |                       |      |                     |      |
| 0                        | 1999           | 5135           | 39.3 | 37.7 | 94   | 34.2        | 225                | 40.2                  | 5.7  | 40.0                | 79.3 |
| 23                       | 2855           | 7286           | 39.9 | 39.5 | 104  | 36.1        | 291                | 49.1                  | 6.3  | 44.7                | 81.9 |
| 46                       | 3431           | 8660           | 40.1 | 40.1 | 108  | 37.2        | 302                | 49.7                  | 6.5  | 45.3                | 84.1 |
| 69                       | 3575           | 8806           | 40.8 | 40.3 | 110  | 37.9        | 304                | 51.9                  | 6.6  | 47.1                | 88.2 |
| 92                       | 3662           | 9217           | 39.7 | 40.2 | 113  | 38.1        | 302                | 52.2                  | 6.8  | 48.9                | 92.7 |
| LSD (0.05)               | 249            | 574            | 2.0  | 1.14 | 4.5  | 1.1         | 23.9               | 4.8                   | 0.18 | 2.2                 | 10.4 |
| 0 vs. N                  | ***            | ***            | NS   | ***  | ***  | ***         | ***                | **                    | ***  | ***                 | *    |
| N linear                 | ***            | ***            | NS   | ***  | ***  | ***         | ***                | **                    | ***  | ***                 | **   |
| N quadratic              | ***            | ***            | NS   | *    | *    | NS          | **                 | NS                    | *    | NS                  | NS   |

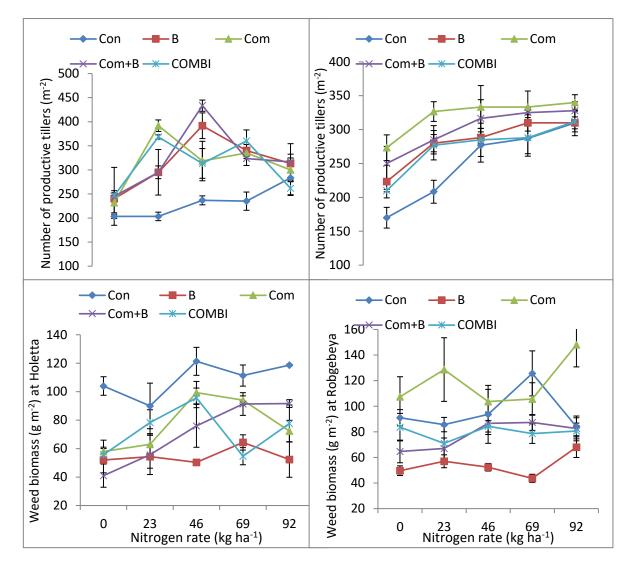
Table 5.4. Table of means for main effects of organic amendments and N fertilizer on barley agronomic parameters at Robgebeya.

Significant at  ${}^{*}p \le 0.05$ ,  ${}^{**}p \le 0.01$ ,  ${}^{***}p \le 0.001$ ; NS: Not significant. B: biochar; Com: Compost; COMBI: Co-composted biochar-compost; HI: Harvest index; TGM: Thousand grain mass; PHT: Plant height; CHLC: Chlorophyll content (SPAD-unit); PTIL: Number of productive tillers (m<sup>-2</sup>); LA: Leaf area; SPL: Spike length; SPKL: Spikelet number; WB: Oven-dried weed biomass; LSD: Least significant difference at 5% level of significance; N: Nitrogen.

Organic amendments and N fertilizer essentially had additive effects on yield and some yield components of barley, and weed biomass. The highest grain yield of 5,381 kg ha<sup>-1</sup> was obtained at Holetta from the applications of Com + B and 69 kg N ha<sup>-1</sup>, followed by Com and 92 kg N ha<sup>-1</sup> with a yield of 5,134 kg ha<sup>-1</sup> (Figure 5.2). In contrast, the highest grain yield at Robgebeya (4,598 kg ha<sup>-1</sup>) was recorded from the addition of Com and 92 kg N ha<sup>-1</sup>, followed by Com with an N fertilizer rate of 46 kg ha<sup>-1</sup> and a yield of 4,235 kg ha<sup>-1</sup> (Figure 5.2). Grain yield responded synergistically to the combined application of organic amendments and N fertilizer. For example, at Robgebeya, the application of Com and 23 kg N ha<sup>-1</sup> more than doubled grain yield (3,321 kg ha<sup>-1</sup>) compared to the yield (1,560 kg ha<sup>-1</sup>) with the same N fertilizer rate only (Figure 5.3). However, the trend in yield increase with N levels was not consistent for all organic amendments at both locations. The number of productive tillers increased consistently with the increase in the N fertilizer levels for all organic amendments at Robgebeya, but the trend at Holetta was not consistent. The highest number of productive tillers (434 m<sup>-2</sup>) and (340 m<sup>-2</sup>) were obtained from Com + B and 69 kg N ha<sup>-1</sup> and Com and 92 kg N ha<sup>-1</sup> at Holetta and Robgebeya, respectively. The application of Com and 92 kg N ha<sup>-1</sup> resulted in the highest weed biomass at Robgebeya (148 g m<sup>-2</sup>) and 46 kg N ha<sup>-1</sup> only at Holetta (121 g m<sup>-2</sup>). In contrast, Com + B only and B + 69 kg N ha<sup>-1</sup> resulted in the lowest weed biomass at Holetta (41 g m<sup>-2</sup>) and Robgebeya 44 g m<sup>-2</sup>), respectively (Figure 5.4).

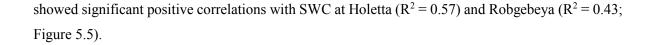


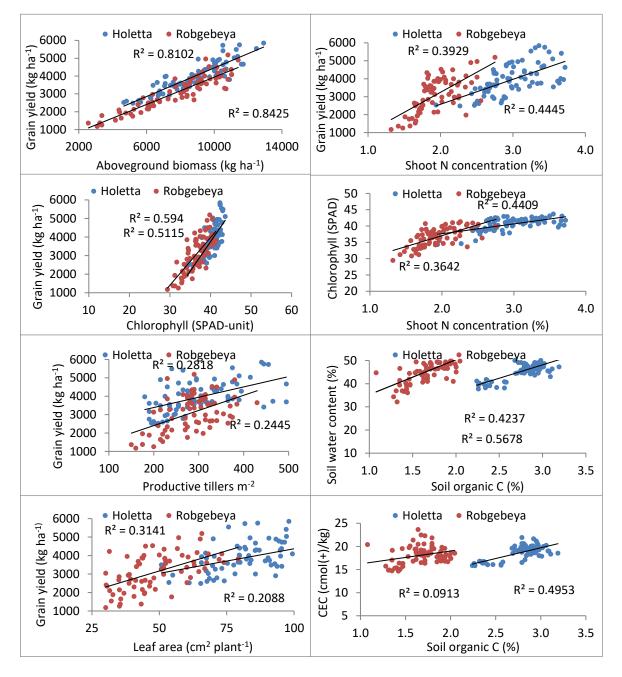
**Figure 5.2.** Barley grain yield as influenced by the interaction of organic amendment and N fertilizer rate at Holetta (left) and Robgebeya (right). Note: Con: Control; B: Biochar; Com: Compost; COMBI: Co-composted biochar-compost. Error bars represent  $\pm 1$  SE.



**Figure 5.3.** Number of productive tillers (upper) and weed biomass (lower) as influenced by the interaction of organic amendments and N fertilizer rate at Holetta (left) and Robgebeya (right). Note: Con: Control; B: Biochar; Com: Compost; COMBI: Co-composted biochar-compost. Error bars represent  $\pm 1$  SE.

The linear regression analysis indicated that grain yield was significantly (p < 0.001) and positively correlated with total biomass, leaf chlorophyll and shoot N concentration at Holetta ( $R^2 = 0.76$ , 0.49 and 0.32, respectively) and Robgebeya ( $R^2 = 0.81$ , 0.57 and 0.41, respectively), corresponding to the application of organic amendments and N fertilizer levels (Figure 5.4). Grain yield was significantly correlated with the number of productive tillers at Holetta ( $R^2 = 0.31$ ) and Robgebeya ( $R^2 = 0.42$ ). Leaf chlorophyll was significantly correlated with shoot N concentration at Holetta ( $R^2 = 0.36$ ) and Robgebeya ( $R^2 = 0.44$ ). The improvement in SOC, due to the addition of organic amendments,





**Figure 5.4.** Relationships among plant parameters, shoot N concentration and soil physicochemical properties (n = 75) tested at factorial combinations of five different organic amendments and five N levels at Holetta and Robgebeya. Note: Significant at  $p \le 0.05$  (R<sup>2</sup> = 0.16-0.34),  $p \le 0.01$  (R<sup>2</sup> = 0.35-0.50),  $p \le 0.001$  (R<sup>2</sup> > 0.50); NS: Not significant.

### 5.3.2. Nutrient uptake

Organic soil amendments significantly ( $p \le 0.05$ ) improved N, P and K concentrations by barley plants (with no significant difference among them), compared to the control (Table 5.5). Shoot N, P and K concentration ranged from 2.65-3.19%, 0.28-0.34% and 3.07-3.63, respectively at Holetta, and 1.75-2.12%, 0.27-0.31% and 2.09-2.39%, respectively at Robgebeya (Table 5.5). At Holetta, Com addition resulted in the highest shoot N, P and K contents of 3.19, 0.34 and 3.63%, respectively, while at Robgebeya Com + B recorded the highest N, P and K concentrations of 2.12, 0.31 and 2.39%, respectively (Table 5). Biochar, Com, Com + B, and COMBI addition increased N concentration by a factor of 1.1, 1.2, 1.2 and 1.1 relative to the control at both locations. Plant C concentration did not significantly respond to organic soil amendments (data not shown).

Nitrogen fertilizer rates resulted in significant (p < 0.01) linear and quadratic responses of shoot N and K contents, which were 2.66-3.22% and 3.11-3.57%, respectively at Holetta and 1.68-2.12% and 2.13-2.43% at Robgebeya (Table 5.5). Shoot P concentration exhibited significant linear and quadratic responses to N fertilizer rate at Robgebeya and a linear response at Holetta (Table 5.5). The maximum shoot N content was noted from the application of 92 kg N ha<sup>-1</sup> at both locations. In general, organic amendments and application of N fertilizer proportionally increased grain yield more than they did shoot nutrient contents. Plant nutrient uptake was not significantly affected by the interaction of organic amendment and N fertilizer rate (Table 5.2). The moisture contents of barley grain were 10-11% (data not shown).

|                          |       | Holetta |       |       | Robgebeya |       |
|--------------------------|-------|---------|-------|-------|-----------|-------|
| Treatments               | N (%) | P (%)   | K (%) | N (%) | P (%)     | K (%) |
| OA                       |       |         |       |       |           |       |
| Control                  | 2.65  | 0.28    | 3.07  | 1.75  | 0.27      | 2.09  |
| В                        | 3.01  | 0.32    | 3.39  | 1.92  | 0.29      | 2.33  |
| Com                      | 3.19  | 0.34    | 3.63  | 2.12  | 0.31      | 2.39  |
| Com + B                  | 3.11  | 0.31    | 3.34  | 2.03  | 0.30      | 2.29  |
| COMBI                    | 3.02  | 0.30    | 3.22  | 1.91  | 0.31      | 2.38  |
| LSD                      | 0.34  | 0.05    | 0.33  | 0.29  | 0.03      | 0.19  |
| OA vs. Con               | ***   | **      | **    | ***   | **        | **    |
| N (kg ha <sup>-1</sup> ) |       |         |       |       |           |       |
| 0                        | 2.66  | 0.30    | 3.11  | 1.68  | 0.27      | 2.13  |
| 23                       | 2.86  | 0.32    | 3.27  | 1.88  | 0.30      | 2.32  |
| 46                       | 3.08  | 0.31    | 3.25  | 1.98  | 0.30      | 2.36  |
| 69                       | 3.10  | 0.31    | 3.44  | 2.03  | 0.29      | 2.26  |
| 92                       | 3.22  | 0.32    | 3.57  | 2.13  | 0.30      | 2.43  |
| LSD (0.05)               | 0.18  | 0.02    | 0.21  | 0.14  | 0.01      | 0.12  |
| Control vs. N            | ***   | NS      | **    | ***   | ***       | ***   |
| N linear                 | ***   | *       | ***   | ***   | ***       | ***   |
| N quadratic              | NS    | NS      | NS    | NS    | ***       | NS    |

**Table 5.5.** Table of means for main effects of organic amendments and N fertilizer on nutrient concentration of barley shoots at booting stage at both sites.

Significant at  ${}^*p \le 0.05$ ,  ${}^{**}p \le 0.01$ ,  ${}^{***}p \le 0.001$ ; NS: Not significant. LSD: Least significant difference at 5% significance level; N: Nitrogen.

### 5.3.3. Soil physicochemical properties

Soil physicochemical properties, measured after harvesting of the barley, differed substantially between organic soil amendments. Soil water content (SWC), soil pH, SOC, total N, available P, exchangeable K, Ca, Mg, and CEC were significantly ( $p \le 0.05$ ) improved by organic amendments at both sites (Table 5.2). Use of B, Com, Com + B, and COMBI increased SWC by 21, 17, 20 and 17% at Holetta and 28, 23, 22 and 25% at Robgebeya, respectively. Addition of B, Com, Com + B, and COMBI considerably increased soil pH, by 0.51, 0.24, 0.29 and 0.27 units at Holetta, and 0.52, 0.35, 0.41 and 0.48 units at Robgebeya, respectively, relative to the control. Soil pH, SOC, total N,

available P, and CEC were higher at Holetta than at Robgebeya, in keeping with the differences in pre-trial soil analysis results, as well as yield and shoot nutrient concentration of barley between sites. SOC content ranged from 2.38 - 2.94% at Holetta and 1.42 - 1.93% at Robgebeya, with the highest values being in plots amended by biochar. Overall, the average SOC content decreased in the order B > Com + B > COMBI > Com > Control at both locations (Tables 5.6 and 5.7). Soil N concentration was 0.22 - 0.28% at Holetta and 0.14 - 0.15% at Robgebeya, and extractable P concentration was 14.6-19.0 mg kg<sup>-1</sup> at Holetta and 6.4 - 11.0 mg kg<sup>-1</sup> at Robgebeya, with the lowest values being from the control and the highest from Com and Com + B soil amendments (Tables 5.6 and 5.7). Biochar addition resulted in the highest CEC at both locations, but significant differences were not observed among organic amendments.

Most soil physicochemical properties did not respond significantly or consistently to N fertilizer rate. Soil Mg and CEC were the only soil chemical properties significantly ( $p \le 0.05$ ) affected by N fertilizer rate at both locations. Soil pH varied significantly between N fertilizer levels only at Robgebeya. SWC and soil pH exhibited significant quadratic, and Mg linear, responses to N fertilizer rate only at Robgebeya (Table 5.7). There was a significant interaction between organic amendment and N fertilizer rate ( $p \le 0.05$ ) for SOC at Robgebeya (Table 5.2). Application of B and 92 kg ha<sup>-1</sup> resulted in the highest SOC (1.96%) at Robgebeya, which was 42% higher than the control.

|                          | SWC  | SBD                   | Soil | SOC  | Total N | Soil P         | Exchangeable cations (cmol(+) kg <sup>-1</sup> ) |       |      |      |      |  |
|--------------------------|------|-----------------------|------|------|---------|----------------|--|-------|------|------|------|--|
| Treatments               | (%)  | (g cm <sup>-3</sup> ) | pН   | (%)  | (%)     | $(mg kg^{-1})$ | К  | Na    | Ca   | Mg   | CEC  |  |
| OA<br>O to 1             | 20.7 | 1.00                  | 5 10 | 2.29 | 0.22    | 14.6           | 0.70   | 0.41  | 7.24 | 2.45 | 16.0 |  |
| Control                  | 39.7 | 1.00                  | 5.12 | 2.38 | 0.22    | 14.6           | 0.79   | 0.41  | 7.34 | 2.45 | 16.2 |  |
| В                        | 47.9 | 0.93                  | 5.63 | 2.94 | 0.27    | 18.3           | 0.93   | 0.44  | 9.10 | 2.77 | 20.1 |  |
| Com                      | 46.6 | 0.94                  | 5.36 | 2.86 | 0.28    | 19.0           | 0.94   | 0.43  | 8.68 | 2.83 | 19.2 |  |
| Com + B                  | 47.7 | 0.91                  | 5.41 | 2.89 | 0.28    | 16.7           | 0.89   | 0.43  | 8.95 | 2.78 | 19.4 |  |
| COMBI                    | 46.5 | 0.90                  | 5.39 | 2.88 | 0.27    | 16.8           | 0.85   | 0.44  | 8.83 | 2.72 | 19.3 |  |
| LSD                      | 2.6  | 0.05                  | 0.26 | 0.10 | 0.01    | 2.76           | 0.08   | 0.03  | 0.48 | 0.15 | 0.94 |  |
| OA vs. Con               | ***  | **                    | ***  | ***  | **      | **             | **   | NS    | **   | **   | **   |  |
| N (kg ha <sup>-1</sup> ) |      |                       |      |      |         |                |  |       |      |      |      |  |
| 0                        | 45.8 | 0.94                  | 5.39 | 2.80 | 0.25    | 16.7           | 0.90   | 0.42  | 8.52 | 2.64 | 18.6 |  |
| 23                       | 45.2 | 0.93                  | 5.34 | 2.75 | 0.26    | 17.0           | 0.87   | 0.44  | 8.61 | 2.69 | 18.8 |  |
| 46                       | 46.3 | 0.94                  | 5.43 | 2.81 | 0.26    | 17.3           | 0.89   | 0.45  | 8.79 | 2.77 | 19.2 |  |
| 69                       | 45.3 | 0.94                  | 5.41 | 2.79 | 0.26    | 16.7           | 0.88   | 0.43  | 8.41 | 2.65 | 18.4 |  |
| 92                       | 45.8 | 0.93                  | 5.31 | 2.80 | 0.27    | 17.5           | 0.86   | 0.43  | 8.56 | 2.68 | 18.6 |  |
| LSD (0.05)               | 1.2  | 0.02                  | 1.5  | 0.08 | 0.006   | 0.94           | 0.06   | 0.016 | 0.29 | 0.09 | 0.56 |  |
| Con vs. N                | NS   | NS                    | NS   | NS   | NS      | NS             | NS   | NS    | NS   | NS   | NS   |  |
| N linear                 | NS   | NS                    | NS   | NS   | NS      | NS             | NS   | NS    | NS   | NS   | NS   |  |
| N quadratic              | NS   | NS                    | NS   | NS   | NS      | NS             | NS   | NS    | NS   | NS   | NS   |  |

**Table 5.6.** Mean soil physicochemical properties as influenced by organic amendments (OA) and N fertilizer rates (main effects) after harvesting barley at Holetta.

Significant at  ${}^{*}p \le 0.05$ ,  ${}^{**}p \le 0.01$ ,  ${}^{***}p \le 0.001$ ; NS: Not significant. OA: Organic amendment; Con: Control; LSD: Least significant difference; SBD: Soil bulk density; SOC: Soil organic carbon; SWC: Soil water content.

|                          | SWC  | WC SBD                |      | SOC  | Total N | Soil P            | Ех   | cchangeable | e cations (c | mol(+) kg <sup>-</sup> | 1)    |
|--------------------------|------|-----------------------|------|------|---------|-------------------|------|-------------|--------------|------------------------|-------|
| Treatments               | (%)  | (g cm <sup>-3</sup> ) | pН   | (%)  | (%)     | $(mg kg^{-1})$    | K    | Na          | Ca           | Mg                     | CEC   |
| OA<br>O (                | 27.0 | 0.00                  | 4.05 | 1.40 | 0.1.4   | <i>C</i> <b>A</b> | 0.64 | 0.22        | ( )1         | 0.07                   | 1.5.4 |
| Control                  | 37.9 | 0.96                  | 4.85 | 1.42 | 0.14    | 6.4               | 0.64 | 0.32        | 6.21         | 2.37                   | 15.4  |
| В                        | 48.7 | 0.89                  | 5.37 | 1.93 | 0.15    | 8.8               | 0.74 | 0.41        | 7.50         | 2.81                   | 19.1  |
| Com                      | 46.8 | 0.88                  | 5.20 | 1.69 | 0.15    | 10.3              | 0.70 | 0.41        | 7.79         | 2.83                   | 18.6  |
| Com + B                  | 46.4 | 0.87                  | 5.26 | 1.81 | 0.15    | 11.0              | 0.71 | 0.40        | 7.49         | 2.80                   | 18.5  |
| COMBI                    | 47.5 | 0.88                  | 5.33 | 1.73 | 0.15    | 9.5               | 0.71 | 0.39        | 7.64         | 2.93                   | 19.0  |
| LSD                      | 3.4  | 0.03                  | 0.15 | 0.21 | 0.006   | 2.6               | 0.07 | 0.03        | 0.93         | 0.31                   | 2.02  |
| OA vs. Con               | **   | **                    | ***  | ***  | *       | ***               | **   | **          | **           | **                     | ***   |
| N (kg ha <sup>-1</sup> ) |      |                       |      |      |         |                   |      |             |              |                        |       |
| 0                        | 46.2 | 0.89                  | 5.25 | 1.65 | 0.14    | 9.6               | 0.69 | 0.37        | 7.20         | 2.67                   | 17.7  |
| 23                       | 44.7 | 0.89                  | 5.15 | 1.71 | 0.14    | 8.9               | 0.70 | 0.38        | 7.41         | 2.71                   | 18.2  |
| 46                       | 44.5 | 0.90                  | 5.13 | 1.63 | 0.15    | 9.1               | 0.72 | 0.39        | 7.48         | 2.83                   | 18.5  |
| 69                       | 45.1 | 0.89                  | 5.26 | 1.68 | 0.15    | 8.8               | 0.69 | 0.39        | 7.15         | 2.72                   | 17.8  |
| 92                       | 46.9 | 0.90                  | 5.22 | 1.71 | 0.15    | 9.7               | 0.71 | 0.38        | 7.40         | 2.82                   | 18.3  |
| LSD (0.05)               | 1.8  | 0.02                  | 0.08 | 0.07 | 0.005   | 1.8               | 0.04 | 0.017       | 0.29         | 0.13                   | 0.64  |
| Con vs. N                | NS   | NS                    | *    | NS   | NS      | NS                | NS   | NS          | NS           | *                      | *     |
| N linear                 | NS   | NS                    | NS   | NS   | NS      | NS                | NS   | NS          | NS           | *                      | NS    |
| N quadratic              | **   | NS                    | *    | NS   | NS      | NS                | NS   | NS          | NS           | NS                     | NS    |

**Table 5.7.** Mean soil physicochemical properties as influenced by organic amendments (OA) and N fertilizer rates (main effects) after harvesting barley at Robgebeya.

Significant at  ${}^{*}p \le 0.05$ ,  ${}^{**}p \le 0.01$ ,  ${}^{***}p \le 0.001$ ; NS: Not significant. OA: Organic amendment; Con: Control; LSD: Least significant difference; SBD: Soil bulk density; SOC: Soil organic carbon; SWC: Soil water content.

### 5.4. Discussion

### 5.4.1. Yield and yield components

In our study, the addition of organic amendments and N fertilizer resulted in clear effects on yield and yield components of barley in the Ethiopian highlands. In this environment, where demographic and economic pressures are intense and most farming is for subsistence, soil fertility depletion is severe and use of external inputs, whether organic or inorganic, is very limited. The farmers' main response to such pressures has been to grow barley every year in a farming system with limited or no amendments to replenish the soil. This strategy appears to be unsustainable because crop yields and soil fertility have been declining (Agegnehu et al., 2014a; Zelleke et al., 2010). In this study, the application of Com and Com + B (without added N) resulted in similar yields to the highest rate of N fertilizer at Holetta, but higher yields than all organic amendments (without added N) at Robgebeya. Importantly, barley yields were substantially improved with all organic amendments, irrespective of the rate of N fertilizer applied. The positive effects of organic amendments on yield resulted from changes to the soil, including lower bulk density, higher water retention and plant available water, and increased micronutrients, soil pH and contents of available P and possibly K. The increase in soil pH was probably a particularly important effect, as the soils at both sites have sufficiently low pH to affect P availability and barley growth. There may also have been an N effect, but it was probably small, since the positive effect of the organic amendments was similar at all N rates.

The positive effect of organic amendment was greater at Robgebeya where the soil is poorer and yield more N-limited than at Holetta. At Holetta, COMBI influenced yield similarly to Com, but at Robgegeya COMBI tended to be less beneficial than Com alone. Com gave similar results to Com + B at both sites, although at Holetta its optimum effect was at a higher N rate than Com + B alone. Despite the same soil type at both sites, the soil at Robgebeya was more N-limited, and Com and COMBI provide energy for soil biota, generally resulting in healthier soil, and B is also known to stimulate biological activity. Several studies indicated that application of biochar to soil improves soil fertility (Biederman and Harpole, 2013; Kookana et al., 2011; Verheijen et al., 2009) as a result of its effects on physicochemical and biological properties, creates a favourable habitat for microorganisms due to improved soil porosity (Lehmann and Joseph, 2015; Steinbeiss et al., 2009), as well as increase plant nutrient availability in nutrient-limited agroecosystems (Major et al., 2010).

Application of compost and biochar, individually or together, when combined with fertilizer enhanced leaf chlorophyll and productive tillers over mineral fertilizer alone. This could also indicate increased nutrient availability, vigorous plant growth and healthier plants, resulting in higher yields. Studies have shown that the nutrient supplying capacity of compost and manure, applied singly or in combination with biochar, are higher than biochar alone (Fischer and Glaser, 2012; Schulz and Glaser, 2012). Liu et al. (2012) also demonstrated a synergistic positive effect of compost and biochar mixtures on soil organic matter content, nutrient content and water-storage capacity of a sandy soil under field conditions. Although all organic soil amendments significantly enhanced shoot nutrient concentration at both sites, the N, P and K concentrations of barley plants were significantly higher at Holetta than at Robgebeya. This implies that poor soils are less responsive to external inputs due to other constraints, besides the nutrients contained in the fertilizer or organic soil amendments. The difference in soil fertility between the fields has led to large gaps in both control and attainable yields, and nutrient use efficiencies. Zingore et al. (2007) indicated that N use efficiency by maize varied from > 50 kg grain kg<sup>-1</sup> N on the more fertile fields, to less than 5 kg grain kg<sup>-1</sup> N in degraded fields.

Soil fertility was sub-optimal for the production of barley, particularly at Robgebeya, so crop growth and yields were directly related to soil properties. Organic amendments increased grain yield by 51-78% at the less fertile Robgebeya site, compared to 30-49% at Holetta. The main benefits of organic amendments compared with mineral fertilizer are that, in addition to supplying nutrients, they improve physical, chemical and biological aspects of soil fertility (Agegnehu et al., 2015a; Bolan et al., 2012; Chen et al., 2013; Dil and Oelbermann, 2014; Domene et al., 2014). The nutrients in the Com, Com + B, or COMBI can enhance soil nutrient content and become available to the crop as the compost or manure mineralizes over time. Application of biochar also possibly enhanced nutrient uptake (N, P and K), as indicated by increased nutrient concentrations in leaves in conjunction with increased shoot biomass. For N and P, this effect is likely to have been at least partly due to changed conditions for root growth rather than supply of the actual nutrients, because the concentrations of these elements were low in the biochar. According to Prendergast-Miller et al. (2014), barley plants in biochar-amended soils had larger rhizosphere zones than the control treatment. Similarly, addition of 4 t ha<sup>-1</sup> of maize cob biochar on weathered soils in Zambia significantly increased the root system and yield of maize (Abiven et al., 2015). This study revealed that biochar soil amendment showed a considerable effect on suppression of weeds, perhaps through an inhibitory effect on weed seed germination. Arif et al. (2012) reported that application of biochar at the rate of 25 t ha<sup>-1</sup> and 5 t ha<sup>-1</sup> of FYM resulted in a lower weed population 30 and 60 days after planting maize. Similarly, reapplication of biochar significantly reduced the cumulative emergence of weed seedlings and total

biomass by day 28 (Quilliam et al., 2012), and weed biomass, measured at harvest, was less in biochar treated plots (Vaccari et al., 2011).

Nitrogen fertilizer application at optimum level is required to enhance the growth and yield of barley. Moderate rates increase yield, but excessively high rates could result in increased plant height and lodging, reduced test weight, and yield (Agegnehu et al., 2014a). In integrated soil fertility management systems, N fertilizer strategies for barley production should ensure adequate amounts of available N for crop establishment and tiller development, and the amount required should be based on the cropping system and soil fertility management practices. The response to N fertilizer (i.e., slope of yield vs application rate) was greater at Robgebya than at Holetta. Nitrogen supply appeared to be a much greater factor limiting yield at Robgebeya than at Holetta, which was related to the soil total N content and more likely the higher available N (extractable nitrate and ammonium) before planting and uptake by plants. It is also worth mentioning that the optimum rate of application does not appear to have been reached at Robgebeya (i.e., > 92 kg N ha<sup>-1</sup>).

In this study, increases in yield and yield components were more pronounced when organic amendments and N fertilizer were both applied, in comparison to one or the other. Application of organic amendments stimulated plant growth and yield, especially when combined with mineral fertilizer, which is in agreement with the results of several previous studies (Alburguerque et al., 2013; Doan et al., 2015; Schulz and Glaser, 2012; Steiner et al., 2008). Organic soil amendments and N fertilizer up to 92 kg ha<sup>-1</sup> (higher than the existing practice) increased yield and yield components of barley. Biochar and N fertilizer had essentially positive and additive effects at both sites, whereas the positive effect of organic amendments containing compost was greatest at moderate levels of N application (23-69 kg ha<sup>-1</sup>) at both sites. This may be due to the fact that the compost and N fertilizer can partially substitute for one another; i.e., both supply N, whereas N supply by biochar is unlikely. Other studies have also shown that addition of manure plus mineral fertilizer significantly increased yields of maize and wheat (Kaur et al., 2008; Meade et al., 2011), and biochar plus mineral fertilizer yield of sorghum (Blackwell et al., 2015) relative to mineral fertilizer alone. Abewa et al. (2013) found that application of 12 t ha<sup>-1</sup> eucalyptus biochar and 40/30 kg ha<sup>-1</sup> N/P fertilizer on acid Nitisols in Ethiopia increased tef yield by 67% relative to the same NP fertilizer rate without biochar. Nutrientpoor soil amended with low C algal biochars, without and with mineral fertilizer, increased sorghum growth rate between 15 and 32 times, respectively, compared to the control without biochar (Bird et al., 2012). Gathorne-Hardy et al. (2009) reported that application of 50 t ha<sup>-1</sup> biochar alone did not result in significant effect on barley yield, but the same amount with 100 kg N ha<sup>-1</sup> significantly increased barley yield by 30%. Overall, inclusion of organic soil amendments in the production system improves fertilizer use efficiency, and ensures yield and sustainability in the long-term.

Correlation of grain yield with total biomass was the highest at both sites (Figures 5.3 and 5.4). A possible reason for the significant relationship of grain yield with SOC, CEC and SWC is that the organic amendments have the capacity to improve the soil water and nutrient content that can be available to the growing crop. The direct effect on the performance of the barley crop of available soil nutrients and plant nutrient uptake from organic soil amendments and N fertilizer exceeded the direct effect of available nutrients and nutrient uptake from N fertilizer alone. Other studies have also shown linear correlations between soil nutrient content, yield, and yield components of peanut and maize as a result of application of biochar, compost and their mixture (Agegnehu et al., 2016a; Agegnehu et al., 2015a), and between soil characteristics, seed germination, root and shoot growth of wheat due to biochar addition (Solaiman et al., 2012).

### 5.4.2. Nutrient uptake and soil physicochemical properties

The significantly higher yield and plant biomass on organic-amended plots was due to increased nutrient uptake, and perhaps increased water uptake and transpiration, by the plants over their growth cycle. The increase in the shoot N concentration of barley with organic amendments was similar to that resulting from N fertilizer addition. However, plant P and K concentrations were higher for organic-amended than N fertilized soil, suggesting that organic treatments improve the supply of other essential macro- and micro-nutrients and water. Although shoot N, P and K concentrations were in the reported sufficiency range for all treatments (Jones Jr, 2003), the trial site Holetta with the higher yield, C and N in the soil had mean shoot N and K concentration of 3.19 and 3.63% for Com and 3.22 and 3.57 % for the 92 kg N ha<sup>-1</sup> compared to shoot N concentration of 2.12 and 2.39% for Com and 2.13 and 2.43% for 92 kg N ha<sup>-1</sup> at Robgebeya. This indicates that the application of organic amendments directly influences the availability of native or applied nutrients. The plant nutrient content at Holetta was higher than at Robgebeya, reflecting differences in yield and soil fertility between the sites. However, in the long term, organic amendments and fertilizer may have substantial effects at less fertile sites. The trend in plant N uptake increases in relation to organic amendments and N levels were similar to the increments in plant growth, yields and soil nutrient status.

The solubility and availability of important nutrients to plants is closely related to the pH of the soil. Excessive soil acidity results in a shortage of available Ca, P and Mo on the one hand, and an excess of soluble Al, Mn and other cations on the other (Fageria and Baligar, 2008). In this study, organic

soil amendments improved plant P-uptake, suggesting that Com and Com + B supplied P to the soil and also improved its availability by reducing sorption and leaching, supporting the results of other studies (Inal et al., 2015; Lehmann et al., 2003). Ferralsols amended with biochar and compost lowers leaching and improves the root-fertilizer contact, thus optimizing the availability of P, B and Mo (Agegnehu et al., 2015a; Van Zwieten et al., 2015) to plants.

The present study also clearly demonstrated that organic amendments substantially improved SWC. The fact that there was greater SWC and greater growth (which entails greater transpiration) must have been due to greater infiltration and/or retention of water during the growing season. Runoff was not measured, but it is common in the Ethiopian highlands. Other studies have also shown that biochar addition increased soil total pore volume and water content and biomass, yield and N use efficiency of plants (Abel et al., 2013; Haider et al., 2015). Baronti et al. (2014) also found that during droughts, application of orchard pruning waste biochar at the rate of 22 and 44 t ha<sup>-1</sup> increased available soil water content by 3.2 and 45% and leaf water potential by 24 and 37%, respectively, compared to control soils.

Organic amendments appear promising as a strategy to address the nutrient deficiencies of Nitisols, which is in conformity with the results of pre-planting soil analysis at both sites, where soil nutrients are particularly low at Robgebeya. In such soils, the proportion of P fertilizer that could be available to a crop becomes inadequate, unless ameliorated with organic and/or liming materials. Experimental plots treated with organic amendments exhibited improvements in soil physicochemical properties. Despite being under the same land-use, similar climatic conditions, and similar pH values, the Nitisol at Holetta has an OC twice that of the Nitisol at Robgebeya. In the current study, various plant nutrient concentrations responded positively to organic amendments, an observation consistent with previous studies in acidic and highly weathered tropical soils (Agegnehu et al., 2015a; Lehmann et al., 2003; Zhao et al., 2014a). Biochar and Com + B addition provided greater benefit to barley yield, possibly by reducing Al toxicity and improving P nutrition.

Soil reaction (pH) is an important characteristic of soils in terms of nutrient availability and plant growth. It is common practice to amend acidic soils by adding agricultural lime to raise the pH, which allows plants to grow at their maximum potential when other requirements, such as water and nutrient availability, are met. Soil pH increased from the initial value of 4.97 to 5.63 at Holetta and from 4.83 to 5.37 at Robgebeya at harvest, due to biochar soil amendment, with significant differences between organic amended and un-amended plots for soil pH at both sites. Similar liming effects of biochar have been recorded in previous studies on similar soils (Lehmann et al., Glaser et al., 2002; 2003).

Dume et al. (2015) reported that acidic Nitisols, amended by coffee husk and maize cob biochars, at rates of 5-15 t ha<sup>-1</sup> increased soil pH from 5.3 to 6.7 compared to the control (5.2). Application of lime + compost + NP fertilizer increased barley yield by 109 and 152%, respectively, in acid Nitisols of Ethiopian highlands (Regassa and Agegnehu, 2011). Biochar applications to soils can improve soil fertility by increasing the CEC and nutrient retention of soils. Soils with low CEC are often low in fertility and vulnerable to soil acidification. The buffering capacity of a soil increases with the increase in CEC and SOC content. Xu et al. (2012b) reported that the enhanced CEC increased soil fertility through greater nutrient availability, as nutrients are retained in the soil against leaching. The application of 20 t biochar ha<sup>-1</sup> to a low-fertility, acidic soil of Colombia led to significant increases in concentrations of several nutrients in soil solution (Major et al., 2012).

Replenishment of organic matter derived from plant, animal and microbial biomass, in all stages of decomposition, is critical to ensuring long-term soil fertility; it provides a balanced medium for nutrients and water for plant growth. Although significant quantities of manures and crop residues are produced in the country as potential feedstock for biochar and compost production, they are not returned to soil due to competing utilization. For example, some estimates suggest the nutrient contents of the crop residues, used as feed and manures as fuel instead of fertilizer, are higher than the quantities applied as fertilizers. In other words, this lack of alternative fuel and feed sources is a significant constraint on productivity and sustainability of the broader agricultural system in the highlands (Zelleke et al., 2010). Application of biochar and biochar-compost mixes could potentially decrease the N fertilizer requirement for crop growth. Another important aspect of lowering N fertilizer dose is slowing the rate of soil acidification, which is a problem in many parts of the world. Decreasing N fertilizer application rate can, in turn, reduce the cost of producing food, while simultaneously mitigating environmental pollution. The effective use of organic resources as nutrient sources is central to achieving the long-term need for increased biomass production for food and soil fertility and, hence, sustainably higher productivity, critical to breaking the poverty cycle. Specific actions include improving the local supply of affordable fuel alternatives, efficiency of stoves and availability of affordable feed and forage sources. Overall, in the highlands, the integrated use of all the available resources, including lime and organic amendments, to improve and sustain soil health and crop yield is of great practical significance.

### 5.5. Conclusions

The results of this study highlight the benefits of the application of organic amendments for improving the quality of Nitisols, and in promoting barley growth and yield. High yields were obtained with

application of Com or Com + B, and the highest yields were achieved in combination with moderate rates of applied N. Important findings in this study were that results displayed significant improvement in the quality characteristics of the soil amended with B, Com, and Com + B. Furthermore, the growth and yield components of barley, supplemented with organic amendments and lower N fertilizer rates, were significantly higher than observed at the highest N rate (92 kg ha<sup>-1</sup>) alone, showing the enhanced synergistic effects of mixed treatments. Biochar and biochar-compost soil amendments increased SOC by 23-34% and CEC by 19-24%, compared to the initial values. The positive effects of organic amendments on soil properties and crop yield were greater at the site with lower soil fertility. Any of the organic-inorganic amendment options identified in this study can be recommended to improve soil fertility and yield of barley. However, although B, Com, Com + B, and COMBI-mix were all good soil fertility ameliorants, Com and Com + B were the best in terms of barley yield. Generally increasing rate of N fertilizer increased yield with optimal fertiliser rates, up to 69 kg ha<sup>-1</sup> at Holetta and 92 kg ha<sup>-1</sup> at Robgebeya, irrespective of the organic amendment also applied.

As crop production in the highlands of the country is entirely rain-fed, and terminal moisture stress is a critical problem at grain filling stage, the positive effects of B or Com + B application on soil moisture retention is likely to be a critical factor. The addition of compost-biochar mixes to soils may have stabilizing effects, especially on easily degradable components of the compost. Overall, the use of organic amendments in a farming system can improve soil biophysical and chemical properties, maintain satisfactory crop yield, reduce the costs of production, therefore, increasing profitability, and enhance long-term sustainability of the production system. Moreover, the interaction between organic amendment and N fertilizer should be tested over longer periods involving representative locations across major barley producing areas of the country.

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### **Overview of Chapter 6**

This chapter is based on a paper published in the *Science of the Total Environment* with minimal formatting changes. I have investigated the interaction effects of organic amendments (compost, biochar, compost, biochar-compost mix and co-composted biochar-compost), and five levels of nitrogen fertilizer on nutrient uptake and nitrogen use efficiency of barley. The trial sites were located in the central highlands of Ethiopia at two sites (one at Holetta Research Center and the other on a farmer's field) on Nitisols.

The study was a factorial experiment in a split plot design, with organic amendments as main plots and N fertilizer levels as sub-plots. I have determined grain N uptake (GNU) and straw N uptake (SNU), the agronomic efficiency (yield increase per unit of N applied, AE), apparent recovery efficiency (increase in N uptake per unit of N applied, ARE), and physiological efficiency (yield increase per unit of N uptake, PE). I organized, analyzed and interpreted the data, and wrote the manuscript for publication. This study is very important in terms of optimizing the N use efficiency of crops through the use of organic soil amendments.<sup>6</sup>

<sup>&</sup>lt;sup>6</sup> Agegnehu, G., Nelson, P. N., and Bird, M. I. (2016). The effects of biochar, compost and their mixture and nitrogen fertilizer on yield and nitrogen use efficiency of barley grown on a Nitisol in the highlands of Ethiopia. *Science of The Total Environment* 569–570, 869–879.

## Chapter 6

# The effects of biochar, compost and their mixture, and nitrogen fertilizer on yield and nitrogen use efficiency of barley grown on a Nitisol in the highlands of Ethiopia

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### Abstract

The effects of organic amendments and nitrogen (N) fertilizer on yield and N use efficiency of barley were investigated on a Nitisol of the central Ethiopian highlands in 2014. The treatments were factorial combinations of no organic amendment, biochar (B), compost (Com), Com + B, and cocomposted biochar (COMBI) as main plots and five N fertilizer levels as sub-plots, with three replicates. Application of organic amendment and N fertilizer significantly improved yield, with grain yield increases of 60% from Com + B + 69 kg N ha<sup>-1</sup> at Holetta and 54% from Com + 92 kg N ha<sup>-1</sup> at Robgebeya, compared to the yield from the maximum N rate. The highest total N uptake was obtained from Com + B + 92 kg N ha<sup>-1</sup> at Holetta (138 kg ha<sup>-1</sup>) and Com + 92 kg N ha<sup>-1</sup> at Robgebeya (101 kg ha<sup>-1</sup>). The agronomic efficiency (yield increase per unit of N applied, AE), apparent recovery efficiency (increase in N uptake per unit of N applied, ARE), and physiological efficiency (yield increase per unit of N uptake, PE) responded significantly to organic amendments and N fertilizer. Mean AE and ARE were highest at B + 23 kg N ha<sup>-1</sup> at Holetta and at B + 23 and B + 46 kg N ha<sup>-1</sup> at Robgebeya. The PE ranged from 19 - 33 grain kg<sup>-1</sup> N uptake at Holetta and 29 - 48 kg grain kg<sup>-1</sup> N uptake at Robgebeya. The effects of organic amendments and N fertilizer on AE, ARE and PE, were greater at Robgebeya than at Holetta. The enhancement of N use efficiency through application of organic amendments emphasizes the importance of balanced crop nutrition, ensuring that barley crops are adequately supplied with N and other nutrients. Overall, the integration of both organic and inorganic amendments may optimize N uptake efficiency and reduce the amount of N fertilizer required for sustainable barley production in the long-term.

**Keywords:** Barley, biochar, compost, co-composted biochar-compost, nitrogen fertilizer; nitrogen use efficiency

### 6.1. Introduction

Soil nutrient depletion and low nutrient use efficiency are major constraints for the productivity of crops in Sub-Saharan Africa. Barley (*Hordeum vulgare* L.) is one of the world's main cereals, ranking fourth in production after wheat, maize and rice (FAO, 2013). It is the fifth most important cereal crop in Ethiopia after tef (*Eragrostis tef*), maize, sorghum, and wheat in production area and tonnage (CSA, 2013). Barley is grown predominantly under rain-fed conditions, by subsistence farmers in the highlands of the country (2,000 to 3,500 m above sea level), in areas receiving mean annual rainfall greater than 1,000 mm (Agegnehu et al., 2014a). The crop covers ~1.018 million ha, but the national average yield is very low at 1.75 t ha<sup>-1</sup> (CSA, 2013) compared to the global average barley yield of

2.6 t ha <sup>-1</sup> (Ullrich, 2011), due to poor soil fertility, particularly N and P, and low N use efficiency (Agegnehu et al., 2011). Depletion of soil nutrients by cropping is a major problem in the country; Haileslassie et al. (2005) reported a depletion rate of 122 kg N ha<sup>-1</sup> yr<sup>-1</sup>, 13 kg P ha<sup>-1</sup> yr<sup>-1</sup>, and 82 kg K ha<sup>-1</sup> yr<sup>-1</sup> at the national level. Although barley grows across a wide range of soil conditions (Agegnehu et al., 2011), highly weathered reddish brown clay soils (Nitisols) are the most important soils for production in Ethiopia (Bekele and Höfner, 1993).

Although there is a considerable potential for increased barley production, productivity is constrained by low soil pH, nutrient deficiencies, and low levels of fertilizer application (Agegnehu et al., 2011). Soil acidity adversely affects morphological, physiological and biochemical processes in plants and, thus, N uptake and use efficiency (Fageria and Baligar, 2005; Marschner, 2011). Inefficient use of fertilizer contributes to the depletion of scarce financial resources, increased unit production costs, and potential environmental risks. Hence, optimizing the efficiency of N fertilizer use must be a major consideration for subsistence producers. Cereal-dominated cropping systems, aimed at meeting farmers' subsistence requirements, coupled with low usage of fertilizer, have led to wide spread depletion of soil nutrients in the major barley growing regions of the country (Agegnehu et al., 2014a). Only 48.3% of the total area of land covered by barley receives fertilizer, which is the lowest among all the cereals grown in Ethiopia (Mulatu and Lakew, 2011). Heavy rains during the main cropping season (June to August) also cause substantial soil nutrient losses, due to intense leaching and erosion on Nitisols (Tarekegne et al., 1997). Several studies have demonstrated considerable potential for increasing barley yields by applying organic and inorganic fertilizers and improved crop management practices (Agegnehu et al., 2014a; Hejcman et al., 2013; Tarekegne et al., 1997), and these practices may also be beneficial for nutrient use efficiency.

Use of nitrogen fertilizer in low-input subsistence farming may improve yields and quality, and lower risk of crop failure, but response to fertilizer may be limited by low inherent physical, biological and chemical fertility of soil. Nitrogen use efficiency in crop plants is defined in several ways (Baligar et al., 2001; Dawson et al., 2008; Fageria and Baligar, 2005). In simple terms, N use efficiency is the ratio of output (economic yield) to input (fertilizers) for a process or system (Dawson et al., 2008; Fageria, 2008). Fertilizer N use efficiency is governed by three major factors, which include N uptake by the crop, N supply from soil and fertilizer, and N losses from soil–plant systems. The crop N requirement is the most important factor influencing N use efficiency (Ladha et al., 2005). Nitrogen use efficiency of cereals is estimated to be 42% and 29% in developed and under-developed nations, respectively (Raun and Johnson, 1999). Worldwide, N use efficiency

of cereals is about 33%, and the 67% not accounted for represents a \$15.9 billion annual loss in N fertilizer costs (Raun and Johnson, 1999). Fan et al. (2004) also reported that the average N fertilizer recovery in cereals in China was 30 - 35%. Thus, improving N use efficiency through a combination of agronomic and soil management methods is of paramount importance in terms of profitability and environmental management. In cereals, nutrient use efficiency has been expressed relative either to the total nutrient supply in the soil and fertilizer (Giambalvo et al., 2004) or to the applied fertilizer nutrient alone (Baligar et al., 2001; Fageria, 2008). Given the difficulty of precise measurements of available soil nutrients and the economic importance of fertilizer nutrients, the latter approach is preferred (Sinebo et al., 2004; Xu et al., 2012a). The components of fertilizer N use efficiency are agronomic efficiency, apparent recovery efficiency and physiological efficiency (Baligar et al., 2001; Fageria, 2008; Xu et al., 2012a). Various authors have reported that apparent N recovery efficiency is a suitable indicator to evaluate the efficiency of fertilizer management practices (Baligar et al., 2001; Shejbalová et al., 2014; Tarekegne and Tanner, 2001).

The rate of N fertilizer application depends on the purpose for which barley is grown. It is desirable to apply N fertilizer at a higher rate for feed or food barley than for malting barley, as protein is more important in food barley than in malting barley, and barley with a high protein content is difficult to malt (Agegnehu et al., 2014a; Jankovic and Ikanovic, 2011). In Ethiopia, where pH, organic carbon, and N content of most soils are low, the N fertilizer rates for barley production typically range between 23 and 46 kg N ha<sup>-1</sup> (Agegnehu et al., 2014a). Soils with low organic carbon contents have low crop yield and low use efficiency of added nutrients. Fertilizer N efficiency is influenced by the long-term dynamics of organic matter of a soil (Bruulsema et al., 2004). Soil organic matter is vital for sustainable yields as it is able to retain water and nutrients, as well as providing habitat and energy for soil biota and improving soil aggregation (Alburquerque et al., 2013; Courtney and Mullen, 2008; Lal, 2009b). To date, fertilizer application in Ethiopia has been limited, and improvement of agricultural productivity necessitates more than the application of inorganic fertilizers alone.

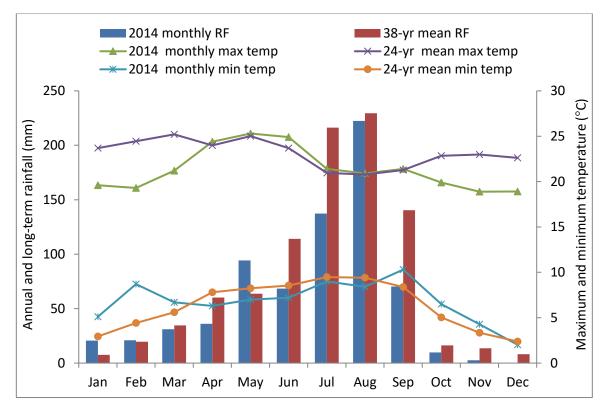
Previous studies indicated that the application of biochar and N fertilizer, on a light soil within a temperate climate, increased yield of spring barley by 30% compared to N fertilizer only (Gathorne-Hardy et al., 2009), and maize biomass by 2.7-3.5 times relative to NPK fertilizer alone on different soil types and agro-climatic conditions in China (Zhu et al., 2015). This indicates that an integrated soil fertility management approach may have more sustainable agronomic and economic impacts, than a focus on inorganic fertilizer alone. No previous studies have been conducted to assess the effects of biochar, compost, biochar-compost, and their interactions with N fertilizer on yield, N uptake, and nitrogen use efficiency of barley in the Ethiopian highlands. The objectives of this study

were to determine the effects of organic amendments, N fertilizer and their interaction on: 1) plant N concentration and uptake; and 2) nitrogen use efficiency of barley under rain-fed conditions on weathered Nitisols.

### 6.2. Materials and methods

### 6.2.1. Characteristics of experimental sites

The experiment was conducted in the main cropping season of 2014 at Holetta Agricultural Research Center (9°3' 19.43" N, 38° 30' 25.43" E, at 2,400 m above sea level), and on a farmer's field at Robgebeya in the central highlands of Ethiopia (9°7' 56.55" N, 38°26' 21.41" E, at 2475 m asl). The long-term average annual rainfall at Holetta is 1,100 mm, of which about 85% falls from June to September, and the remainder from January to May. The year 2014 had a total rainfall of 714 mm, or about 78% of the 38-year average rainfall (924 mm). Average maximum and minimum air temperatures are 22.1°C and 6.2°C, respectively (Figure 6.1). The soil type is acidic Eutric Nitisol at both sites. Nitisols are well drained soils with a deep clayey sub-surface horizon that have typical nutty, or polyhedric blocky peds with shiny faces (IUSS Working Group WRB, 2014). The soil at Holetta has higher organic C, total N, available P, exchangeable K, and CEC than that at Robgebeya (Table 1). Micronutrients, including Cu, Zn and B, appear to be deficient in most Nitisols of the Ethiopian highlands, including at our study sites (Tulema et al., 2007).



**Figure 6.1.** Monthly total rainfall, monthly maximum and minimum temperatures for the 2014 crop growing season including the long-term 38-year average rainfall and 24-year average maximum and minimum temperatures. Note: RF: rainfall; max temp: maximum temperature; min temp: minimum temperature.

Three weeks prior to planting, soil samples were collected from 0 - 20 cm depth at each experimental site. Six sampling locations were selected by dividing the trial area into six cells ( $17 \text{ m} \times 10 \text{ m}$ ). Three sampling points were randomly selected within each of the six cells. At each sampling point, surface organic litter was cleared away, and four samples from a 3-m radius were collected using a manual auger with a 20-cm core barrel with a 6-cm internal diameter. The twelve samples were combined, making a bulk sample for each cell, resulting in six composite samples per site for analysis. After manual homogenization, the samples were ground to pass a 2-mm sieve. Soil samples were analyzed for pH using a ratio of 2.5 ml water to 1 g soil (McLean, 1982); extractable P using Bray 2 solution as extractant; i.e., ammonium fluoride and concentrated hydrochloric acid (Bray and Kurtz, 1945). Soil organic carbon was analyzed by wet oxidation using hydrogen peroxide, chromic acid and sulfuric acid (Walkley and Black, 1934); total N content using Kjeldahl digestion with concentrated H<sub>2</sub>SO<sub>4</sub> and a K<sub>2</sub>SO<sub>4</sub>-catalyst mixture (He et al., 1990); exchangeable cations and CEC using ammonium acetate method (Black, 1965), at the soil and plant analysis laboratory of Holetta Agricultural Research Center. Table 6.1 shows the pre-planting soil chemistry of the trial soil.

### 6.2.2. Experimental set-up and procedure

Biochar, compost and co-composted biochar-compost were produced at Holetta. The feedstock for biochar production was acacia (Acacia spp.), derived from stem, bark and branches. The pyrolysis was undertaken in earth kilns. The earth formed the necessary gas-tight insulating barrier below which carbonization could take place without inflow of air, allowing the biomass to burn away. The kiln was fired and the biomass heated up to allow pyrolysis (~350 - 450 °C for six days). After cooling, the kilns were opened and the biochar was removed. The biochar was crushed to a particle size < 25mm prior to field application. The biochar mass was approximately 15-20% of the original biomass. Compost was prepared following standard procedures (Tulema et al., 2007). A homogenized mixture of farmyard manure, feed leftovers from dairy cattle, and bedding materials was composted in pits 1.5 m  $\times$  2.4 m wide by 1.0 m deep at Holetta. Fresh and decomposing materials were placed in alternating layers, with fresh and straw materials constituting the bottom layer. The compost was turned every two weeks and was mature after fifteen weeks, which was estimated by the temperature and water content of the compost at the composting site (Chikae et al., 2006). The co-composted biochar-compost (COMBI) was prepared following the same procedure, using a ratio of 1:5 biochar to compost on a dry weight basis. The final compost and COMBI had lower C:N ratios (20 and 26, respectively) and higher pH values than the starting feed mixture (Fischer and Glaser, 2012). Biochar, mature compost and COMBI samples were collected from all pits for analysis of chemical properties before they were applied in the field (Table 6.1). The samples were analyzed following the methods used for soil analysis.

The experimental sites were prepared for sowing using standard cultivation practices. Tractormounted disk plowing and disk harrowing was carried out in May and June at the research station, while the on-farm trial field was plowed using oxen-drawn implements. The experiment was a factorial split-plot design with five organic amendments as main plots, and five N fertilizer levels (0, 23, 46, 69 and 92 kg N ha<sup>-1</sup>) as sub-plots, with three replicates for each treatment. Treatment sequencing was randomized and the main and sub-plot areas were  $62.5 \text{ m}^2 (12.5 \times 5 \text{ m})$  and  $10 \text{ m}^2 (5 \times 2 \text{ m})$ , respectively. A space of 0.5 m was left between plots and 1.0 m between blocks, to avoid border effects of treatments. The total area of each trial site was ~1300 m<sup>2</sup>. The organic amendments were: 1) control without amendment; 2) acacia biochar at 10 t ha<sup>-1</sup> (B); 3) compost at 10 t ha<sup>-1</sup> (Com); 4) compost plus biochar at 10 t ha<sup>-1</sup> Com + 2 t ha<sup>-1</sup> B (Com + B) mixed onsite before application; and 5) co-composted biochar-compost applied at 10 t ha<sup>-1</sup> (COMBI). Application rates of B, Com, and COMBI were calculated on an air-dry weight basis. The total N supply from each of B, Com, Com + B, and COMBI was calculated based on the total N concentration of each amendment (Table 6.1), which was 56, 125, 136 and 115 kg ha<sup>-1</sup>, respectively.

| Parameter                              | Unit                     | Com   | В     | COMBI |         | Soil      |
|--|--------------------------|-------|-------|-------|---------|-----------|
|  |                          |       |       |       | Holetta | Robgebeya |
| Physical properties                    |                          |       |       |       |         |           |
| Soil bulk density                      | g cm <sup>-3</sup>       |       |       |       | 1.02    | 0.98      |
| Soil water content                     | %                        |       |       |       | 16.5    | 15.4      |
| Clay                                   | %                        |       |       |       | 56.1    | 53.6      |
| Silt                                   | %                        |       |       |       | 28.6    | 31.5      |
| Sand                                   | %                        |       |       |       | 15.4    | 14.9      |
| <b>Chemical properties</b>             |                          |       |       |       |         |           |
| pH (H <sub>2</sub> O)                  |                          | 7.54  | 8.27  | 7.87  | 4.97    | 4.83      |
| Organic carbon (OC)                    | %                        | 27.8  | 51.6  | 32.7  | 2.32    | 1.44      |
| Total nitrogen (N)                     | %                        | 1.36  | 0.56  | 1.25  | 0.23    | 0.15      |
| Bray-2 phosphorus (P)                  | g kg <sup>-1</sup>       | 9.3   | 0.90  | 8.3   | 0.015   | 0.007     |
| Exchangeable potassium (K)             | cmol(+) kg <sup>-1</sup> | 3.62  | 7.17  | 4.86  | 0.98    | 0.71      |
| Exchangeable sodium (Na)               | cmol(+) kg <sup>-1</sup> | 1.15  | 0.59  | 0.78  | 0.41    | 0.37      |
| Exchangeable calcium (Ca)              | cmol(+) kg <sup>-1</sup> | 5.83  | 3.97  | 4.41  | 12.2    | 11.6      |
| Exchangeable magnesium (Mg)            | cmol(+) kg <sup>-1</sup> | 2.65  | 2.42  | 2.28  | 2.67    | 2.85      |
| Cation exchange capacity (CEC)         | cmol(+) kg <sup>-1</sup> | 13.3  | 14.2  | 12.3  | 16.2    | 15.5      |
| Ammonium nitrogen (NH <sub>4</sub> -N) | mg kg <sup>-1</sup>      | 147   | trace | 63    | 21.0    | 16.3      |
| Nitrate nitrogen (NO <sub>3</sub> -N)  | mg kg <sup>-1</sup>      | trace | 77.0  | trace | 100.3   | 88.7      |

**Table 6.1.** Physicochemical properties of compost (Com), acacia biochar, co-composted biocharcompost (COMBI) and pre-planting soil (0 - 20 cm depth) at the two sites.

The organic amendments were applied manually and evenly to the main plots, fourteen days before sowing, and thoroughly mixed in the upper 5 cm of soil. The plots were sown with six-row food barley (variety *HB-1307*) at the rate of 90 kg ha<sup>-1</sup> in ten rows 5 m long and 0.2 m apart. Sowing took place at the onset of rainfall at each site, this being on the 24<sup>th</sup> of June 2014 at Holetta and the 30<sup>th</sup> of June 2014 at Robgebeya. The preceding crop was potato at both sites. Phosphorus fertilizer was applied to all plots at planting, as triple superphosphate (TSP) at the recommended rate (20 kg P ha ha<sup>-1</sup>), in a band in the row. To minimize losses and increase efficiency, all the N fertilizer (urea) was applied along the row in two applications: half at planting and the other half 40 days after planting,

during the maximum growth period of the crop at full tillering stage, after the first weeding and during light rainfall to minimize loss of N to the atmosphere. Other recommended agronomic practices were applied during the crop growth. Harvesting took place at physiological maturity, on the 4<sup>th</sup> November 2014 at Holetta and on the 1<sup>st</sup> December 2014 at Robgebeya. The central six rows of the plots (5 m  $\times$  1.2 m) were harvested.

### 6.2.1. Sample analyses and calculation of nutrient use efficiency

Roughly 15 g each of the grain and straw samples were subsampled to determine the total N concentration and N isotope composition ( $\delta^{15}$ N). The subsamples were oven-dried at 70 °C for 72 h and, subsequently, ground using a Karl Kolb-RETSCH grinder (Scientific Technical Supplies, Germany) to pass through a 0.5 mm mesh. Total N and isotopic composition of grain and straw samples were determined using the ECS 4010 CHNSO Analyzer and a ThermoFinnigan DeltaVPLUS Continuous-Flow Isotope Ratio Mass Spectrometer (EA-IRMS), at the Advanced Analytical Centre of James Cook University, Cairns, Australia. Stable isotope compositions are reported as per mil (‰) deviations from the VPDB reference standard for  $\delta^{13}$ C, and from the international air standard for  $\delta^{15}$ N. Precisions on internal standards were better than ±0.2‰ for both isotope determinations. Grain protein concentration was calculated by multiplying grain N concentration by 5.83 (Mariotti et al., 2008). Grain N uptake (GNU) and straw N uptake (SNU), as kg ha<sup>-1</sup>, were calculated by multiplying total grain yield by grain N and straw N concentration, respectively. Total N uptake is the sum of GNU and SNU. Nitrogen harvest index (NHI) was computed as follows:

$$NHI(\%) = \frac{GNU}{TNU} \times 100 \tag{1}$$

Fertilizer N use efficiency is a complex term with many components. The following parameters were used to express and quantify the relative grain N uptake and N use efficiency of barley

Agronomic efficiency (AE), defined as grain production per unit of N applied, was computed as:

$$AE (kg kg^{-1}) = \frac{GY_{f} - GY_{u}}{N_{a}}$$
(2)

Where  $GY_f$  is the grain yield of the fertilized plot (kg),  $GY_u$  is the grain yield of the unfertilized plot (kg) for each replicate, and  $N_a$  is the quantity of N applied as organic amendments and N fertilizer (kg).

Apparent nitrogen recovery efficiency (ARE), also referred to as applied N uptake efficiency, is the ratio of the difference in N uptake between the fertilizer treated and non-treated control plot to the application rate. It was calculated as:

$$ARE (\%) = \frac{N_f - N_u}{N_a} \times 100$$
(3)

Where  $N_f$  is the N uptake (grain plus straw) of the fertilized plot (kg),  $N_u$  is the N uptake (grain plus straw) of the unfertilized plot (kg) for each replicate, and N<sub>a</sub> is the quantity of N applied (kg).

Physiological efficiency (PE), which denotes the ability of a plant to transform N acquired from fertilizer into grain yield, is defined as grain yield obtained per unit of nutrient uptake. It was computed as:

$$PE (kg kg^{-1}) = \frac{GY_{f} - GY_{u}}{N_{f} - N_{u}}$$
(4)

Where  $GY_f$  is the grain yield of the fertilized plot (kg),  $GY_u$  is the grain yield of the unfertilized plot (kg) for each replicate,  $N_f$  is the N uptake (grain plus straw) of the fertilized plot (kg),  $N_u$  is the N uptake (grain plus straw) of the unfertilized plot (kg) for each replicate.

### 6.2.1. Data analysis

The data were subjected to analysis of variance using the general linear model procedure (PROC GLM) of SAS statistical package version 9.1 (SAS Institute Cary, NC). The total variability for each trait was quantified using the model:

$$Y_{ijk} = \mu + R_i + A_j + R(A)_{ij} + N_k + AN_{(jk)} + \mathcal{C}_{ijk}$$
(5)

Where  $Y_{ijk}$  is the observed value,  $\mu$  is the grand mean,  $R_i$  is the effect of the *i*<sup>th</sup> replicate,  $A_j$  is the effect of the *j*<sup>th</sup> organic amendment,  $N_k$  is the effect of the *k*<sup>th</sup> nitrogen level, AN is the interaction effect, and  $R(A)_{ij}$  and  $e_{ijk}$  are the variations due to random error for the main and sub-plots, respectively. Significance of the A effect was tested against the  $R(A)_{ij}$  mean square, as an error term. All other effects were tested against the residual. Means for the main effects of organic amendments and N fertilizer levels (n = 5) were compared using the MEANS statement with the least significant difference (LSD) test at the 5% level. Single degree of freedom orthogonal contrasts was also performed to determine the nature of the crop response to the rates of applied N fertilizer. Means for the interactions were compared using the PDIFF STDERR option in the least squares means (LS MEANS) statement of the GLM procedure of SAS; in particular, specifying the R(A) as an appropriate error term for separating LS MEANS for the interaction of organic amendment and N fertilizer rate. Correlation analysis was performed among N uptake and N use efficiency parameters (n = 75), following the SAS CORR procedure (SAS Institute, 2008).

### 6.3. Results and discussions

### 6.3.1. Effect of organic amendments on N concentration and uptake of barley

Organic amendments significantly (p < 0.05 and p < 0.01) increased grain and straw N concentrations, grain and straw uptake, and significantly affected  $\delta^{15}N$  composition at both locations (with no significant difference among them), compared to the control (Table 6.2). At Holetta, mean grain N concentration was 1.53% in the control and 1.65% in Com + B, and at Robgebeya it was 1.40% in the control and 1.58% in Com + B. Similarly, straw N concentration ranged from 0.65% in the control to 0.81% in Com at Holetta and 0.45% in the control to 0.54% in Com at Robgebeya (Tables 6.3 and 6.4). The increase in grain and straw N concentration with organic amendments was similar to that resulting from N fertilizer addition at both sites. Similarly, Agegnehu et al. (2016a) reported that application of Com, and a compost + biochar mixture resulted in the same concentration of N in maize grain as when mineral fertilizer was applied. The grain and straw N concentrations were higher at Holetta than at Robgebeya, reflecting differences in pre-trial soil nutrient contents, particularly for mineral N and yield between the sites. Application of B, Com, Com + B, and COMBI (without mineral fertilizer) increased grain and straw N concentration by 6.5, 7.2, 7.8 and 7.2%, and 11, 25, 23 and 17%, respectively at Holetta, and by 2.1, 11, 13 and 3%, and 11, 20, 18 and 4.4%, respectively at Robgebeya, relative to the control. Barley grain protein was significantly ( $p \le 0.05$ ) improved by organic amendments at Robgebeya only (Table 6.4). Grain protein ranged from 8.9 to 9.6% at Holetta, and 8.2 to 9.2% at Robgebeya, with the highest being from Com + B at Holetta and from Com at Robgebeya. Nitrogen harvest index (NHI) did not differ significantly among organic amendments at either location (Table 6.2). NHI was the highest in the control (no organic amendment) at Holetta, and Com treatment at Robgebeya (Tables 6.3 and 6.4).

|                      |    |     | Hole | ta   |      |     | Robgebeya |      |      |          |  |  |
|----------------------|----|-----|------|------|------|-----|-----------|------|------|----------|--|--|
|                      |    |     |      |      | Root |     |           |      |      |          |  |  |
| Parameter            | OA | Ν   | OA×N | CV   | MSE  | OA  | Ν         | OA×N | CV   | Root MSE |  |  |
| Grain yield          | ** | *** | *    | 8.1  | 318  | **  | ***       | **   | 10.9 | 338      |  |  |
| Straw yield          | *  | *** | *    | 11.2 | 532  | **  | ***       | *    | 13.0 | 613      |  |  |
| Grain $\delta^{15}N$ | *  | **  | *    | 11.4 | 0.30 | **  | ***       | ***  | 14.5 | 0.14     |  |  |
| Straw $\delta^{15}N$ | ** | *** | ***  | 14.2 | 0.15 | *** | ***       | ***  | 15.3 | 0.04     |  |  |
| Grain N (%)          | NS | *** | NS   | 4.2  | 0.07 | *   | **        | NS   | 7.2  | 0.11     |  |  |
| Straw N (%)          | ** | *   | *    | 11.1 | 0.08 | *   | ***       | **   | 11.3 | 0.06     |  |  |
| Grain N-uptake       | *  | *** | *    | 9.5  | 6.1  | **  | ***       | *    | 13.7 | 6.38     |  |  |
| Straw N-uptake       | ** | *** | NS   | 15.4 | 5.6  | **  | ***       | *    | 18.0 | 4.27     |  |  |
| Total N-uptake       | *  | *** | *    | 9.0  | 9.1  | **  | ***       | **   | 9.5  | 6.67     |  |  |
| NHI (%)              | NS | NS  | NS   | 5.8  | 3.7  | NS  | NS        | *    | 7.8  | 5.19     |  |  |
| GPC (%)              | NS | *** | NS   | 4.1  | 0.41 | *   | **        | NS   | 7.2  | 0.62     |  |  |
| AE                   | *  | *** | ***  | 10.6 | 1.17 | **  | ***       | **   | 14.3 | 2.11     |  |  |
| ARE                  | *  | *** | **   | 13.2 | 5.10 | **  | ***       | *    | 13.8 | 5.19     |  |  |
| PE                   | NS | *   | **   | 11.8 | 3.34 | *   | NS        | *    | 11.3 | 4.20     |  |  |

**Table 6.2.** Significance of the effects of organic amendments (OA), nitrogen fertilizer rates (N) and their interaction (OA  $\times$  N) on plant and soil parameters at the two sites.

Significant at  ${}^{*}p < 0.05$ ,  ${}^{**}p < 0.01$ ,  ${}^{***}p < 0.001$ ; NS: Not significant. OA: Organic amendment; NHI: Nitrogen harvest index; GPC: Grain protein content; AE: Agronomic efficiency; ARE: Apparent nitrogen recovery efficiency; PE: Physiological efficiency; N: Nitrogen; CV: Coefficient of variation.

The  $\delta^{15}$ N composition of grain and straw were measured as a means of inferring relative uptake of N from the different sources, including organic amendments, N fertilizer, and soil. Grain and straw  $\delta^{15}$ N were the highest (+2.9 and +1.14‰, respectively) with Com + B at Holetta, and with Com (+1.2 and +0.4‰, respectively) at Robgebeya, with marked differences between the sites (Tables 6.3 and 6.4). Higher grain and straw  $\delta^{15}$ N values at Holetta than at Robgebeya may indicate differences in native soil fertility at the two sites, in conformity with the initial soil N content. Biochar, Com, Com + B, and COMBI increased grain  $\delta^{15}$ N by 1.2, 2.3, 11.5 and 3.9%, respectively at Holetta, and by 27, 82, 46 and 58% at Robgebeya, relative to the control. This implies greater utilization of organic-derived N (which has higher  $\delta^{15}$ N than control N) in compost, Com + B, and COMBI treatments than in the other treatments, in agreement with the findings of other studies (Agegnehu et al., 2016a; Montemurro

et al., 2006). Choi et al. (2002) also reported that the  $\delta^{15}N$  of leaves and grains of maize amended with composted pig manure were significantly higher than those with urea only.

Adequate availability of nitrogen is a crucial factor for optimal N uptake by plants. Higher N uptake by the crop implies that the organic-amended soil maintained a higher concentration of this nutrient in the soil solution. Possibly but could also be that the faster growing plants were a larger sink for N and depleted the pool more rapidly, causing greater mass flow of N. Grain and straw N uptake were 45.8-73.4 and 22.3-42.7 kg ha<sup>-1</sup>, respectively at Holetta, and 29.4 - 56.5 and 15.7 - 29.7 kg ha<sup>-1</sup> at Robgebeya (Tables 6.3 and 6.4). The highest grain and straw N uptake were obtained from Com + B at Holetta, and from Com and Com + B, respectively at Robgebeya. Biochar, Com, Com + B, and COMBI additions increased grain and straw N uptake by 37, 50, 59 and 52%, and 64, 82, 95 and 86%, respectively at Holetta, and by 62, 97, 93 and 52%, and 50, 69, 87 and 44% at Robgebeya, compared to the control without soil amendment (Tables 6.3 and 6.4). The effect of organic amendments on grain N uptake was greater at Robgebeya than at Holetta, and vice versa for straw N concentration. The reason for higher grain N content because of the addition of organic amendments could be due to initial lower soil N content at Robgebeya than at Holetta. In contrast, grain and straw C content and  $\delta^{13}$ C showed virtually no response to N fertilizer, organic amendment, or their interaction at either location (data not shown).

|                                 | Isotope N (‰)        |                      | N concentrat | ion (%) | N Uptake (kg ha <sup>-1</sup> ) |        | (%)  |         |
|---------------------------------|----------------------|----------------------|--------------|---------|---------------------------------|--------|------|---------|
| Treatments                      | Grain $\delta^{15}N$ | Straw $\delta^{15}N$ | Grain N      | Straw N | GNU                             | SNU    | NHI  | GPC     |
| Organic amendment               |                      |                      |              |         |                                 |        |      |         |
| Control                         | 2.57 b               | 0.87 c               | 1.53         | 0.65 c  | 45.8 b                          | 22.3 b | 67.4 | 8.94    |
| В                               | 2.60 b               | 1.02 b               | 1.63         | 0.72 b  | 62.9 ab                         | 36.1 a | 63.5 | 9.47    |
| Com                             | 2.66 ab              | 1.11 ab              | 1.64         | 0.81 a  | 69.3 a                          | 39.7 a | 63.6 | 9.54    |
| Com + B                         | 2.88 a               | 1.14 a               | 1.65         | 0.80 a  | 73.4 a                          | 42.7 a | 63.3 | 9.57    |
| COMBI                           | 2.66 ab              | 1.02 b               | 1.64         | 0.76 ab | 70.2 a                          | 40.6 a | 63.2 | 9.55    |
| LSD (0.05)                      | 0.27                 | 0.11                 | 0.15         | 0.06    | 22.1                            | 8.2    | 7.2  | 0.89    |
| Nitrogen (kg ha <sup>-1</sup> ) |                      |                      |              |         |                                 |        |      |         |
| 0                               | 2.89 a               | 1.15 b               | 1.51 d       | 0.68 b  | 49.6 d                          | 27.6 c | 62.2 | 8.79 d  |
| 23                              | 2.94 a               | 1.28 a               | 1.58 c       | 0.74 ab | 59.2 c                          | 33.8 b | 63.7 | 9.22 c  |
| 46                              | 2.64 b               | 1.01 c               | 1.63 bc      | 0.76 a  | 65.9 b                          | 41.0 a | 61.6 | 9.47 bc |
| 69                              | 2.55 bc              | 0.95 c               | 1.67 ab      | 0.77 a  | 71.7 a                          | 39.5 a | 64.6 | 9.73 ab |
| 92                              | 2.35 c               | 0.77 d               | 1.69 a       | 0.79 a  | 75.3 a                          | 39.3 a | 65.5 | 9.85 a  |
| LSD (0.05)                      | 0.22                 | 0.11                 | 0.05         | 0.06    | 4.5                             | 5.0    | 2.7  | 0.30    |
| N linear                        | ***                  | ***                  | ***          | **      | ***                             | ***    | ***  | ***     |
| N quadratic                     | ***                  | ***                  | NS           | NS      | NS                              | NS     | NS   | ***     |

Table 6.3. Table of means for main effects of organic amendments and N fertilizer on barley agronomic parameters at Holetta.

Within each column, means with different letters are significantly different at p < 0.05. Significant at p < 0.05, p < 0.01, p < 0.001, respectively; NS: Not significant. B: Biochar; Com: Compost; COMBI: Co-composted biochar-compost; GNU: Grain nitrogen uptake; SNU: Straw nutrient uptake; NHI: Nitrogen harvest index; GPC: Grain protein content; LSD: Least significant difference at 5% level of significance.

|                                 | Isotop               | e N (‰)              | N concentration (%) |         | N Uptake (kg h | a <sup>-1</sup> ) | %       |        |
|---------------------------------|----------------------|----------------------|---------------------|---------|----------------|-------------------|---------|--------|
| Treatments                      | Grain $\delta^{15}N$ | Straw $\delta^{15}N$ | Grain N             | Straw N | GNU            | SNU               | NHI     | GPC    |
| Organic amendments              |                      |                      |                     |         |                |                   |         |        |
| Control                         | 0.66 d               | 0.33 b               | 1.40 b              | 0.45 b  | 29.4 c         | 15.7 c            | 66.7    | 8.2 ab |
| В                               | 0.84 c               | 0.19 d               | 1.43 ab             | 0.50 ab | 47.4 ab        | 23.7 b            | 66.9    | 8.4 b  |
| Com                             | 1.20 a               | 0.38 a               | 1.54 ab             | 0.54 a  | 56.5 a         | 26.5 ab           | 67.5    | 9.0 ab |
| Com + B                         | 0.96 bc              | 0.24 c               | 1.58 a              | 0.53 a  | 55.8 a         | 29.7 a            | 65.2    | 9.2 a  |
| COMBI                           | 1.04 ab              | 0.27 c               | 1.44 ab             | 0.47 ab | 43.4 b         | 23.1 b            | 65.5    | 8.4 ab |
| LSD (0.05)                      | 0.17                 | 0.035                | 0.18                | 0.08    | 12.0           | 5.13              | 6.6     | 1.02   |
| Nitrogen (kg ha <sup>-1</sup> ) |                      |                      |                     |         |                |                   |         |        |
| 0                               | 1.52 a               | 0.35 a               | 1.34 c              | 0.42 c  | 27.0 d         | 13.1 c            | 65.7 ab | 7.8 c  |
| 23                              | 1.03 b               | 0.29 b               | 1.44 bc             | 0.50 b  | 40.9 c         | 22.5 b            | 65.8 ab | 8.4 b  |
| 46                              | 0.95 b               | 0.32 b               | 1.50 ab             | 0.56 a  | 51.9 b         | 28.4 a            | 64.6 b  | 8.8 ab |
| 69                              | 0.59 c               | 0.25 c               | 1.54 ab             | 0.50 b  | 55.5 ab        | 26.2 a            | 68.1 a  | 9.0 ab |
| 92                              | 0.63 c               | 0.21 d               | 1.57 a              | 0.51 b  | 57.3 a         | 28.3 a            | 67.5 ab | 9.1 a  |
| LSD (0.05)                      | 0.10                 | 0.032                | 0.08                | 0.04    | 4.8            | 3.1               | 3.3     | 0.46   |
| N linear                        | ***                  | ***                  | ***                 | NS      | ***            | ***               | NS      | ***    |
| N quadratic                     | ***                  | NS                   | NS                  | NS      | **             | **                | NS      | NS     |

Table 6.4. Table of means for main effects of organic amendments and N fertilizer on barley agronomic parameters at Robgebeya.

Within each column, means with different letters are significantly different at p < 0.05. Significant at p < 0.05, p < 0.01, p < 0.001, respectively; NS: Not significant. B: Biochar; Com: Compost; COMBI: Co-composted biochar-compost; NS: GNU: Grain nitrogen uptake; SNU: Straw nutrient uptake; NHI: Nitrogen harvest index; GPC: Grain protein content; LSD: Least significant difference at 5% level of significance.

#### 6.3.2. Effects of N fertilizer on N concentration and uptake of barley

The application of N fertilizer resulted in significant responses of N uptake at both sites. At Holetta, there were significant (p < 0.01) linear responses for grain and straw N concentration, grain and straw N uptake, and linear and quadratic (p < 0.001) responses for grain and straw  $\delta^{15}$ N and grain protein content. Grain and straw N concentrations were increased by 4.6-12% and 2.8-8.3%, respectively, at Holetta in response to the applied fertilizer N, relative to the control (Table 6.3). At Robgebeya, N fertilizer addition resulted in significant (p < 0.01) linear responses for grain N concentration, straw  $\delta^{15}$ N and grain protein content, and linear and quadratic (p < 0.001) responses for grain and straw N uptake and grain  $\delta^{15}$ N. Grain and straw N concentrations were increased by 5.8-12% and 6.5-20%, respectively at Robgebeya, compared with no N fertilizer application (Table 6.4). The higher grain and straw N concentrations at Holetta were related to higher initial soil N content or more likely the mineral N compared to Robgebeya. The apparent positive effect of N fertilizer application on N concentration in the barley grain is in accordance with other studies (Pettersson and Eckersten, 2007; Sinebo et al., 2004; Tarekegne and Tanner, 2001). Although the response to organic amendments was more pronounced than the response to N fertilizer for straw N concentration at both locations, the straw N concentration response to N fertilizer was greater at Robgebeya than at Holetta. Interestingly, the substantial improvement of straw N concentration, due to organic amendments and N fertilizer at both locations, led to a preferred and more palatable livestock feed than other cereal straws (Tables 6.3 and 6.4). The application of N fertilizer at the rates of 23, 46, 69 and 92 kg N ha<sup>-1</sup> increased mean grain and straw N content by 18, 32, 44 and 50%, and 21, 46, 43 and 39%, respectively at Holetta, and by 52, 93, 107 and 111%, and 77, 115, 100 and 115% at Robgebeya (Tables 6.4 and 6.5), relative to the control without N fertilizer.

Nitrogen is a primary constituent of the nucleotides and proteins essential for life. Nitrogen fertilizer application significantly ( $p \le 0.01$ ) improved barley grain protein content at both locations (Table 6.2). Grain protein ranged between 8.8 and 9.9% at Holetta, and between 7.8 and 9.1% at Robgebeya, in the control and highest N fertilizer rate (92 kg N ha<sup>-1</sup>). The increase in grain protein content was consistent with the increase in the N rate at both sites (Tables 6.3 and 6.4). Grain protein content is an important quality component of cereals (Xu et al., 2012a). Since food barley is a major source of protein in the highland areas of Ethiopia, high grain N concentration is an important quality attribute, in addition to high grain yield. The highest grain N concentrations attained (1.69% at Holetta and 1.58% at Robgebeya), equivalent to the respective grain protein contents of 9.9% and 9.1%, were achieved with the application of 92 kg N ha<sup>-1</sup>. Grain  $\delta^{15}$ N decreased significantly ( $p \le 0.05$ ) in

response to N fertilizer application at both locations (Tables 6.3 and 6.4), reflecting the low  $\delta^{15}$ N of fertilizer. The effect of N fertilizer on NHI was significant only at Robgebeya ( $p \le 0.05$ ). Mean NHI values were 62 - 66% for Holetta and 65 - 68% for Robgebeya, with the highest values being from 92 kg N ha<sup>-1</sup> at Holetta, and from 69 kg N ha<sup>-1</sup> at Robgebeya. Other studies also reported N harvest index values ranging from 79 - 82% (Shejbalová et al., 2014) and 53 - 64% (Montemurro et al., 2006) for barley, and 78-81% for bread wheat (Tarekegne and Tanner, 2001). High N harvest index is associated with efficient utilization of applied N (Fageria, 2014) as is discussed next.

# 6.3.3. Effect of organic amendment by N fertilizer interaction on yield and N use efficiency of barley

Organic amendment by N interaction was significant (p < 0.05) for grain and straw yields at both sites (Table 6.2). Straw was considered to be all the non-grain dry matter, which included rachis, glumes, awns, leaves and culms. Grain yield was considerably improved with the combined application of organic amendments and N fertilizer at both locations. Interestingly, the control yield at Holetta was substantially higher than the national average yield, but lower at Robgebeya where the soil is poorer, and yield more N-limited, than at Holetta. The highest grain yields resulted from application of Com + B and 69 kg N ha<sup>-1</sup> at Holetta, and Com + 92 kg N ha<sup>-1</sup> at Robgebeya, with corresponding yield increments of 60% and 54%, compared to the highest N rate (92 kg ha<sup>-1</sup>) only, without organic amendment (Table 6.5). Previous studies have also shown that applications of biochar, compost, or their combination, together with N fertilizer, markedly increased barley grain yield in temperate (Gathorne-Hardy et al., 2009) and tropical (Agegnehu et al., 2016b) agroecosystems. Gathorne-Hardy (2012) also found that the combined application of biochar and nitrogen fertilizer significantly increased crop yields and nitrogen use efficiency. Grain protein content was higher for the combined application of organic amendments and N fertilizer than either on their own, but the interaction was not significant.

|         |        | Holetta site | Robgebeya site |             |             |  |  |  |
|---------|--------|--------------|----------------|-------------|-------------|--|--|--|
| OA      | N rate | Grain yield  | Straw yield    | Grain yield | Straw yield |  |  |  |
| Control | 0      | 2511±16.5    | 2660±286.4     | 1261±54.6   | 1747±279.9  |  |  |  |
| Control | 23     | 2654±47.7    | 2886±142.8     | 1560±121    | 2033±314.7  |  |  |  |
| Control | 46     | 3008±232     | 3668±423.2     | 2182±59.5   | 3382±552.8  |  |  |  |
| Control | 69     | 3367±187.7   | 3980±674.1     | 2236±187.4  | 3251±506.3  |  |  |  |
| Control | 92     | 3368±234.9   | 3776±444.9     | 2982±289.6  | 4839±601.1  |  |  |  |
| В       | 0      | 3266±132.7   | 4162±361.5     | 2253±191.5  | 3309±155.9  |  |  |  |
| В       | 23     | 3837±225.7   | 4398±675.1     | 3315±191.6  | 4967±392.5  |  |  |  |
| В       | 46     | 3716±169.7   | 5503±573.9     | 3612±154.7  | 6207±568.5  |  |  |  |
| В       | 69     | 4247±278.6   | 5632±483.9     | 3782±383.8  | 5316±170.7  |  |  |  |
| В       | 92     | 4270±102.3   | 5308±447.9     | 3935±78.4   | 5079±186.6  |  |  |  |
| Com     | 0      | 3450±102.3   | 4402±88.7      | 2124±339.6  | 3202±262.3  |  |  |  |
| Com     | 23     | 3823±69.5    | 4877±111.1     | 3321±150    | 4680±191.2  |  |  |  |
| Com     | 46     | 4139±73.0    | 4891±62.8      | 4235±374.4  | 5681±432.4  |  |  |  |
| Com     | 69     | 4736±183.5   | 5166±108.5     | 3874±672.3  | 5423±290.1  |  |  |  |
| Com     | 92     | 5134±423.8   | 5057±149.4     | 4598±186.1  | 5492±507.3  |  |  |  |
| Com + B | 0      | 3693±264     | 4177±812.8     | 2272±183.1  | 3617±402.2  |  |  |  |
| Com + B | 23     | 4020±362.4   | 4790±1123      | 3237±243.3  | 5619±166.7  |  |  |  |
| Com + B | 46     | 4498±363.7   | 6772±309.6     | 3981±69.5   | 5728±961.5  |  |  |  |
| Com + B | 69     | 5381±278.5   | 5837±623.3     | 4123±133.2  | 6145±654.9  |  |  |  |
| Com + B | 92     | 4534±497.4   | 4936±681.9     | 3750±253.7  | 6822±405.6  |  |  |  |
| COMBI   | 0      | 3427±541.7   | 4288±591.6     | 2082±172.2  | 3805±194.4  |  |  |  |
| COMBI   | 23     | 4283±305.7   | 5509±323.3     | 2842±258.7  | 4856±539.2  |  |  |  |
| COMBI   | 46     | 4708±485     | 5898±530.8     | 3145±239.3  | 5145±918.2  |  |  |  |
| COMBI   | 69     | 4488±637     | 5716±227.8     | 3863±51.2   | 6015±284.7  |  |  |  |
| COMBI   | 92     | 4263±618.4   | 5171±432       | 3045±265.1  | 5543±335.7  |  |  |  |

**Table 6.5.** Interaction effects of organic amendments and nitrogen fertilizer on grain and straw yield (kg ha<sup>-1</sup>) of barley.

OA: Organic amendment; Control (0 kg ha<sup>-1</sup>); B: Biochar (56 kg N ha<sup>-1</sup>); Com: Compost (125 kg N ha<sup>-1</sup>); Com + B (136 kg N ha<sup>-1</sup>); COMBI: Co-composted biochar-compost (115 kg N ha<sup>-1</sup>).

The N uptake parameters responded positively to combined application of organic amendments and N fertilizer at both locations, as a result of the increased plant N content and the significant

enhancement of grain and straw yields. In the current study, increases in grain and straw N uptake were more pronounced when organic amendments and N fertilizer were both applied, in comparison to one or the other (Figures 6.2 and 6.3). Total N uptake responded significantly ( $p \le 0.01$ ) to the interaction of organic amendments and fertilizer N at both locations (Table 6.2). The highest mean total N uptake values were achieved with the combined application of organic amendments and N fertilizer at both sites, although there were differences in soil fertility between the sites. Mean values of total N uptake increased from 51 kg N ha<sup>-1</sup> in the control to 138 kg N ha<sup>-1</sup> in Com + B and 92 kg N ha<sup>-1</sup> at Holetta, and 22 kg N ha<sup>-1</sup> in the control to 101 kg N ha<sup>-1</sup> in Com + 92 kg N ha<sup>-1</sup> at Robgebeya, as a result of higher grain and straw N concentrations (Figures 6.2 and 6.3). The difference between sites was consistent with higher grain and straw yields at Holetta than at Robgebeya (Table 6.5). The total N uptake values in this study are comparable with the results of previous studies, which reported values ranging from 54-105 kg N ha<sup>-1</sup> (Shejbalová et al., 2014) and 97 - 160 kg N ha<sup>-1</sup> (Montemurro et al., 2006) for barley, under organic and inorganic fertilization. Studies with wheat on various soils with various N applications rates reported total N uptake values of 23-64 kg N ha<sup>-1</sup> on Vertisol and 47 - 103 kg N ha<sup>-1</sup> on Nitisol (Tarekegne and Tanner, 2001), 98 - 112 kg N ha<sup>-1</sup> (Ruisi et al., 2015) and 101-162 kg N ha<sup>-1</sup> (Guarda et al., 2004).

The agronomic and apparent recovery efficiency of N responded significantly ( $p \le 0.01$ ) to the organic amendment by N fertilizer interaction at both locations (Table 6.2). Based on the total amount of N applied, mean agronomic efficiency was 6.2 - 16.8 kg grain kg<sup>-1</sup> N applied at Holetta, and 6.9-26 kg grain kg<sup>-1</sup> N applied at Robgebeya (higher at the low fertility site), while the mean apparent recovery efficiencies of N were 27 - 51% and 18 - 59%, respectively (Figures 6.2 and 6.3). The highest agronomic and recovery efficiency of N were obtained from the application of B + 23 kg N ha<sup>-1</sup> at Holetta, and from B + 23 and B + 46 kg N ha<sup>-1</sup> at Robgebeya, with agronomic efficiency increment over the highest N fertilizer rate only (92 kg N ha<sup>-1</sup>) of  $\sim$ 81% and 28% respectively, and N recovery efficiency of 39 and 18%, respectively. In this study, effects of the treatments on N use efficiency of barley were consistent with the findings of previous studies (Raun et al., 2002; Ruisi et al., 2015). According to Shejbalová et al. (2014), mean agronomic efficiency of 22 - 29 kg kg<sup>-1</sup> and N recovery efficiency of 46 -60% were obtained for barley grown in organic and inorganic amended Cambisol. For wheat, Guarda et al. (2004) reported mean agronomic efficiency of 2 - 18 kg kg<sup>-1</sup> N and N recovery efficiency of 14 - 56%, and (Tarekegne and Tanner, 2001) reported agronomic efficiency of 12.6 - 29 and 15 - 26 kg yield increase kg<sup>-1</sup> applied N and recovery efficiency of 32 - 5 6% and 42 -63% on a Vertisol and Nitisol, respectively. Similarly, a 59% and 37% recovery efficiency of applied N in wheat and barley, respectively, were reported in response to slurry and mineral N interaction

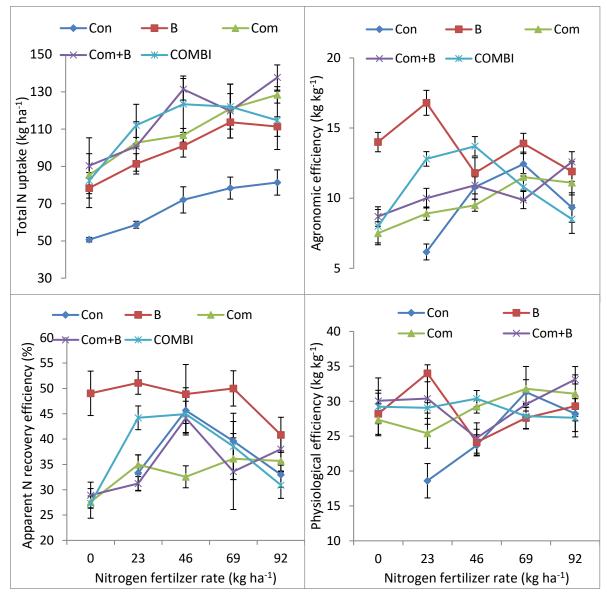
(Sieling et al., 1998). Although grain, straw and N yields of barley were the highest at Holetta, the response to N fertilizer (i.e., slope of both agronomic and recovery efficiency of N vs application rate) was greater at Robgebeya than at Holetta (Figures 6.2 and 6.3). This is due to the higher yield obtained in the control at Holetta than at Robgebeya. At the lowest and highest N rates (0 and 92 kg N ha<sup>-1</sup>) there was a negative effect of Com, Com + B, and COMBI on the agronomic and recovery efficiency of N at both locations (Figures 6.2 and 6.3). However, at intermediate rates of N application (23 and 46 kg N ha<sup>-1</sup>), the organic amendments had a positive effect.

Analysis of the interaction revealed rate-dependent effects on the agronomic and apparent recovery efficiency of N; both parameters tended to decrease with higher N rates (organic plus inorganic sources) at both locations. An increase in the rate of N application usually decreases the agronomic and recovery efficiency of N in cereals (Limon-Ortega et al., 2000; Sieling et al., 1998). Both low and high rates of crop recovery efficiency of applied organic and inorganic fertilizer nutrients have been previously reported for rice (Baligar et al., 2001), maize and barley (Montemurro et al., 2006). These trends, however, vary considerably with the yield potential of the crop variety (Dawson et al., 2008; Guarda et al., 2004; Sinebo et al., 2004), cropping history of the site, moisture availability, nutrient status, and nutrient retention capacity of the soil (Baligar et al., 2001; Fageria and Baligar, 2005). Shi et al. (2012) found that increasing N rate not only resulted in low N utilization efficiency and grain yield, but also resulted in high leaching loss of NO<sub>3</sub>-N. Baligar et al. (2001) also reported that increasing applications of N from 0 to 210 kg ha<sup>-1</sup> reduced overall N use efficiency in lowland rice, with the N recovery efficiency of 32% at 210 kg N ha<sup>-1</sup>. Such low N recoveries may be related to N losses from soil via denitrification, ammonia volatilization and  $NO_3^-$  leaching. In the current study, the enhancement of the agronomic and recovery efficiency of N by applying organic amendments and fertilizer N together emphasizes the importance of balanced crop nutrition. To optimize biological and economic efficiency of applied fertilizer, adequate supply of N and other nutrients must be ensured.

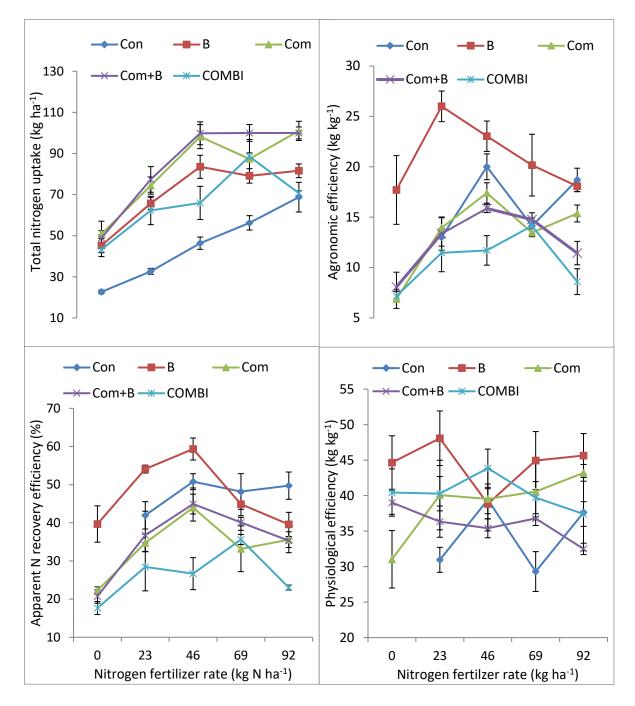
Physiological efficiency represents the fraction of plant acquired N that is converted to grain yield. Physiological efficiency was significantly ( $p \le 0.05$ ) affected by the interaction between organic amendment and N fertilizer application at both locations (Table 6.2). It was 19 - 33 at Holetta and 29 - 48 kg grain kg<sup>-1</sup> N uptake at Robgebeya (Figures 6.2 and 6.3). Despite similar effects of all organic amendments on physiological efficiency of N, the maximum values were with biochar. Biochar + 23 kg N ha<sup>-1</sup> resulted in the highest physiological efficiency of 33 kg kg<sup>-1</sup> N at Holetta and 48 kg kg<sup>-1</sup> N at Robgebeya. The findings of Paneque et al. (2016) have shown that the shortage of rain during the last weeks of sunflower culture caused a water shortage and greater loss of the photosystem II (QY) loss in plants not amended by biochar. Better growth of amended plants during the drought period correlated with higher reduction of stomatal conductance, indicating that the greater water use efficiency is at the origin of the better crop performance of biochar-amended plants. The increase in physiological efficiency was not consistently related to N fertilizer rate at either location (Figures 6.2 and 6.3). Nutrient use efficiency is a function of the capacity of a soil to supply adequate levels of nutrients and the ability of plants to acquire them. Interaction of plants with environmental factors, such as solar radiation, rainfall, temperature, plant response to diseases, insects, allelopathy, and root microbes all have a great influence on N use efficiency in plants (Baligar et al., 2001; Xu et al., 2012a). The partial factor productivity of applied N, grain yield produced per unit of applied N, was 20 - 115 and 15 - 68 kg grain kg<sup>-1</sup> N at Holetta and Robgebeya, respectively (data not shown). Zingore et al. (2007) reported partial factor productivity ranging from more than 50 kg grain kg<sup>-1</sup> applied N for maize on more fertile fields to less than 5 kg grain kg<sup>-1</sup> N in degraded fields of African smallholder farms.

The N use efficiency of a cereal crop is a function of its ability to capture N from the available supply, and the efficiency with which N is utilized to make grain (Bingham et al., 2012; Huggins and Pan, 2003). For most annual crops, N-uptake from soil at optimum fertilizer rates occurs for only 8 - 12 weeks, and the discrepancy between N availability with crop need is probably the greatest contributor to excess N losses (Robertson and Vitousek, 2009). Nitrogen in applied organic amendments may be prone to immobilization and, as a result, plants usually recover low amounts of organic amendment N in the short-term. Burger and Jackson (2003) reported high rates of microbial NO<sub>3</sub><sup>-</sup> immobilization relative to the gross nitrification rates in an organic cropping system. On the other hand, N is susceptible to losses through leaching or denitrification, so rates of recovery are low under conditions of high rainfall and/or impeded drainage (Tarekegne and Tanner, 2001; Xu et al., 2012a). In this study, the relatively low N use efficiency at the high yielding site (Holetta) is presumably due to the reasonably high N uptake and high grain yield in the control, rather than the low N uptake and yield at the high N rate. Under these conditions, more N may be lost by NH<sub>3</sub> volatilization, leaching, and denitrification. Eagle et al. (2000) indicated that additional available soil N from applied N resulted in increased N uptake without a corresponding increase in rice grain yield, which decreased N use efficiency. At both of our locations, the physiological efficiency of N by barley generally decreased with increasing N rates from mineral and organic sources, indicating that the decrease in N use efficiency may have been due to the increased soil N supply, in agreement with other findings (Baligar et al., 2001; Eagle et al., 2000). Nutrient use efficiency is generally greatest under conditions of high yield response to an applied nutrient as demonstrated by the results of this study and other studies (Sieling et al., 1998).

In our study, the relative contributions of the two components of N use efficiency varied with N supply, consistent with other studies (Huggins and Pan, 2003; Le Gouis et al., 2000). Application of organic amendments improved fertilizer use efficiency, especially when combined with mineral fertilizer, which is in agreement with the results of previous studies (De Boer, 2008; Shejbalová et al., 2014; Steiner et al., 2008). Biochar and N fertilizer had essentially positive and additive effects at both sites, whereas the positive effect of organic amendments containing compost was greatest at moderate levels of N application (23 - 69 kg ha<sup>-1</sup>) at both sites. According to Steiner et al. (2008), the total N recovery in sorghum grain was significantly higher with compost (16.5%), biochar (18.1%), and compost + biochar treatments (17.4%) than with mineral-fertilized plots (10.9%). Overall, inclusion of organic soil amendments in the production system improves fertilizer use efficiency and ensures good yield and sustainability in the long-term.



**Figure 6.2.** Nitrogen use efficiencies as influenced by organic amendments and N fertilizer at Holetta. Note: Con: Control; B: Biochar; COMBI: Co-composted biochar-compost. Error bars represent ± 1 SE.



**Figure 6.3.** Nitrogen use efficiencies as influenced by organic amendments and N fertilizer at Robgebeya. Note: Con: control; B: biochar; COMBI: co-composted biochar-compost. Error bars represent  $\pm 1$  SE.

The relationship between yield and nutrient use efficiency differed between the two sites. The most important feature of the various components of N use efficiency is the N requirement for producing the highest potential yield, which is the combination of N uptake and physiological efficiency (Chien

et al., 2009; Xu et al., 2012a). In agreement with the study of Bingham et al. (2012), the correlation of grain yield with physiological efficiency was greater than with apparent recovery efficiency of applied N at Holetta, implying greater importance of physiological efficiency in enhancing yield, possibly because of greater N fertility (Table 6.6). In contrast, the correlation of grain yield with apparent recovery efficiency of applied N was stronger than with physiological efficiency of N at Robgebeva, signifying the greater importance of apparent N recovery efficiency to maximizing grain yield, i.e. actual N supply is important (Table 6.6). Sinebo et al. (2004) also found that grain yield was more strongly correlated with N recovery efficiency than with physiological efficiency. The presence of a stronger association of grain or straw yield with the respective grain or straw N uptake, than the correlation of grain or N concentration with the N uptakes, suggests that the relationships were greatly dictated by the variability in dry matter yields, relative to by the variability in plant N concentration. In cereal crops, grain protein content and grain yield generally show a negative relationship (Bogard et al., 2010). High grain yields are known to dilute the quantity of protein in the grain. Relatively lower protein content (a low seed N concentration) represents higher N physiological use efficiency, as the N concentration in the vegetative organs at later developmental stages is much lower than it is in the seeds, and most of the N taken up by cereals is allocated to grain (Xu et al., 2012a). Thus, as most of the N in cereal crops is transported into grain, decreasing the content of nonessential grain protein components without affecting yield could be an alternative strategy for improving N use efficiency. In general, N use efficiency can be increased by improving the grain yield per unit of N application.

|                      | Grain              | Straw        | Grain               | Straw               | Grain               | Straw               |                     |                     |                     |                     |                     |                     |                     |
|----------------------|--------------------|--------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
|                      | yield              | yield        | $\delta^{15}N$      | $\delta^{15}N$      | Ν                   | Ν                   | GNU                 | SNU                 | TNU                 | GPC                 | AE                  | ARE                 | PE                  |
| Grain yield          |                    | 0.75***      | -0.18 <sup>ns</sup> | -0.12 <sup>ns</sup> | 0.68**              | 0.44*               | 0.98***             | 0.71***             | 0.94***             | 0.68**              | 0.58**              | 0.51**              | 0.57**              |
| Straw yield          | 0.80**             |              | -0.11 <sup>ns</sup> | -0.05 <sup>ns</sup> | 0.61**              | 0.41*               | 0.75***             | 0.91***             | 0.88***             | 0.60**              | 0.56**              | 0.55**              | 0.34*               |
| Grain $\delta^{15}N$ | -0.42*             | -0.39*       |                     | 0.35*               | -0.16 <sup>ns</sup> | -0.03 <sup>ns</sup> | -0.18 <sup>ns</sup> | -0.07 <sup>ns</sup> | -0.15 <sup>ns</sup> | -0.18 <sup>ns</sup> | -0.07 <sup>ns</sup> | -0.05 <sup>ns</sup> | -0.08 <sup>ns</sup> |
| Straw $\delta^{15}N$ | -0.51**            | -0.51**      | 0.34*               |                     | -0.24*              | 0.06 <sup>ns</sup>  | -0.16 <sup>ns</sup> | -0.02 <sup>ns</sup> | -0.12 <sup>ns</sup> | -0.24*              | 0.02 <sup>ns</sup>  | -0.01 <sup>ns</sup> | 0.16 <sup>ns</sup>  |
| Grain N              | $0.40^{*}$         | 0.38*        | 0.04 <sup>ns</sup>  | -0.14 <sup>ns</sup> |                     | 0.30*               | 0.81***             | 0.56**              | 0.77***             | 0.99***             | 0.41**              | 0.45**              | 0.26*               |
| Straw N              | 0.16 <sup>ns</sup> | 0.70***      | 0.05 <sup>ns</sup>  | 0.06 <sup>ns</sup>  | 0.35*               |                     | 0.42*               | $0.70^{***}$        | 0.56**              | 0.30*               | 0.26*               | 0.25*               | 0.41*               |
| GNU                  | 0.96***            | $0.78^{***}$ | -0.38*              | -0.54**             | 0.68**              | 0.35*               |                     | 0.73**              | 0.95***             | 0.81***             | 0.61**              | 0.50**              | 0.50**              |
| STU                  | 0.74***            | $0.88^{***}$ | -0.33*              | -0.45*              | $0.48^{*}$          | 0.59**              | 0.76***             |                     | 0.89***             | 0.55**              | 0.46**              | 0.49**              | 0.35*               |
| TNU                  | 0.94***            | 0.86***      | -0.39*              | -0.54**             | 0.64**              | 0.45*               | 0.97***             | 0.89***             |                     | 0.77***             | 0.59**              | 0.53**              | 0.47**              |
| GPC                  | $0.46^{*}$         | 0.41*        | -0.15 <sup>ns</sup> | -0.22 <sup>ns</sup> | 0.99***             | 0.35*               | 0.68**              | $0.48^{*}$          | 0.64**              |                     | 0.41*               | 0.45*               | 0.26*               |
| AE                   | 0.52**             | 0.39*        | -0.42*              | -0.35*              | 0.05 <sup>ns</sup>  | 0.32*               | 0.42*               | 0.41*               | $0.44^{*}$          | 0.05 <sup>ns</sup>  |                     | 0.85***             | 0.72***             |
| ARE                  | 0.53**             | 0.38*        | -0.33*              | -0.17 <sup>ns</sup> | $0.28^{*}$          | 0.53**              | 0.41*               | 0.49**              | 0.45*               | 0.29*               | 0.88***             |                     | 0.57**              |
| NUtE                 | 0.44*              | 0.37*        | -0.16 <sup>ns</sup> | -0.56**             | -0.09 <sup>ns</sup> | -0.05 <sup>ns</sup> | 0.36*               | 0.24*               | 0.35*               | -0.09 <sup>ns</sup> | 0.61**              | 0.35*               |                     |

**Table 6.6.** Coefficients of correlation for N uptake and N use efficiency of barley grown with five organic amendments and five N levels at Holetta (upper right) and at Robgebeya (lower left).

Significant at  ${}^{*}p < 0.05$ ,  ${}^{**}p < 0.01$ ,  ${}^{***}p < 0.001$ ; ns: not significant. GNU: grain nitrogen uptake; STU: straw nitrogen uptake; TNU: total nitrogen uptake; GPC: grain protein content; AE: agronomic efficiency; ARE: apparent N recovery efficiency; PE: Physiological efficiency.

#### 6.4. Conclusions

Our experiments with barley showed high uptake, agronomic, and apparent recovery efficiency of applied N with application of B, Com, Com + B, or COMBI (at similar N rates). The highest yields were achieved using organic amendments in combination with moderate rates of N fertilizer. All N uptake parameters responded significantly to applied organic amendments at both locations, reflecting the positive effects of organic amendments on barley grain and straw yields, and the enhancement of grain and straw N contents. Most N-use efficiency components were greater when organic amendments and N fertilizer were applied together than when they were applied alone. The positive effects of organic amendments on N use efficiency were greater at the site with lower soil fertility than at the site with higher fertility. Any of the organic-inorganic amendment options identified in this study can be recommended to improve N use efficiency and yield of barley. However, although B, Com, Com + B, and COMBI were all good soil fertility ameliorants, the best treatments in terms of N use efficiency were B, Com, and Com + B, together with 23 kg N ha<sup>-1</sup> at Holetta, and 46 kg N ha<sup>-1</sup> at Robgebeya. Thus, when used together with organic amendments, less N fertilizer is required to achieve a given yield than when N is applied alone. This interaction may increase profitability and enhance long-term sustainability of the production system, while simultaneously mitigating environmental pollution.

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# Chapter 7

# **General Discussions and Conclusions**

### 7.1. Relative performance of biochar, compost and their mixture

The biochars used in this study were all from woody lingo-cellulosic feedstock and were similar to each other, in that they were all comparatively high carbon. However, willow biochar had relatively higher nutrients than acacia biochar. The major difference between these biochars was their density, which varied from 0.19 g cm<sup>-3</sup> for willow, and 0.83 g cm<sup>-3</sup> for acacia biochar. Bulk density affects porosity and surface area - a characteristic of importance to soil biota, nutrient, and water retention. These biochars have shown significant carbon sequestration potential as a soil amendment as all were produced at a high temperature, enough to convert a large proportion of the original carbon to stable polycyclic aromatic carbon (McBeath et al., 2015). The long-term carbon sequestration potential of these biochars ranged from 41-67% of the mass of biochar added to the soil. Addition of biochar to the composts, to make the COMBI amendment, significantly increased the pH of the product, which is of potential benefit to ameliorate acid soils. Biochar also increased the rate of composting, as measured by the C:N ratio, which is consistent with previous studies (Zhang et al., 2014; Zhang and Sun, 2014). Curbing the period of composting required for maturation through the addition of biochar may be a beneficial possibility for increasing output and productivity for composting operations. Thus, there are potential advantages in co-composting biochar with organic waste in terms of higher pH, faster composting, and possibly increased nitrogen retention.

Substantial improvements were observed in soil properties and crop yield associated with the application of organic amendments compared with the fertilizer treatment alone. However, a clear outcome in this study was that none of the organic amendments systematically outperformed any other. For example, all organic amendments significantly increased maize total biomass over the fertilizer treatment, but significant differences were not observed among them. In the same trial, biochar-containing compost resulted in significantly higher soil water content, compared to both the biochar only and the fertilizer treatment. In contrast, for the peanut trial, the COMBI treatment led to significantly higher NO<sub>3</sub>-N levels in the soil than all other organic amendments, but the biochar only treatment resulted in significantly higher soil water content and peanut nodule dry weights per plant than the other organic amendments. However, all organic amendments performed similarly in peanut pod and seed yields.

The results suggested that the use of COMBI (co-composted biochar-compost) has no immediate advantage, at least in the first year of application, in terms of either soil fertility or crop yield, over compost only, or over mixing compost and biochar together during application. The use of biochar has distinct advantages in terms of carbon sequestration, and the use of compost in soil health and crop yield. In general, this study has demonstrated that the additional application of organic amendments with conventional fertilizer, gave improvements in soil fertility and crop performance compared to fertilizer alone. Previous studies have shown that the impact of the combined use of compost and biochar on soils and crop yields can be significant in some cases (Lashari et al., 2015; Luo et al., 2016), and negligible in others (Schmidt et al., 2014). The effect of compost, biochar, and mixes of the two, appears to be dependent on interactions between the amendments, soil and crop types. Recent work has suggested that a minimum proportion of biochar may be required to be present in the compost to obtain the synergistic benefits of both (Schulz et al., 2014). Although Schulz et al. (2014) found significant increases in crop performance with co-composted mixtures of biochar and compost with up to 50% biochar, a minimum 3 t ha<sup>-1</sup> of biochar, regardless of compost application rate, was required to induce an effect from the biochar, additional to that derived from the compost. In this study, the biochar application rate with the compost (both COMBI and mixed on site) was 2.5 t ha<sup>-1</sup>.

#### 7.2. Impact of the amendments on soil properties

The results of the study indicate that there are no systematic differences in soil response between the different organic amendments, but differences were observed between the organic amendments as a group and the fertilizer treatment alone. The soil carbon stocks to 30 cm, prior to initiation of the trails, varied widely among the trails. The red soils of the Atherton Tablelands in North Queensland, Australia planted to maize and peanut had lower soil carbon stocks of 14.6 - 37.6 t ha<sup>-1</sup>, while the Nitisols in Ethiopia planted to barley had soil carbon stocks of 28.8-46.4 t ha<sup>-1</sup>. The impacts of the organic treatments on SOC were variable across the trials and over time. At trial mid-points, soil carbon contents and stocks were significantly higher in all organic treatments compared to fertilizer treatments for both Ferralsols and Nitisols, but significant differences were not observed among them. The biochar C addition ranged between 52 - 92% for the biochars used in the current study. According to McBeath et al. (2015), the minimum long-term carbon sequestration potential of the biochars averaged 0.54 t carbon per ton of biochar added (0.41 - 0.67 t), with this component representing the minimum real long-term carbon sequestration potential of the biochar.

The decomposition of organic matter in soils is dependent on temperature, moisture, nitrogen, and soil type (clay content), amongst other things (Conant et al., 2011; Krull et al., 2004; Wang et al., 2014). For the relatively hot and seasonally wet conditions pertaining for these trials it is reasonable to assume that, conservatively, 10% of the compost (with an average 140 kg C in the wet compost) added to the soil was still present in the soil at the end of the trials. Consequently, this figure would reduce further at a slower rate beyond the end of the trials. Combining this figure, with the carbon added as biochar, where appropriate, using the average of 67% of the biochar mass as carbon yields, the following potential carbon stock increases for the organic amendments over the fertilizer treatment only. The amount of carbon in the soil at the trial end period can be calculated based on the application rates used in this study. The carbon contents of biochar, compost, biochar + compost mixture, and co-composted biochar-compost (COMBI) were 6.7, 0.35, 2.0 and 2.0 t C ha<sup>-1</sup>, respectively, compared to the fertilizer treatment alone at the end of the trials. The long-term carbon sequestration potential of the single application of the organic amendments, at the rates used in this study, will be sequestered in the longer-term were 5.4, 0.035, 1.4, and 1.4 t C ha<sup>-1</sup> for biochar, compost, biochar + compost, and COMBI, respectively, assuming an average 0.54 t C per ton biochar and 1% of carbon derived from compost.

The values given above for carbon sequestration potential are indicative only and vary depending on the type of compost and biochar used, as well as interactions between the organic amendments and the soil. The figures for biochar carbon sequestration are, however, reasonably secure, and the figures for compost, while conjectural, are plausible. Longer-term stabilization of compost derived carbon is likely to be linked to clay mineral content (Lehmann et al., 2007; Trigalet et al., 2014) and, in this regard, the red soils of the Atherton Tablelands have the most potential for building soil carbon stocks. It should also be noted that significant increases in biomass occurred in trials and the un-harvested component of this biomass was incorporated into the soil. For example, in maize there was a 12 - 27% increase in non-cob biomass left as stover on the field after harvest. A sustained increased in stover left on the field after harvest could increase the equilibrium carbon stocks of the soil over time.

Soil water content exhibited a significant response to most organic amendments in most trials. All organic amendments retained significantly more soil moisture compared to fertilizer alone. For example, Sorrenti and Toselli (2016) have demonstrated that the combined application of biochar and compost amendments resulted in the highest soil WHC, even significantly greater than compost alone, indicating an additive effect. This synergism suggests that mixing biochar with compost represents a strategy to enhance the SWC. Overall, this study provides strong evidence that even the single application of the organic amendments used here will provide improved water retention over a

cropping cycle, noting that the magnitude is likely to be dependent on soil type. This result is significant given the importance of improved water use efficiency to agriculture (Castellini et al., 2015; Grafton et al., 2013) in the face of climate change, as well as future increases in utilization and greater climate variability (Steffen et al., 2011). Further, this result is particularly significant in areas where crops are subject to seasonally dry conditions and any increase in the duration or intensity of soil moisture deficiency is likely to have a detrimental effect on crop performance.

The addition of organic amendments improved most soil characteristics over the pre-plant soil conditions across all trials. Thus, pH, SOC, CEC, extractable cations, Colwell P, and nitrogen (total N, NH<sub>4</sub>-N and NO<sub>3</sub>-N) were increased, and bulk density decreased in the mid-point soil results, compared to the pre-plant soil results for most amendments in most trials, with some exceptions. Organic amendments significantly reduced leaching of NO<sub>3</sub><sup>-</sup>-N and basic cations on Ferallsols. For example, Haider et al. (2017) reported that soil NO<sub>3</sub><sup>-</sup>-N concentrations in topsoil (0 – 15 cm) remained significantly higher, and increased with increasing rate of biochar addition, throughout the study period. Soil pH was significantly improved by the organic amendments on Nitisols in Ethiopia, but not on Ferralsols in Australia in the maize and peanut trials.

While there was a tendency for biochar and biochar and compost mixture soil amendments to have a pH higher than the fertilizer treatment, there were no significant differences in maize and peanut trials, except for barley where organic amendments resulted in significantly higher pH values after harvesting. The liming effect of biochar was the highest on Nitisols, where the pH of this soil was suboptimal for the production of various crops. Studies indicated that the exchangeable base cations, effective cation exchange capacity, and base saturation have been increased, owing to the addition of biochar; while soil exchangeable Al and exchangeable acidity have significantly been decreased (Xu et al., 2016; Yuan and Xu, 2011). Organic amended soils had significantly higher CEC at trial end points. The exchangeable cations showed significant increases at the trial end in all trials, in spite of differences between trials. As a result, exchangeable Ca, Mg and/or K, as well as NH<sub>4</sub>-N and NO<sub>3</sub>-N, were significantly increased in organic amended soils of all trials. Colwell P was significantly increased in organic treatments in maize and positive effects on soil biophysical and chemical properties (Deenik and Cooney, 2016; Sorrenti and Toselli, 2016).

In general, the clay-rich Ferralsols and Nitisols exhibited a significant response to the organic amendments. Crane-Droesch et al. (2013) conducted a global meta-analysis of crop yield responses to biochar application and concluded that the best opportunities to improve yields through improving soil condition was in the humid tropics on soils with low CEC, low SOC, and perhaps, with lower pH

and heavier textures. Alling et al. (2014) found that application of wood biochar on acid tropical soils in Asia and Africa increased NO<sub>3</sub>-N, exchangeable K and Mg, and extractable P, but had little effect on NH<sub>4</sub>-N. Thus, the Ferralsols and Nitisols in this study fit these characteristics as soils, where the benefits of organic amendments might be relatively significant. Application of pineapple residue compost on a sandy loam soil of pH 4.0 markedly decreased the soil bulk density, and increased the contents of available P and K, the abundance of bacteria and actinomycetes, the activities of catalase, acid phosphatase, and invertase in the soil (Liu et al., 2014).

Application of biochar, compost, and biochar compost mixture significantly reduced the amount of leachate volume, leaching of NO<sub>3</sub>-N, P, and exchangeable cations compared to the untreated control and fertilizer treated soils. Other studies have also shown that application of biochar decreased the leaching of nitrate, ammonium, total nitrogen (Pratiwi et al., 2016; Xu et al., 2016), P, Ca, Mg, and K (Major et al., 2012; Zhao et al., 2014b).

#### 7.3. Impact of amendments on crop performance and nutrient uptake

As with other parameters discussed above, organic amendments resulted in significantly improved plant nutrient uptake, crop performance, and yield, compared to the fertilizer treatment only. Those plant performance indicators that responded significantly to the organic amendments tended to do so most obviously in all trials where a significant difference in both soil properties and crop yield were also observed. Thus, leaf chlorophyll significantly increased as growth of plants progressed, for all organic treatments across trials. Leaf N, P and K were increased in organic amendments in all trials relative to fertilizer treatment. Higher uptake of nutrients by plants suggests that higher concentrations of N, P and K were maintained in the soil solution and were available to plants. In general, the most obvious differences were observed in all trials where differences in soil condition and in crop yield were also observed. Significant improvements in yield were observed in association with the use of the organic amendments in this study. Where improvements in yield were observed, these were accompanied by significant improvements in some soil and plant performance parameters, with this congruence providing increased confidence in the yield results. In maize, increases in yield for the organic amendments, relative to fertilizer treatment, ranged from 12-27% (mass of cobs), with the higher yields associated with the amendments that included compost. In peanut, increases in pod yield for the organic amendments ranged from 17-24%. In neither of these cases was the quality of the crop different between treatments (as measured average grain weight for corn and barley, grade distribution for peanuts, and nitrogen content of grain and kernels).

#### 7.4. Impact of amendments on nitrogen use efficiency

Organic treatments that included compost and N fertilizer significantly increased yield and N use efficiency of barley on Nitisols. Organic amendment by N fertilizer interaction significantly improved barely grain yield, with yield increases of 54 - 60%. Agronomic efficiency and apparent recovery efficiency of N ranged from 6.2 - 26 kg kg<sup>-1</sup> N and 18-59%, respectively. It should be noted that while this site was not on a Ferralsol, the clay content of the soil was high at ~54%, a characteristic shared with the Ferralsols. The reduction of fertilizer rates to 50% of the recommended amount is of particular significance in terms of reducing nutrient runoff to watercourses. Organic amendments increase greater root growth and activity (McNeill and Penfold, 2009; Ouédraogo et al., 2001; Yu et al., 2016). Overall, a qualitative link can be drawn between significant changes in soil water content associated with the organic amendments relative to the fertilizer treatment across the trials. Thus, significant increases in soil water, associated with the organic amendments, were accompanied by yield increases in maize, barley and peanut. Again, this suggests that one of the major benefits of the use of organic amendments is in the potential to increase soil water content in the seasonally dry tropics.

#### 7.5. Impact of amendments on greenhouse gas emissions

The addition of organic amendments to the soil decreased the cumulative emissions of  $CO_2$  and  $N_2O$  from the soil in the peanut trial, but increased them in the maize trial. In all treatments, the emission of both gases increased up to a certain point after application of the amendments, showing that carbon mineralization was triggered. Other researchers have also reported similar findings of (Bass et al., 2016; Hewage, 2016; Zhang et al., 2016). For the maize trial, the median differences in the rates of  $CO_2$  emissions were +12.6%, +37.3%, +23.1% and +51.0% for biochar, compost, compost + biochar, and COMBI, respectively. The increases observed in  $CO_2$  flux are to be expected with the addition of fresh labile carbon to the system, as a result of both biotic factors associated with the stimulation of microbial respiration by the addition of labile carbon and potentially the provision of new microbial habitat associated with the stimulation of microbial respiration by the stimulation of microbial respiration by the stimulation of microbial respiration by biochar surfaces. In the biochar containing treatments, there is potentially also a priming effect associated with the stimulation of microbial respiration by biochar from native carbon in the soil (Luo et al., 2011; Wang et al., 2016; Zimmerman et al., 2011). According to Ventura et al. (2015), only 7% and 3% of the biochar carbon added was decomposed after 245 and 164 days in Italy and United Kingdom sites, respectively.

Differences in  $CO_2$  fluxes between treatments also exhibited strong temporal variability, likely associated with the utilization and subsequent exhaustion of the newly available labile carbon in compost treatments and a priming effect in the biochar containing treatments. In general, the magnitude of the differences between organic treatments and the fertilizer treatment declined over time. This suggests that the elevated  $CO_2$  flux is largely restricted to the initial period following application of the organic amendment and, once the labile carbon is utilized, a longer-term stable component remains. This long-term potential sequestration of the more refractory carbon pool will, to varying degrees, partially offset initial elevated  $CO_2$  emissions measured in these trials. Overall, this study provides evidence that, while the application of organic amendments can lead to elevated  $CO_2$  emission rates in the short-term, the limited duration of this increase, coupled to the long-term storage potential of the refractory components (particularly with biochar), mean there is overall sequestration potential for carbon in the soils. It is worth noting that, whatever their end-use, the compost components would ultimately be re-mineralized to  $CO_2$  and/or methane, so the emissions would occur regardless of whether the composts were incorporated into the soil in these trials.

Published evidence regarding the effects of biochar and compost amendments on N<sub>2</sub>O soil emissions is contradictory, with reduced (Cayuela et al., 2014; Felber et al., 2014; Martin et al., 2015; Rose et al., 2016; Spokas and Reicosky, 2009; Zhang et al., 2010; Zhang et al., 2016), elevated, or negligible (Angst et al., 2014; Suddick and Six, 2013) fluxes reported. In this study, there was a general decrease in  $N_2O$  fluxes, relative to the fertilizer treatment, with median emission rates reduced by 31.2%, 23.3%, 27.8 % and 20.0% for biochar only, compost only, compost + biochar, and COMBI, respectively. A meta-analysis of 296 observations across 61 studies also showed that biochar application led to a significant reduction of 16% for N<sub>2</sub>O emissions (Song et al., 2016). However, with the maize trial, all organic treatments recorded average N<sub>2</sub>O emissions up to an order of magnitude greater than the fertilizer treatment only. Temporal variations in N<sub>2</sub>O fluxes were useful in defining the finer details of the influence that the organic amendments had on fluxes. In general, fluxes were highest at trial initiation, as expected, when concentrations of soil N are high following fertilizer application. Fluxes then drop significantly over a period of a few weeks and, in many cases, remain close to zero thereafter. This is not uncommon, as N<sub>2</sub>O emissions are generally brief and strongly linked to parameters, such as soil water and available nitrogen (Allen et al., 2010; Deng et al., 2015; Macdonald et al., 2014a; Singh et al., 2010b); the former at least, varying dramatically in north Queensland on both the short timescales of rain events and over the course of the seasonal cycle between wet and dry seasons.

What is of most significance from this study is that, following the initial period of 4 - 6 weeks where fluxes from all treatments were elevated,  $N_2O$  emissions tended to be suppressed in the organic treatments, particularly in those treatments containing biochar. In general, where emissions were noticeably reduced relative to the fertilizer treatment, there was a concurrent increase in soil N retention. It is hypothesized that biochar can potentially lower the availability of this retained nitrogen to plants/microbes, which can, consequently, lower total N denitrified in the soil and favor the last step of denitrification; thus, reducing the amount of  $N_2O$  available to be subsequently emitted.

The magnitude of such an effect is likely to be soil, crop, and climate dependent. Overall, in the peanut field, the fluxes of  $CO_2$  and  $N_2O$  were generally lower in organic-amended plots than the control, while the in maize field, emissions of  $CO_2$  and  $N_2O$  relative to yield from all organic amendments were similar to, or higher than, the control. However, the magnitudes of these differences were not constant across each trial. The site of application, as well as the organic amendment selected for application, must be carefully considered if  $N_2O$  emission reduction is a primary aim of amendment application. Temporal variability must also be carefully considered for two reasons:

- The effects of the organic amendments on N<sub>2</sub>O fluxes were not consistent over time. Initially, elevated fluxes, in general, declined below fertilizer fluxes as the time since application increased. It is, therefore, possible that the longer the organic amendment is in the soil, the greater the cumulative emission reduction effect.
- Spikes in N<sub>2</sub>O emissions related to rainfall are generally short-lived and, therefore, easy to miss in sampling campaigns with significant time gaps between sampling.

#### 7.6. Future research needs

This research has investigated the short-term carbon sequestration, soil resilience, and crop yield impacts of the application of compost, biochar, and their mixture to tropical agricultural soils. The long-term carbon-sequestration benefits of biochar addition are simple to predict, given the resistance of biochar to degradation (Bird et al., 2015; McBeath et al., 2015), and results of this study indicated a minimum of 41-67% of the biochar carbon added to the soils in these trials will be stable on centennial timescales. The longer-term impacts of compost addition on carbon sequestration are less clear, as a significant component of the compost will be re-mineralized over annual timescales, with only a small fraction ultimately stabilized by organo-mineral interactions as soil humus. Many of the other potential benefits of compost and biochar additions, in terms of improved soil health and resilience and improved yields, where these occur, also accrue gradually over an extended period involving multiple applications (Diacono and Montemurro, 2010).

Therefore, further research is needed to translate these experiments into longer-term trials conducted at the sites that have shown the most promising preliminary results. Repeated application of compost and biochar, with monitoring of soil condition, plant performance and crop yield over a number of cropping cycles, will be required to properly evaluate the benefits of adoption. Previous studies have also shown that soil humus is built at a faster rate in finer-textured soils (Diacono and Montemurro, 2010; Krull et al., 2004). Given the restricted capacity to supply organic amendments at the present time, the results from this study, combined with previous research elsewhere, suggests that both the Ferralsols and Nitisols have the most potential for improvement through application of compost and biochar (alone or in combination) and, hence, would be the priority for the establishment of longer-term trials. The Ferralsols of the Atherton Tablelands, under original vegetation, have 7-11% organic carbon in mineral soil to 10 cm depth (Maggs and Hewett, 1993); considerably more than is currently the case in cropped soils (< 2% in this study over 30 cm). This suggests a significant capacity to build and stabilize soil carbon over time through organic amendment additions.

Future research should also address the issue of optimizing application rates of organic amendments, as the rates used in this study were simply a single estimate based on previous work as to what might be appropriate. Whereas, application rate (of compost, biochar, or mixes of the two) is known to impact agronomic performance (Fischer and Glaser, 2012; Schulz et al., 2013, 2014), and is likely to vary both by soil and crop type (Biederman and Harpole, 2013; Crane-Droesch et al., 2013; Ding et al., 2016; Jeffery et al., 2011). Moreover, several studies conducted so far on biochar, compost, and their mixture, have employed piecemeal, short-term and single factor. No previous studies have been conducted to assess the interactive effects of biochar, biochar-compost and inorganic fertilizers, particularly nitrogen and phosphorus, on yield, nutrient uptake and nutrient use efficiency of crops. Such studies will enable to determine the amount of inorganic fertilizers reduced due to the use of organic amendments and the rates required to achieve an optimal yield.

A further research need, which was not met by the present study, is the development of a mechanistic understanding of the interactions between biochar, compost, biochar-compost, soil, water, microbes, and crops that underpinned yield improvements and enhancements in soil quality. Such research would require a detailed set of investigations at a limited number of locations, and would be key to developing a predictive capacity for determining the impact of organic amendments on soils, GHG emissions and crops over time. Such a capacity is required to optimize application rates and also to predict what crops and soils might benefit most from the use of such amendments. The development of such a capacity is also key to enhancing adoption of organic amendments into mainstream farming practice, where their use can be demonstrated to be economically viable; in the short term by

increased yield, and in the longer term by improved soil quality and, hence, sustainability of the farming enterprise. As noted above, this is likely to be closely linked to other changes in current practices towards reduced or strategic tillage (Dang et al., 2015; Martinsen et al., 2014). In short, the future research needs in the area of this study can be summarized as the need for a limited number of longer-term field trials, intensively instrumented and monitored, to assess the long-term impact of organic amendments and tillage practices on carbon sequestration, soil quality, crop performance, GHG emissions and nutrient leaching, and also elucidating the biogeochemical processes underpinning observed impacts. These sites would be best focused on the Ferralsols of the Atherton Tablelands in North Queensland, Australia, and on the Nitisols of Ethiopian highlands.

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## Appendices

**Table A1.** Effect of treatments on ratios of exchangeable cations in soil (as moles of charge) under greenhouse condition (chapter 2).

| Treatment          | Ca: ECEC | Mg: ECEC | K:    | Na: ECEC | Ca:   |
|--------------------|----------|----------|-------|----------|-------|
|                    |          |          | ECEC  |          | Mg    |
| Control            | 73.8b    | 14.4d    | 4.8c  | 1.6b     | 5.1ab |
| F                  | 78.5a    | 14.7cd   | 5.0c  | 1.8b     | 5.3a  |
| F + Com            | 74.1b    | 18.5a    | 6.4ab | 2.5a     | 4.0d  |
| F + WB             | 74.2b    | 15.8bc   | 7.0a  | 1.8b     | 4.7bc |
| F + WB + Com       | 73.1b    | 16.5b    | 7.2a  | 2.3a     | 4.4cd |
| F + AB             | 78.9a    | 14.5cd   | 5.2c  | 1.8b     | 5.4a  |
| F + AB + Com       | 76.8ab   | 15.2bc   | 5.8bc | 2.0ab    | 5.0ab |
| Significance level | *        | ***      | **    | *        | ***   |
| LSD (0.05)         | 4.1      | 1.4      | 1.1   | 0.48     | 0.50  |
| CV (%)             | 3.6      | 5.9      | 12.5  | 16.5     | 4.9   |

Significant at \*p < 0.05, \*\*p < 0.01, \*\*\*p < 0.001. Within each column, means with different letters are significantly different at p < 0.05. ECEC effective cation exchange capacity

| Parameter                         | Prin1  | Prin2  | Prin3    | Prin4  |
|-----------------------------------|--------|--------|----------|--------|
| Eigen value                       | 19.88  | 5.66   | 2.75     | 1.71   |
| % variance                        | 66.27  | 18.88  | 9.17     | 5.68   |
| Cumulative                        | 66.27  | 85.15  | 94.32    | 100    |
|                                   |        | Eiger  | ivectors |        |
| Seed yield                        | 0.223  | -0.038 | -0.018   | 0.007  |
| Pod yield                         | 0.221  | -0.066 | 0.032    | 0.054  |
| Chlorophyll content               | 0.206  | 0.106  | -0.183   | 0.007  |
| Nodulation number                 | 0.202  | -0.153 | 0.077    | 0.154  |
| Nodulation dry weight             | -0.179 | 0.249  | 0.009    | 0.070  |
| Specific leaf weight              | 0.220  | -0.025 | -0.107   | -0.047 |
| Mid-point leaf C content          | 0.184  | 0.230  | 0.028    | 0.109  |
| Mid-point leaf N content          | 0.208  | 0.140  | -0.061   | 0.109  |
| Mid-point leaf NO3-N content      | 0.045  | 0.314  | 0.343    | 0.210  |
| Mid-point leaf P content          | 0.194  | 0.209  | -0.028   | -0.045 |
| Mid-point leaf K content          | 0.099  | 0.315  | 0.214    | -0.259 |
| Seed C content                    | -0.014 | 0.132  | -0.415   | 0.499  |
| Seed N content                    | 0.008  | -0.327 | 0.320    | -0.257 |
| Seed 13C content                  | -0.019 | 0.417  | 0.030    | -0.058 |
| Seed 15N content                  | -0.177 | -0.256 | -0.034   | 0.059  |
| Mid-point pH (H <sub>2</sub> O)   | -0.220 | 0.025  | 0.107    | 0.047  |
| Mid-point SOC                     | 0.220  | -0.074 | 0.053    | 0.017  |
| Mid-point total soil N            | 0.195  | 0.015  | 0.256    | -0.192 |
| Mid-point Available soil P        | 0.156  | 0.068  | 0.210    | 0.466  |
| Mid-point soil NH <sub>4</sub> -N | 0.207  | -0.151 | 0.069    | 0.053  |
| Mid-point soil NO <sub>3</sub> -N | 0.220  | 0.018  | -0.009   | -0.136 |
| Mid-point exchangeable Ca         | 0.199  | -0.056 | -0.261   | -0.076 |
| Mid-point exchangeable Mg         | 0.220  | 0.023  | 0.043    | -0.131 |
| Mid-point exchangeable K          | 0.182  | 0.156  | 0.271    | 0.022  |
| Mid-point CEC                     | 0.214  | 0.003  | -0.167   | -0.089 |
| Mid-point exchangeable Al         | -0.167 | 0.260  | 0.119    | -0.124 |
| Mid-point EC                      | 0.220  |        | -0.107   | -0.047 |
| Average SWC                       | 0.193  |        | 0.070    | 0.109  |
| Average CO <sub>2</sub> -flux     | -0.014 |        | -0.401   | -0.408 |
| Average N <sub>2</sub> O-flux     | -0.220 |        | 0.107    | 0.047  |

**Table A2.** Percentage, cumulative variances and eigenvectors on the first 4 principal components for 30 parameters in 5 treatments (peanut trial, chapter 4).

| Fac | tor | δG <sup>1</sup> | <sup>5</sup> N | $\delta S^{12}$ | <sup>5</sup> N | GN   | IC   | SN   | IC   | TN   | JU    | A    | E    | AF   | E    | PE   | Ξ    |
|-----|-----|-----------------|----------------|-----------------|----------------|------|------|------|------|------|-------|------|------|------|------|------|------|
| OA  | Ν   | Mean            | ±SE            | Mean            | ±SE            | Mean | ±SE  | Mean | ±SE  | Mean | ±SE   | mean | ±SE  | Mean | ±SE  | Mean | ±SE  |
| 1   | 1   | 2.78            | 0.12           | 0.69            | 0.04           | 1.43 | 0.03 | 0.57 | 0.04 | 51   | 1.20  |      |      |      |      |      |      |
| 1   | 2   | 2.47            | 0.23           | 1.33            | 0.12           | 1.51 | 0.01 | 0.63 | 0.03 | 59   | 1.86  | 6.2  | 0.57 | 33.3 | 3.56 | 16.0 | 2.32 |
| 1   | 3   | 2.57            | 0.08           | 0.79            | 0.11           | 1.52 | 0.04 | 0.67 | 0.06 | 72   | 7.03  | 10.8 | 0.87 | 45.7 | 4.48 | 32.7 | 2.41 |
| 1   | 4   | 2.60            | 0.22           | 0.76            | 0.05           | 1.59 | 0.04 | 0.63 | 0.03 | 78   | 5.90  | 12.4 | 0.85 | 39.6 | 3.86 | 31.6 | 1.96 |
| 1   | 5   | 2.58            | 0.41           | 0.80            | 0.10           | 1.6  | 0.02 | 0.74 | 0.09 | 81   | 6.77  | 9.3  | 1.05 | 33.0 | 2.48 | 21.4 | 0.29 |
| 2   | 1   | 2.89            | 0.19           | 0.94            | 0.01           | 1.52 | 0.03 | 0.75 | 0.01 | 78   | 5.37  | 13.5 | 0.69 | 48.8 | 4.39 | 40.6 | 1.82 |
| 2   | 2   | 2.90            | 0.41           | 1.44            | 0.13           | 1.59 | 0.04 | 0.67 | 0.02 | 91   | 5.46  | 16.8 | 0.89 | 51.1 | 2.26 | 38.8 | 2.16 |
| 2   | 3   | 2.69            | 0.17           | 0.95            | 0.05           | 1.64 | 0.05 | 0.71 | 0.01 | 101  | 6.03  | 11.8 | 1.09 | 49.0 | 5.82 | 39.7 | 1.86 |
| 2   | 4   | 2.38            | 0.10           | 0.89            | 0.03           | 1.67 | 0.03 | 0.76 | 0.01 | 114  | 8.42  | 13.9 | 0.72 | 50.1 | 3.48 | 37.7 | 2.61 |
| 2   | 5   | 1.96            | 0.13           | 0.87            | 0.15           | 1.70 | 0.03 | 0.72 | 0.02 | 111  | 5.24  | 11.9 | 0.70 | 40.8 | 3.47 | 29.8 | 3.39 |
| 3   | 1   | 2.73            | 0.16           | 1.35            | 0.09           | 1.50 | 0.03 | 0.77 | 0.02 | 85   | 1.33  | 7.5  | 0.82 | 27.6 | 1.17 | 21.6 | 0.66 |
| 3   | 2   | 2.92            | 0.07           | 1.32            | 0.02           | 1.62 | 0.08 | 0.84 | 0.06 | 103  | 2.85  | 8.9  | 0.47 | 34.9 | 1.91 | 23.8 | 0.99 |
| 3   | 3   | 2.76            | 0.17           | 1.33            | 0.18           | 1.60 | 0.03 | 0.82 | 0.05 | 107  | 3.67  | 9.5  | 0.43 | 32.6 | 2.17 | 22.6 | 0.45 |
| 3   | 4   | 2.41            | 0.04           | 0.87            | 0.02           | 1.68 | 0.11 | 0.80 | 0.01 | 121  | 8.01  | 11.5 | 0.94 | 36.1 | 4.12 | 24.4 | 1.27 |
| 3   | 5   | 2.48            | 0.17           | 0.68            | 0.08           | 1.77 | 0.04 | 0.82 | 0.02 | 128  | 4.41  | 11.1 | 0.87 | 35.6 | 2.00 | 22.1 | 1.51 |
| 4   | 1   | 2.78            | 0.16           | 1.49            | 0.05           | 1.55 | 0.07 | 0.91 | 0.05 | 90   | 14.99 | 8.7  | 0.69 | 28.9 | 2.57 | 19.8 | 7.48 |
| 4   | 2   | 3.42            | 0.25           | 1.36            | 0.08           | 1.60 | 0.03 | 0.76 | 0.03 | 101  | 13.26 | 9.5  | 0.70 | 31.2 | 1.36 | 22.9 | 9.51 |
| 4   | 3   | 2.83            | 0.16           | 0.90            | 0.12           | 1.73 | 0.05 | 0.64 | 0.04 | 131  | 5.79  | 10.9 | 0.61 | 44.1 | 3.33 | 33.6 | 1.41 |
| 4   | 4   | 2.81            | 0.12           | 1.05            | 0.03           | 1.68 | 0.07 | 0.88 | 0.02 | 120  | 14.51 | 9.9  | 0.61 | 33.6 | 7.50 | 21.0 | 5.23 |
| 4   | 5   | 2.56            | 0.13           | 0.89            | 0.01           | 1.67 | 0.01 | 0.83 | 0.08 | 138  | 6.75  | 12.6 | 0.73 | 38.0 | 3.16 | 26.6 | 3.94 |
| 5   | 1   | 3.27            | 0.13           | 1.26            | 0.06           | 1.54 | 0.11 | 0.71 | 0.06 | 82   | 14.45 | 8.0  | 1.19 | 27.2 | 2.92 | 22.1 | 1.91 |
| 5   | 2   | 2.99            | 0.10           | 0.95            | 0.07           | 1.59 | 0.09 | 0.79 | 0.02 | 112  | 11.28 | 12.8 | 0.52 | 44.2 | 8.10 | 33.6 | 4.52 |
| 5   | 3   | 2.33            | 0.15           | 1.08            | 0.14           | 1.63 | 0.10 | 0.75 | 0.01 | 123  | 15.12 | 13.6 | 0.69 | 44.9 | 3.63 | 33.8 | 1.59 |
| 5   | 4   | 2.56            | 0.10           | 1.17            | 0.09           | 1.72 | 0.10 | 0.78 | 0.02 | 122  | 12.03 | 10.7 | 0.95 | 38.6 | 6.54 | 27.4 | 3.91 |
| 5   | 5   | 2.16            | 0.19           | 0.62            | 0.07           | 1.71 | 0.06 | 0.79 | 0.04 | 115  | 15.64 | 8.5  | 1.01 | 30.8 | 7.57 | 20.6 | 0.82 |

Table A3. Interaction effects of organic amendments and nitrogen fertilizer on nitrogen use efficiency of barley at Holetta (Chapter 6).

OA: organic amendments; OA1: control; OA2: biochar (56 kg ha<sup>-1</sup>); OA3: compost (125 kg N ha<sup>-1</sup>); OA4: Com+B (136 kg N ha<sup>-1</sup>); OA5: COMBI (115 kg ha<sup>-1</sup>). N1: nitrogen (0); N2: nitrogen (23 kg N ha<sup>-1</sup>); N3: nitrogen (46 kg N ha<sup>-1</sup>); N4: nitrogen (69 kg N ha<sup>-1</sup>); N5: nitrogen (92 kg N ha<sup>-1</sup>).

| Fac | tor | $\delta G^{15} N$ |      | $\delta S^1$ | <sup>5</sup> N | GN   | lС   | SN   | С    | TN   | JU   | AF   | 3   | AR   | E   | PI   | E   |
|-----|-----|-------------------|------|--------------|----------------|------|------|------|------|------|------|------|-----|------|-----|------|-----|
| OA  | Ν   | Mean              | ±SE  | mean         | ±SE            | Mean | ±SE  | Mean | ±SE  | Mean | ±SE  | Mean | ±SE | Mean | ±SE | Mean | ±SE |
| 1   | 1   | 0.86              | 0.15 | 0.35         | 0.02           | 1.26 | 0.02 | 0.39 | 0.00 | 23   | 0.88 |      |     |      |     |      |     |
| 1   | 2   | 1.02              | 0.05 | 0.46         | 0.01           | 1.46 | 0.12 | 0.48 | 0.04 | 33   | 1.45 | 13.0 | 0.9 | 42   | 4.5 | 25   | 2.9 |
| 1   | 3   | 0.95              | 0.11 | 0.51         | 0.01           | 1.46 | 0.01 | 0.43 | 0.04 | 46   | 3.53 | 20.0 | 1.3 | 51   | 2.1 | 56   | 3.0 |
| 1   | 4   | 0.61              | 0.08 | 0.33         | 0.06           | 1.51 | 0.10 | 0.69 | 0.03 | 56   | 2.97 | 14.1 | 0.8 | 48   | 4.2 | 36   | 0.9 |
| 1   | 5   | 0.78              | 0.07 | 0.26         | 0.03           | 1.46 | 0.13 | 0.52 | 0.04 | 69   | 7.23 | 18.7 | 1.2 | 50   | 7.9 | 52   | 4.5 |
| 2   | 1   | 1.41              | 0.07 | 0.16         | 0.01           | 1.32 | 0.03 | 0.47 | 0.01 | 45   | 3.47 | 17.7 | 3.4 | 40   | 5.8 | 46   | 5.3 |
| 2   | 2   | 0.55              | 0.08 | 0.17         | 0.01           | 1.31 | 0.05 | 0.45 | 0.05 | 66   | 3.00 | 26.0 | 1.5 | 54   | 3.9 | 67   | 1.4 |
| 2   | 3   | 0.57              | 0.06 | 0.32         | 0.02           | 1.45 | 0.13 | 0.50 | 0.01 | 84   | 5.61 | 23.1 | 1.5 | 59   | 2.6 | 67   | 0.9 |
| 2   | 4   | 0.30              | 0.02 | 0.13         | 0.01           | 1.44 | 0.08 | 0.46 | 0.01 | 79   | 3.52 | 20.2 | 3.1 | 45   | 2.8 | 49   | 3.1 |
| 2   | 5   | 0.49              | 0.10 | 0.16         | 0.02           | 1.47 | 0.14 | 0.47 | 0.01 | 82   | 3.34 | 18.1 | 0.5 | 40   | 2.2 | 41   | 1.7 |
| 3   | 1   | 1.86              | 0.04 | 0.52         | 0.01           | 1.56 | 0.06 | 0.55 | 0.05 | 51   | 6.25 | 6.9  | 0.9 | 22   | 1.8 | 19   | 2.3 |
| 3   | 2   | 1.31              | 0.07 | 0.27         | 0.01           | 1.52 | 0.09 | 0.51 | 0.03 | 74   | 4.18 | 13.9 | 1.0 | 35   | 2.8 | 34   | 1.0 |
| 3   | 3   | 1.31              | 0.01 | 0.33         | 0.03           | 1.54 | 0.01 | 0.58 | 0.05 | 98   | 5.85 | 17.4 | 1.0 | 44   | 5.5 | 40   | 2.2 |
| 3   | 4   | 0.70              | 0.07 | 0.27         | 0.01           | 1.52 | 0.08 | 0.52 | 0.04 | 87   | 9.42 | 13.5 | 0.4 | 33   | 7.1 | 32   | 1.3 |
| 3   | 5   | 0.82              | 0.26 | 0.24         | 0.02           | 1.57 | 0.03 | 0.51 | 0.06 | 101  | 4.67 | 15.4 | 0.8 | 36   | 2.1 | 33   | 1.5 |
| 4   | 1   | 1.61              | 0.02 | 0.35         | 0.01           | 1.36 | 0.03 | 0.50 | 0.03 | 49   | 3.52 | 8.1  | 1.5 | 21   | 1.4 | 23   | 1.0 |
| 4   | 2   | 0.97              | 0.14 | 0.24         | 0.00           | 1.45 | 0.07 | 0.54 | 0.03 | 77   | 6.23 | 13.4 | 1.7 | 37   | 4.2 | 40   | 2.6 |
| 4   | 3   | 0.84              | 0.06 | 0.19         | 0.06           | 1.63 | 0.08 | 0.61 | 0.10 | 100  | 5.57 | 15.9 | 0.4 | 45   | 3.3 | 39   | 2.1 |
| 4   | 4   | 0.79              | 0.08 | 0.23         | 0.01           | 1.70 | 0.09 | 0.50 | 0.04 | 100  | 4.10 | 14.8 | 0.7 | 40   | 2.1 | 37   | 3.9 |
| 4   | 5   | 0.61              | 0.07 | 0.18         | 0.01           | 1.75 | 0.04 | 0.50 | 0.01 | 100  | 2.91 | 11.4 | 1.2 | 35   | 1.4 | 35   | 0.9 |
| 5   | 1   | 1.86              | 0.08 | 0.36         | 0.01           | 1.34 | 0.05 | 0.41 | 0.01 | 43   | 3.47 | 7.1  | 0.4 | 18   | 0.7 | 25   | 1.1 |
| 5   | 2   | 1.29              | 0.08 | 0.29         | 0.03           | 1.43 | 0.03 | 0.44 | 0.01 | 62   | 6.90 | 11.5 | 1.9 | 28   | 4.9 | 34   | 5.6 |
| 5   | 3   | 1.07              | 0.08 | 0.22         | 0.03           | 1.41 | 0.02 | 0.42 | 0.02 | 66   | 8.09 | 11.7 | 1.5 | 27   | 4.9 | 33   | 6.7 |
| 5   | 4   | 0.52              | 0.05 | 0.28         | 0.04           | 1.48 | 0.06 | 0.52 | 0.06 | 89   | 1.77 | 14.1 | 0.3 | 36   | 0.9 | 37   | 1.8 |
| 5   | 5   | 0.47              | 0.01 | 0.20         | 0.04           | 1.53 | 0.02 | 0.44 | 0.04 | 71   | 1.16 | 8.6  | 1.3 | 23   | 0.5 | 27   | 0.6 |

Table A4. Interaction effects of organic amendments and nitrogen fertilizer on nitrogen use efficiency of barley at Robgebeya (Chapter 6)

| Event                   | Sample date | Event                   | Sample date |
|-------------------------|-------------|-------------------------|-------------|
| Soil sampling           |             | Soil sampling           |             |
| Pre-plant               | 09/12/2013  | Pre-plant               | 09/12/2013  |
| Mid-point               | 23/04/2014  | Mid-point               | 23/04/2014  |
| End-point               | 09/07/2014  | End-point               | 27/05/2014  |
| <b>Plant Biometrics</b> |             | <b>Plant biometrics</b> |             |
| 1st                     | 25/03/2014  | 1st                     | 25/03/2014  |
| 2nd                     | 03/04/2014  | 2nd                     | 03/04/2014  |
| 3rd                     | 15/04/2014  | 3rd                     | 15/04/2014  |
| 4th                     | 23/04/2014  | 4th                     | 23/04/2014  |
| final                   | 12/05/2014  | final                   | 12/05/2014  |
|                         |             | GHG                     |             |
| GHG measurements        |             | measurements            |             |
| Date 1                  | 18/02/2014  | Date 1                  | 15/01/2014  |
| Date 2                  | 27/02/2014  | Date 2                  | 22/01/2014  |
| Date 3                  | 06/03/2014  | Date 3                  | 05/02/2014  |
| Date 4                  | 28/03/2014  | Date 4                  | 18/02/2014  |
| Date 5                  | 29/04/2014  | Date 5                  | 28/03/2014  |
| Date 6                  | 22/05/2014  | Date 6                  | 30/04/2014  |
| Date 7                  | 08/07/2014  | Date 7                  | 15/05/2014  |

**Table A5.** Significant measurement dates during the maize (left, chapter 3) and peanut trials (right, chapter 4).

| Depth (m) | Horizon | Description (maize trial field)   |
|-----------|---------|---|
| 0.0-0.2   | А       | Dark reddish brown (2.5YR3/4, dry; 2.5YR2.5/3, moist) light clay with       |
|           |         | strong structure; zero coarse fragments or segregations; field pH 5.0;      |
|           |         | diffuse to:   |
| 0.2-0.5   | B1      | Dark reddish brown (2.5YR3/4, dry, moist) medium clay; with moderate        |
|           |         | structure; zero coarse fragments or segregations; field pH 5.5; diffuse to: |
| 0.5-1.0   | B2      | Dark red (2.5YR3/6, dry) to dark reddish brown (2.5YR3/4, moist)            |
|           |         | medium clay; with moderate structure; zero coarse fragments or              |
|           |         | segregations; field pH 6.0.   |
| Depth (m) | Horizon | Description (peanut trial field)  |
| 0.0-0.25  | А       | Dark reddish brown (5YR3/4, dry; 2.5YR3/4, moist) medium clay; with         |
|           |         | moderate structure; zero coarse fragments; very few fine-medium             |
|           |         | ferromanganiferous nodules; field pH 6.5; diffuse to:                       |
| 0.25-1.0  | В       | Red (2.5YR4/6, dry) to dark red (2.5YR3/6, moist) medium clay; with         |
|           |         | moderate structure; zero coarse fragments; very few fine-medium             |
|           |         | ferromanganiferous nodules; field pH 6.0.                                   |

**Table A6.** Maize and peanut field soil core descriptions from 0 to 1.0 m depth (chapter 3 and 4).

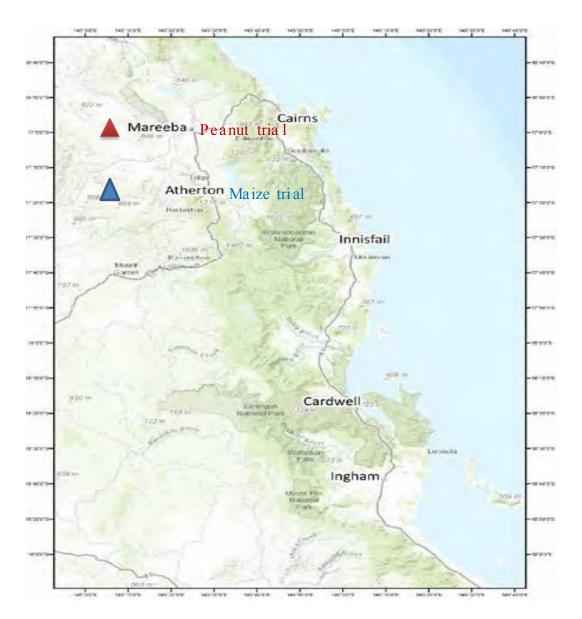


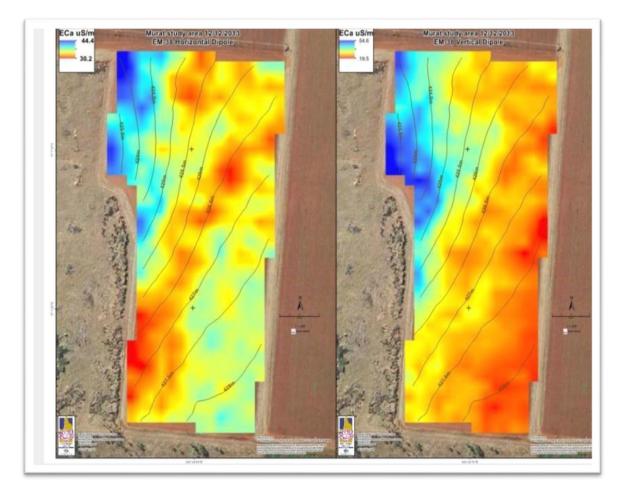
Figure A1. Locations of maize and peanut trials in North Queensland, Australia

|             |             | 6 Rov       | WS          |             |             |             |             | Tre         | atmen       | t 3         |             |             |             |             |  |  |  |
|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|--|--|--|
| ŝ           |             | 6 Roy       | WS          |             |             |             |             | Tre         | atmen       | t 1         |             |             |             |             |  |  |  |
| Replicate 3 |             | 6 Roy       | WS          |             |             |             |             | Tre         | atmen       | t 2         |             |             |             |             |  |  |  |
| Rep]        | -           | 6 Roy       | WS          | Treatment 5 |             |             |             |             |             |             |             |             |             |             |  |  |  |
|             |             | 6 Roy       | WS          | Treatment 4 |             |             |             |             |             |             |             |             |             |             |  |  |  |
|             |             | 6 Roy       | WS          | Treatment 2 |             |             |             |             |             |             |             |             |             |             |  |  |  |
| e 2         |             | 6 Roy       | WS          |             |             |             |             | Tre         | atmen       | t 5         |             |             |             |             |  |  |  |
| Replicate 2 |             | 6 Roy       | WS          |             |             |             |             | Tre         | atmen       | t 4         |             |             |             |             |  |  |  |
| Rep         |             | 6 Roy       | WS          | Treatment 3 |             |             |             |             |             |             |             |             |             |             |  |  |  |
|             |             | 6 Roy       | WS          | Treatment 1 |             |             |             |             |             |             |             |             |             |             |  |  |  |
|             |             | 6 Roy       | WS          | Treatment 4 |             |             |             |             |             |             |             |             |             |             |  |  |  |
| te 1        |             | 6 Roy       | WS          | Treatment 2 |             |             |             |             |             |             |             |             |             |             |  |  |  |
| Replicate 1 |             | 6 Roy       | WS          | Treatment 1 |             |             |             |             |             |             |             |             |             |             |  |  |  |
| Ret         | -           | 6 Roy       |             | Treatment 3 |             |             |             |             |             |             |             |             |             |             |  |  |  |
|             |             | 6 Rov       |             | Treatment 5 |             |             |             |             |             |             |             |             |             |             |  |  |  |
|             | Re          | eplicat     | e 3         |             |             | Re          | plicat      | e 2         | -           |             | Re          | plicat      | e 1         | -           |  |  |  |
| 4 rows      |  |  |  |
| Treatment 1 | Treatment 3 | Treatment 5 | Treatment 2 | Treatment 4 | Treatment 3 | Treatment 4 | Treatment 1 | Treatment 5 | Treatment 2 | Treatment 4 | Treatment 2 | Treatment 3 | Treatment 5 | Treatment 1 |  |  |  |

**Figure A2.** Maize trial layout, row length was 240 m. A total of 90 rows were included with a spacing of 0.9 m between each row (upper). Peanut trial layout, row length was 360 m. 4 guard rows were present between treatments, meaning the trial area included 120 rows spaced 0.9 m apart (lower).



**Figure A3.** Aerial view of the maize trial site in N-S orientation, showing the replicate boundaries. The shaded black area at the western end shows the area where the intensive sampling, including soil sampling, measurements of plant biometrics and GHG fluxes (upper). Aerial view of the peanut trial site in a N-S orientation, showing the replicate boundaries. The dark shaded area at the northern end of the plot shows the area where the intensive sampling, including soil sampling, biometric analysis and GHG fluxes.



**Figure A4.** Vertical (left panel) and horizontal (right panel) EM38 surveys of the peanut field plot carried out on 12/12/2013. A general trend of increasing EC from east to west was measured, though the most rapid transition point was past the far western trial edge. The area of intensive measurement in the NE corner is comparatively uniform in conductivity.



Figure A5. Maize crop at stalk elongation stage as influenced by different treatments



**Figure A6.** (a) Amendment application method using a truck mounted, computer controlled spreader bin and (b) Activity 10, peanut crop 4 weeks after planting, showing row spacing and lateral boom irrigation.



Figure A7. Growth of peanut plants under the different treatment regimes.

|          |    |     |    |    |    |    |            |     |    |     | B  | lock I | II |     |    |     |     |     |    |    |    |     |    |    |
|----------|----|-----|----|----|----|----|------------|-----|----|-----|----|--------|----|-----|----|-----|-----|-----|----|----|----|-----|----|----|
| OA4 OA3  |    |     |    |    |    |    |            | OA1 |    |     |    | OA5    |    |     |    | OA2 |     |     |    |    |    |     |    |    |
| N1       | N2 | N4  | N3 | N5 | N5 | N2 | N3         | N1  | N4 | N2  | N5 | N3     | N1 | N4  | N3 | N1  | N2  | N4  | N5 | N2 | N4 | N3  | N5 | N1 |
|          |    |     |    |    |    |    |            |     |    |     |    |        |    |     |    |     |     |     |    |    |    |     |    |    |
| 51       | 52 | 53  | 54 | 55 | 56 | 57 | 58         | 59  | 60 | 61  | 62 | 63     | 64 | 65  | 66 | 67  | 68  | 69  | 70 | 71 | 72 | 73  | 74 | 75 |
| Block II |    |     |    |    |    |    |            |     |    |     |    |        |    |     |    |     |     |     |    |    |    |     |    |    |
|          |    | OA2 |    |    |    |    | OA4        |     |    | OA3 |    |        |    | OA5 |    |     |     | OA1 |    |    |    |     |    |    |
| N1       | N4 | N3  | N5 | N2 | N5 | N2 | N3         | N1  | N4 | N2  | N4 | N1     | N3 | N5  | N3 | N5  | N1  | N4  | N2 | N4 | N2 | N1  | N3 | N5 |
|          |    |     |    |    |    |    |            |     |    |     |    |        |    |     |    |     |     |     |    |    |    |     |    |    |
| 50       | 49 | 48  | 47 | 46 | 45 | 44 | 43         | 42  | 41 | 40  | 39 | 38     | 37 | 36  | 35 | 34  | 33  | 32  | 31 | 30 | 29 | 28  | 27 | 26 |
|          |    |     |    |    | 1  | 1  |            | 1   |    |     | ]  | Block  | I  |     |    |     |     |     |    |    |    |     |    |    |
|          |    | OA3 |    |    |    |    | <b>OA1</b> |     |    |     |    | OA4    |    |     |    |     | OA5 |     |    |    |    | OA2 |    |    |
| N2       | N5 | N3  | N1 | N4 | N4 | N1 | N3         | N5  | N2 | N1  | N3 | N5     | N4 | N2  | N5 | N3  | N1  | N2  | N4 | N3 | N2 | N1  | N4 | N5 |
|          |    |     |    |    |    |    |            |     |    |     |    |        |    |     |    |     |     |     |    |    |    |     |    |    |
| 1        | 2  | 3   | 4  | 5  | 6  | 7  | 8          | 9   | 10 | 11  | 12 | 13     | 14 | 15  | 16 | 17  | 18  | 19  | 20 | 21 | 22 | 23  | 24 | 25 |

**Figure A8.** Treatments laid out in randomized split-plot design with organic amendments (OA1-OA5) as main plots and nitrogen levels (N1-N5) as sub-plots. OA1: Control; OA2: Biochar: OA3: Compost; OA4: Biochar + compost; OA5: Co-composted biochar-compost. N1: 0; kg N ha<sup>-1</sup>, N2: 23 kg N ha<sup>-1</sup>, N4: 46 kg N ha<sup>-1</sup>, N5: 92 kg N ha<sup>-1</sup>). Blue numbers 1-75 are plot numbers.



**Figure A9.** (a) Composting, biochar, compost and row marking for barley planting (upper), (b) barley trial layout under the different treatments (lower).



Figure A10. Barley plants at different growth stages under different treatment regimes.