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MEASUREMENT OF CO_2 EMISSION FROM BIO-CHAR-AMENDED RICE PADDY FIELD IN THE COASTAL SAVANNAH ZONE OF GHANA.

BY

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DECLARATION

I hereby declare that this thesis has been written by me and that it is a record of my own research work. It has neither in whole nor in part been presented for another degree elsewhere. Works by other researchers have been duly cited by references to the respective authors and all assistance received acknowledged accordingly.

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-- --

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DEDICATION

This work is dedicated to the almighty God for his uncountable grace and glory conferred on me all these years. It is also dedicated to my dad Samuel Koomson, my mum Mary Koomson and Elizabeth Koomson who contributed immensely towards my success.

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ABSTRACT

Agricultural practices affect the production and emission of Carbon dioxide (CO_2) from paddy soils. Evidence now exists on increased CO_2 concentration in the atmosphere as a result of decomposition of organic matter which impacts on climate change and global warming. The release of CO_2 from soil (soil CO_2 efflux or soil respiration) is the largest source of carbon to the atmosphere. In Ghana, the major challenge is lack of data on greenhouse gas (GHG) emission to form the basis of scientific debate. Furthermore, the efficacy of interventions such as bio-char application to reduce GHG emission from crop fields has not been investigated. This research presents a nine (9) months field and screen house experimental study aimed at measuring CO_2 emission from vertisols in the coastal savannah zone of Ghana and to assess the effect of bio-char in combination with irrigation treatments on the emission of CO_2 of the paddy system.

The treatments for the field experiment were established following a split-plot design with soil amendment as main plot treatment and levels of N fertilization as the subplot treatment. The main plots were control (C), bio-char rice husk (BRH) and raw rice husk (RH). The subplots treatments were 0, 45, 90 and 120 kg N/ha applied as urea. The subplots were replicated three times and completely randomized in each main plot. The screen house study was set out in a Completely Randomized Design (CRD) with three types of soil amendment (BRH, RH and C), two fertilizer levels (0 and 90 kg N/ha) and two irrigation treatments (0 cm water head and 5 cm water head). These treatments were replicated three times. Soil CO_2 efflux was measured in-situ using closed chamber method during the whole study period.

Generally, low CO_2 emissions were observed for BRH amended plots followed by C with relatively high emissions from RH amended plots. Soil CO_2 efflux increased with increasing level of N fertilizer. Higher emissions were recorded in the afternoon particularly, in mid-

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morning to afternoon (10-2 pm). Average weekly CO_2 efflux was higher in Season I than Season II over the nine-month study period. Comparing the soil amendment types, the highest mean CO_2 emission of 369 mg m^{-2}/4hrs was recorded from RH treatment whereas, CO_2 emission of 362 and 294 mg m^{-2}/4hrs was recorded from C and BRH amendment respectively during Season I. The lowest average CO_2 efflux of 205 mg m^{-2}/4hrs was recorded on the bio-char amended plots followed by RH and C plots with CO_2 efflux of 293 and 349 mg m^{-2}/4hrs respectively. The lowest average CO_2 emission of 199 mg m^{-2}/4hrs was also recorded on BRH amended plots.

In the screen house study, lower CO_2 effluxes were recorded for flooded treatments than those kept at 0 cm water head. The regression of soil temperature on soil CO_2 efflux showed a positive correlation with CO_2 evolution increasing as temperature increases, and temperature contributing to 61% of the CO_2 efflux, as shown by the regression equation.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

Climate change and global warming continue to be topics of considerable scientific interest and public concern. Greenhouse gases (GHGs) and their interactions with radiation are major causes of global warming that has become a major threat to development and food security especially in the tropics.

Globally, the agricultural sector accounts for 13.5% of GHG emissions while land use, land use change and forestry (LULUCF) represent 17.4% of GHG emissions (IPCC, 2007). The six main types of greenhouse gases are carbon dioxide (CO_2), methane (CH_4), nitrous oxide (N_2O), hydrofluorocarbon (HFC), perfluorocarbon (PFC) and sulfurhexafloride (SF_6) (UNFSCCC, 2011), which results from both industrial and agricultural activities. Carbon dioxide is one of the most significant greenhouse gas contributors to regional and global warming as well as climate change, accounting for 60 % of global warming or total greenhouse effect (Rastogi *et al.,* 2002). Soil is considered as major source and/or sink for atmospheric CO_2 (Jensen *et al*., 1996). Globally, soil is estimated to hold more carbon (1,l00 Gt) than the atmosphere (750 Gt) and the terrestrial biosphere (550 Gt) (Post *et al.,* 1990; Sundquist, 1993). Understanding the controls on soil CO_2 emissions is critical because relatively small changes in soil CO_2 fluxes may drastically alter atmospheric concentrations of CO_2 as well as rates of soil carbon sequestration.

The global atmospheric concentration of CO_2 has increased from a pre-industrial value of 280 parts per million (ppm) to 379 ppm in 2005 (WCCA, 2010). The release of CO_2 from soil

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(soil CO_2 efflux or soil respiration) is the largest source of carbon to the atmosphere in most terrestrial ecosystems (Schlesinger and Andrews, 2000). Soil CO_2 efflux or soil respiration is a major component of the biosphere's carbon cycle because it may constitute about three-quarters of total ecosystem respiration (Law *et al.,* 1999). In recent years, soil CO_2 efflux has been the subject of intense studies because of its potential role in amplifying global warming (Trumbore *et al.,* 1996; Liski *et al.,* 1999; Cox *et al.,* 2000; Luo *et al.,* 2001).

Whereas it is desirable to increase soil carbon storage for beneficial reasons, accelerated decomposition of soil organic matter due to improper soil management depletes the soil of carbon. Coupled with other human activities such as burning of fossil fuels and industrial activities globally, depletion of soil carbon has increased the CO_2 load of the atmosphere from an average value of 315 ppm to 350 ppm over a period of thirty (30) years (IPCC, 2001).

Efforts are underway at finding solutions of mitigating factors that would reduce the CO_2 load in the atmosphere. One of such approaches involves the promotion of high biomass plants to act as CO_2 sinks, fixing CO_2 from the atmosphere in the process of photosynthesis. However, it has been argued that plant conversion of CO_2 into biomass would not constitute a permanent reduction of the CO_2 in the atmosphere so far as deforestation and use of wood as fuel continues.

One other approach of reducing the increased atmospheric CO_2 load is the conversion of CO_2 to soil organic matter through the addition of plants residues. Whether this approach of soil carbon sequestration would be effective or not will depend on several soil, plant and management factors. Some of these factors are: (i) soil factors e.g. soil temperature (Dalias *et al.,* 2001) and moisture (Angle *et al.,* 1984), (ii) plant factors e.g. residue type, C:N ratio etc. (Myers *et al.,* 1997) and (iii) Soil and fallow management (Myers *et al.,* 1997)

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Application of fertilizer has also been proposed to be an important factor for soil organic matter sequestration through increased residue production (Alvarez, 2005; Wilson and Al-Kaisi, 2008), but still unclear is the effect of N fertilization on soil CO_2 fluxes from agricultural ecosystems ((Alluvione *et al*., 2009). Non-significant effects of mineral N fertilization on soil CO_2 fluxes is commonly reported (Liu *et al*., 2006; Amos *et al*., 2005; Almaraz *et al*., 2009), but N fertilization tends to decrease (Al-Kaisi *et al*., 2008, Wilson and Al-Kaisi, 2008) or increase (Sainju *et al*., 2008) CO_2 emissions in some studies.

The application of bio-char to soils is currently gaining considerable interest globally due to its potential to sustainably store soil carbon for extended time periods thereby reducing GHG's emission (Verheijen *et al*., 2010; Lehmann *et al*., 2006; Lehmann, 2007; Downie *et al*., 2009 and Quayle, 2010). Wide variations in the rates of CO_2 emissions from soils treated with bio-char have been reported (Kimetu and Lehmann, 2010; Karhu *et al*., 2011; Zimmerman *et al*., 2011). Global CO_2 flux from soils to the atmosphere is mainly the result of roots and microbial respiration within the soil system as the microbes decompose soil organic matter (Sullivan et al., 2010). Components of bio-char are considered to be considerably more recalcitrant than soil organic matter and as such are only decomposed very slowly, over a time frame of hundreds or thousands of years. This implies that bio-char allows carbon input into the soil to be increased greatly compared to the carbon output through soil microbial respiration and this is the basis behind its potential for mitigating climate change.

1.2 Problem statement

For many years most tropical countries such as Ghana have considered themselves as net carbon sinks or in the worse carbon neutral. This assertion is based on the low level of industrialization. But given the extensive land use change including deforestation, land degradation through poor

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management and periodic bush fires, it is conceivable that their GHG emissions are increasing. The major challenge is the lack of data on GHG emissions to form the bases of scientific debate. Whereas the Environmental Protection Agency (EPA) of Ghana (2000) shows gradual increase in GHG emissions and projects further increase, these are based only on "best guesses" or by the use of emission factors (EFs) published by the IPCC. There is hardly any published measured data to validate these EFs. Furthermore, the efficacy of interventions such as bio-char application to reduce GHG emission from crop fields has not been investigated.

1.3 Aim

This Thesis is set out to address some of the gaps in knowledge outlined in the problem statement. It aims at:

i. Measuring CO_2 emissions from paddy fields within the coastal savannah zone of Ghana;
ii. Assessing the effect of bio-char in combination with irrigation treatments on the emission of CO_2 of the paddy systems.

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CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Climate Change

Humanity is confronted with an urgency of crisis that links today to tomorrow. That crisis is climate change and its implications for human survival and development. Agriculture and climate change are inextricably linked. Nelson (2009) observed that "Agriculture is part of the climate change problem, contributing about 13.5% of annual greenhouse gas emissions (with forestry contributing an additional 19%) compared with 13.1% from transportation. Agriculture is however, also part of the solution, offering promising opportunities for mitigation through carbon sequestration, soil and land use management and biomass production.

The term "climate change" is often used interchangeably with the term "global warming". Climate change refers to any significant change in measures of climate (such as temperature, precipitation, wind e.t.c) lasting for an extended period (decades or longer). Global warming on the other hand refers to an average increase in the temperature of the atmosphere near the earth's surface, which can contribute to changes in global climate pattern. Rising temperatures are just one aspect of climate change. Global warming has been identified as the main cause of fluctuations in climate variables including precipitation and temperature. In Ghana for instance, EPA (2000) has indicated that over the past thirty (30) years, the average temperatures of most places have increased by 1°C and the average precipitation reduced by 1 % in most areas.

Greenhouse gas (GHG) emissions and their interactions with radiation are major causes of global warming. Climatologists generally agree that the concentrations of GHG's (carbon

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dioxide (CO_2), methane (CH_4), oxides of nitrogen (N_2O, NO_2), hydro-and per-fluorocarbons) in the atmosphere are increasing at alarming rates. Anthropogenic activities such as agriculture have been identified among the major causes for the increase of these GHG's namely Carbon dioxide (CO_2), nitrous oxide (N_2O) and methane (CH_4). Whereas much of the historical emissions of GHGs may be attributed to fossil fuel burning, land use change accounts for recent increases in emissions from fertilizer application, rice fields, domestic animals and biomass burning. Greenhouse gases that are generated from paddy field activity are CO_2, CH_4 and N_2O (Pantawat, 2012). Carbon dioxide from paddy rice field is as a result of rice photosynthesis and respiration, the soil microbes and loss of soil organic carbon. Methane evolves under anaerobic conditions and through organic decomposition. Nitrous oxide results from the general use of nitrogen fertilizers, which decompose into nitrous oxide through the denitrification process caused by flooding (Shioiri, 1942).

Among these GHG's implicated in global warming, CO_2 is one of the most significant greenhouse gas contributors to regional and global warming as well as climate change, accounting for 60% of global warming or total greenhouse effect (Rastogi *et al.,* 2002). The Intergovernmental Panel on Climate Change, IPCC (2001) also stated that CO_2 is one of the leading compounds causing global warming. The level of CO_2 in the atmosphere is believed to be on an average value of 280±10 ppmV during the pre-industrial period and rose to 315 ppmV in 1957 when accurate monitoring started. Since then, a dramatic rise has been observed, leading to current level of about 380 ppmV (Mark, 2003).

Carbon dioxide has a relatively long residence time in the atmosphere and traps heat that is radiated to the atmosphere from the earth. As indicated by Emiliani (1992), if the earth did not have an atmosphere or if it had an atmosphere without water and CO_2, the average surface

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temperature would be the same as that of the moon (around -18 °C) and everything would be frozen. Fortunately water vapour and CO_2 in the atmosphere keep the earth warm. While playing this essential role, the balance of these gases in the atmosphere is of utmost importance. Gas inputs into the atmosphere are mainly from volcanoes, agriculture, vehicles and industry and output are absorption by plants and precipitation followed by storage as coal, fossil fuels or limestone. If this balance shifts towards higher CO_2 concentration (i.e. too much volcanic activity, decomposition of organic matter in agriculture or emission from vehicles), temperature rises, the ocean warms up and more water vapour is added to the atmosphere through evaporation from oceans. On the other hand, if ice forms on land (temperate region) the earth's albedo increases, temperature drops, the oceans cool and more CO_2 and water vapour returns to them. This decreases the atmospheric temperature further, thus producing glaciations. The current general consensus is that the inputs outweigh the outputs and therefore global warming will continue to be a major threat to atmospheric stability.

Due to the presence of enormous soil carbon in the first one meter depth of soil (Esteban and Jackson, 2000), greenhouse gas induced climate change caused by influx of soil organic carbon in mineralized form would be undoubtedly pronounced. This will have significant serious implications on global temperatures as well as impacts on soil fertility and agricultural productivity.

2.2 Soil CO_2 efflux

Soil CO_2 efflux is the result of complex interactions between climate and soil biological, chemical and physical properties (Oorts *et al*., 2007). The release of CO_2 from soil (soil CO_2 efflux) is the largest source of carbon to the atmosphere (Schlesinger and Andrews, 2000). Soil surface CO_2 efflux is a major component of biosphere's carbon cycle because it may constitute

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about three-quarters of total ecosystem respiration (Law, 2001). In recent years, soil CO_2 efflux has been subject of intense studies because of its potential role in amplifying global warming (Trumbore *et al*., 1996; Liski *et al*., 1999; Cox *et al*., 2000; Giardina and Ryan, 2000; Kirschbaum, 2000).

Soil CO_2 efflux is dependent on land use and land management. In Ghana, common land use systems include forestry, agriculture, paddy rice and animal husbandry. Poor land use such as deforestation and excessive tillage lead to net CO_2 release into the atmosphere (Robertson *et al*.,2000; Oorts *et al*., 2007). Understanding the controls on soil CO_2 emissions is critical because relatively small changes in soil CO_2 fluxes may dramatically alter atmospheric concentrations of CO_2.

2.3 Soil as Sink and Source of GHG's

Greenhouse gases (GHG's) are gases that make the atmosphere function like the glass in a greenhouse. They trap the sun's shortwave energy and re-emit it as heat producing long wave radiation, causing an increase in atmospheric temperature. Under the United Nations' Framework Convention on Climate Change (UNFCCC), a SOURCE of a greenhouse gas in the atmosphere can be defined as an activity or process that contributes to atmospheric emission of that gas, while a SINK is defined as any process, activity or mechanism that removes these gases from the atmosphere into plants and soils.

In this regard, while the soil constitutes the most important source of three biologically generated GHG's, it at the same time remains the greatest sink for CO_2, which alone constitutes about 60 % of all the GHG's effect in the atmosphere (Rastogi et al., 2002). One most important substance or matter in soil described as the "life blood" of soils; organic matter influences the

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physical, chemical and biological properties of soil and performs the dual role as a source and sink of CO_2. During decomposition, soil organic matter serves as a source since CO_2 is lost into the atmosphere. However, when it becomes stable in soil (resistant to decomposition), it serves as a sink of soil carbon.

Soil organic matter comprises the sum of all organic substances in the soil and is defined as a "mixture of plant and animal residues at various stages of decomposition, of substances synthesized microbiologically and/or chemically from the breakdown products, and of the bodies of micro-organisms and small animals and their decomposing products" (Schnitzer, 1991).

2.4 Carbon in Agricultural Soils

World soils constitute a principal carbon pool of 1,500 to 2,000 Pg (1pecagram = 1 billion metric tonnes) in soil organic carbon and 800 to 1,000 Pg as soil inorganic carbon or carbonate carbon (Post *et al*., 1982; Eswaran and Berg, 1993). Soil holds over three time as much carbon as the atmosphere (Lehmann and Joseph, 2009), more than the earth's vegetation and atmosphere combined, and have the capacity to hold much more (Lal, 2004).

The soil organic carbon content is generally high in virgin soils under grass or forest vegetation. Carbon stocks in terrestrial ecosystems have been greatly depleted since the beginning of the Industrial Revolution, with changes in land use and deforestation responsible for the emission of over 498 gigatons of CO_2 to the atmosphere (IPCC, 2000), approximately half of which has been lost from soils (IPCC, 2000). Cole *et al*., (1993) and Lal (1995) also suggested that losses of soil organic carbon have been accentuated to low production levels, intensive tillage, inadequate use of fertilizers and organic amendments, removal of crop residue, biomass burning, lack of soil protection against erosion and other degradative processes. Estimates of loss

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of soil organic carbon from cultivated soils of the world (croplands) range from 41 Pg (Houghton and Skole, 1990) to 55 Pg (Cole, 1996).

Soil carbon comprises soil organic carbon (SOC) and soil inorganic carbon (SIC). Soil organic carbon is a complex and dynamic group of compounds formed from the carbon originally harvested from the atmosphere by plants. During photosynthesis, plants transform atmospheric carbon into forms useful for energy and growth (Schlesinger, 1997). Organic carbon then cycles from the plant to the soil where it becomes an important source of energy for the soil ecosystem, driving many other nutrients cycles. Soil inorganic carbon is the result of mineral weathering and forms a small proportion of many productive soils.

2.5 Dynamics of Soil Carbon Sequestration

Food and Agriculture Organization, FAO (2000) defines carbon sequestration as the capture and secure storage of carbon in plants and soils that would otherwise be emitted to or remain in the atmosphere. In the face of climate change and increasing CO_2 levels in the atmosphere, the global carbon cycle, soil organic carbon sequestration, and the role of different world biomes as potential sources and sinks of carbon are receiving increasing attention (Feller and Bernoux, 2008). Carbon sequestration in plant and soil systems offers an opportunity for mitigating the greenhouse effect (Lal, 2004). In soil systems effective and efficient management of the soil carbon store-house is thus essential for maintaining soil fertility and sustaining high yields.

Soil organic carbon (SOC) is a dynamic group of compounds that have their origin in the photosynthetic activity of trees, grasses, shrubs, forbs and legumes. The carbon in these compounds cycles through solid forms back to the atmosphere at different rates, with turnover times ranging from months to hundreds of years (Davidson and Janssens, 2006; Six and Jastrow,

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2002). During photosynthesis, plants reduce carbon from its oxidized form into the organic forms useful for growth and energy storage (Schlesinger, 1997). Some of this carbon fixed from the atmosphere in time becomes soil carbon through the processes of above and below ground decomposition, root die-off, and the release of sap exudates from plant roots into the soil (exudates contain carbohydrates). Photosynthesis also provides the raw materials for indirect imports of Carbon-rich materials onto and into the soil.

Soil carbon includes soil inorganic carbon (SIC) in the form of carbonates. Soil inorganic carbon is the result of mineral weathering, and is less responsive to management than SOC, turning over much more slowly (Izaurralde, 2005). Soil inorganic carbon content is low in many productive soils and lower than microbial carbon. Soil microbial biomass carbon forms 1 – 3 % of total soil carbon. Soil organic matter determines soil fertility and stability (Herrick and Wander, 1998). Most SOC is found in the top of the soil profile, due to the presence and influence of biotic processes there, with approximately 64% of soil carbon in the top 50 cm (Conant *et al.,* 2001). Soil organic carbon accumulation is positively correlated with precipitation and negatively correlated with temperature (Jones, 2007). The stock of SOC accumulation is highest in cool, wet conditions (Schlesinger, 1997) and lowest in deserts. Soil carbon stocks are positively correlated with the presence of clay and iron and negatively correlated with the bulk density of soil.

The rate of carbon sequestration is determined by the net balance between carbon inputs and carbon outputs. Carbon inputs and outputs are affected by management and by two biotic processes: (i) production of organic matter in the soil and (ii) decomposition of organic matter by soil microorganisms. The biotic processes are strongly controlled by physical, chemical and

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biological factors including biome, climate, soil moisture, nutrient availability, plant growth and erosion (Post *et al*., 2001; Derner and Schuman, 2007; Jones, 2007; Ingram *et al*., 2008).

Soil CO_2 is the main end product of the decay of SOC. Under aerobic conditions CO_2 is produced by respiration of bacteria and protozoa in the guts of insects, and also by bacteria and fungi in the soil (Singer and Munns, 1987). Soil CO_2 production accelerates with temperature and with exposure of soil organic matter to air in pore spaces and on the surface of the soil. When decomposition and soil CO_2 production is slowed, the net rate of the soil carbon accumulation and storage may increase.

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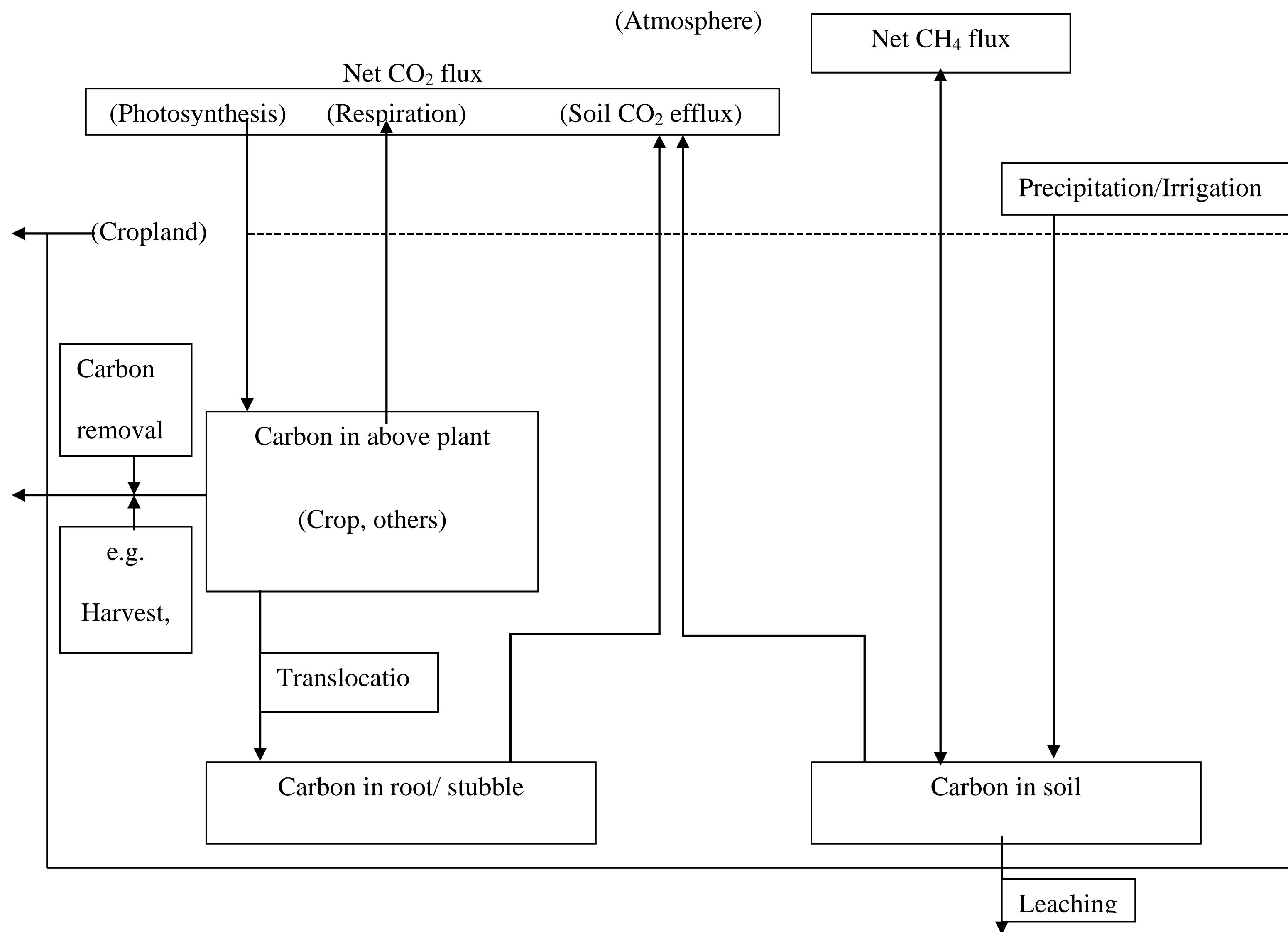

Fig. 2. 1 Schematic diagram of carbon dynamics in cropland ecosystems. Adapted from Nishimura *et al.* (2008).

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2.6 Carbon Dynamics in paddy fields

The dynamics of carbon in paddy fields significantly differs from that in fields with upland crop cultivation in which aerobic decomposition process is dominant. During the submerged period of paddy rice cultivation, CO_2 production in the soils is severely restricted under anaerobic condition. Instead, CH_4 is actively produced in the soil and emitted to the atmosphere mainly through the rice plants. However, when irrigation is withheld to conserve water which is a recommended practice, CO_2 production increases.

The processes of carbon dynamics in submerged paddy soil have been investigated in the previous studies. Kimura *et al.* (2004) recently summarized a review paper in which individual anaerobic decomposition processes of plant residue and soil organic matter, and fates of photosynthesized carbon of rice in the soil were described. Studies on the CH_4 emission have also been intensively conducted including many field experiments (Sass *et al*., 1992; Yagi *et al*., 1996; Corton *et al*., 2000; Wang *et al*., 2000; Wassmann *et al*., 2000; Yu *et al*., 2004) and regional-scale estimations with mathematical models (Mitra *et al*., 2002; Park and Yun, 2002; Liu and Wu, 2004). However, studies on comprehensive carbon dynamics are limited in paddy fields. In particular, studies on CO_2 flux on the flood water surface are limited (Kimura *et al*., 2004), with only a few reported (Koizumi, 2001; Usui *et al*., 2003). Therefore, soil carbon budget in paddy fields has not been sufficiently clarified to date (Nishimura *et al*., 2008).

2.7 Soil Carbon pools and Carbon fraction

Soil organic matter consists of different pools that vary in their intrinsic decay rates and in the factors which control decomposition rates (McGill, 1996). Researchers employ the concept of carbon pools to distinguish between carbon that cycle at different rates in the ecosystem. To

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facilitate the study of organic matter, the general approach is to classify organic matter into various pools (Jackson *et al*., 2002). As yet there is no general agreement on the number of pools of organic matter. However, for the purpose of this research, three main pools may be recognized namely the (i) Active, (ii) Passive (intermediate) and (iii) Slow pool. Carbon in each pool has a different turnover time or Mean Residence Time (MRT).

Carbon pools are not distinct groups of carbon compounds, which are called fractions. There are two soil fractions, the light fraction and the heavy fraction, which are further classified and range from free light fraction to the heavy occluded fraction. Light fractions are composed of fresh plant materials that are subject to rapid decomposition, with turnover from a few months to a few years. Early changes in SOC due to management often occur in the small light fraction, which is known for its spatial and temporal variability. Because most of turnover of SOM is in the light fractions, it is important to include this fraction within any chosen quantification methodology (Post *et al*., 2001). Accumulation of light fraction carbon can be quite large in permanently vegetated soils (i.e. forest and grasslands).

Carbon in the heavy occluded fraction has a MRT from hundreds to over a thousand years. Soil organic carbon and soil organic matter in this fraction are less susceptible to decomposition than in the light fraction. The heavy fraction is composed of polysaccharides (sugar) and humic materials often stabilized in complexes with clay minerals and silt-sized particles (Schlesinger, 1997). One very chemically recalcitrant portion of the heavy fraction has turnover times of 1,500 to 3,500 years (Post *et al*., 2001)

In furtherance of the soil carbon pool and fraction concept, SOC and SOM can be protected from microbial metabolization or decomposition through three ways (Jastrow and Miller, 1998):

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(i) Biochemical recalcitrance occur due to the chemical characteristics of carbon substrate and because substrates are not consumed by microbes, they remain un-decayed compounds and become progressively less decomposable. (ii) Chemical stabilization occurs with the bonding of positively charged cations associated with SOC to negatively charged ion and clay anions. (iii) Physical protection of SOM occurs within soil aggregates, held together by "aggregate glues" such as glomalin, a sticky substance produced by soil fungi that is 30 – 40 % carbon by weight. Soil organic carbon lower in the profile tends to be protected from microbial decomposition due to chemical stabilization. Physical protection can vary by depth and soil type (Del Galdo *et al.*, 2003).

2.8 Factors affecting Soil CO_2 efflux

The critical factors reported to influence soil CO_2 efflux rates include temperature and moisture, nutrient content, root respiration, microbial processes, organic matter, soil aeration, soil porosity, net primary productivity (NPP) and vegetation type (Lundegordh, 1927; Reiners, 1968; Rixon, 1968; Edwards, 1974; Weber, 1990; Johnson, 1993; Mielnick and Dugas, 1999; Maier and Kress, 2000; Schlesinger and Andrew, 2000)

2.8.1 Temperature effects on soil CO_2 efflux

Temperature is an important parameter known to affect most of the major processes in the carbon cycle. It affects the allocation of carbon between roots and shoots (Farrar, 1988), respiratory losses of carbon by plants (Ryan, 1991) and mineralization potentials of the microbial populations (Ross *et al.*, 1999). The exact nature of the relationship between soil respiration and temperature is still not clear. However, understanding how temperature affects soil respiration is vital for predicting soil response to climate change.

Under field conditions, soil temperature and moisture exhibit large amplitude with respective influences on soil respiration. The increase in the reaction rate per 10 °C increase in temperature is known as the respiratory quotient (Q_{10}). The exponential function of Q_{10} is commonly used to express the relationship between soil biological activity and temperature, although Holland *et al.* (1995) have shown that estimates of global soil respiration are very sensitive to the selected Q_{10} value for various biomes. Respiratory quotient values vary considerably among ecosystems and across temperature ranges because of the different temperature sensitivities showed by various components of soil respiration. These components include respiration by live roots, associated mycorrhizae, root exudates and humified organic matter by soil heterotrophs (Trumbore *et al.,* 1996).

Rising temperatures cause an exponential increase in microbial respiration over a range of temperatures. This speeds up the mineralization of organic carbon to CO_2 provided these temperatures do not inhibit the activity of the decomposing community. Studies by Vasconcellos (1994) using tropical and temperate soil showed higher mineralization rates at 35 °C than at 15 °C. He also noted that carbon turnover at 15 °C occurred 2 – 5 times slower than at 35 °C.

2.8.2 Moisture effect on Soil CO_2 efflux

Soil CO_2 efflux could be altered dramatically by changes in soil moisture since moisture affects rooting depth, root respiration and soil microbial community composition. An optimum moisture condition in most soil is when one-half to two-thirds of the pore space is filled with water (Angle *et al*., 1984). Scientists have debated the effect of moisture availability on soil metabolic activity. Raich and Potter (1995) synthesized three phases of moisture effects on soil biota that were identified over time : i) when soils are relatively dry, metabolic activity increases with increasing

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moisture availability; ii) when soils are 50 – 80 % saturated, soil biological activity is almost at its potential; iii) when soils are too wet , oxygen deficiencies inhibit aerobic respiration.

The effect of drying and rewetting soils (Birch, 1958) showed an increased rate of CO_2 evolution after rewetting was compared to soils kept continuously moist, due to the well-established influence of a wetting-drying carbon cycle on microbial activity (Orchard and Cook, 1983). In agreement with this statement, Rochette *et al*., (1991) found high CO_2 fluxes from soil following rainfall after a dry period. In three hours after the rainfall, soil respiration was nine times higher than before and gradually decreased with time.

Haynes (1986) noted that drying-wetting cycles tend to stimulate the mineralization of labile substrates (e.g. hemicelluloses), which are broken down largely by rapidly growing bacteria. Drying-wetting cycles also retard the mineralization of recalcitrant ones (e.g. lignin), which are broken down by slow-growing fungi. It was also observed that under very high soil moisture conditions, water acts as an effective barrier to oxygen supply to decomposers in soils or in wet microsites within well-drained soil. This tends to slow down the mineralization of carbon in soils.

2.8.3 Effect of soil porosity on soil CO_2 efflux

Johnson (1993) suggested the importance of incorporating soil air-filled macro-porosity in CO_2 flux models. Generally, transport of gases through soil is greater in a porous soil than in less porous soil, and this accelerates soil respiration and increases CO_2 efflux from the soil surface. Bouma and Bryla (2000) pointed out that CO_2 efflux from the soil surface was restricted after watering in finer textured soils than in sandy soils because of reduced macro-porosity. The

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dependence of soil respiration on soil porosity is improbable to be linear and cannot be expressed simply (Fang *et al*., 1998).

2.8.4 Effect of nutrient levels on soil carbon storage

Fertilization is a primary means used to increase plant productivity and crop yield. Any increase in biomass also offers increased scope for carbon sequestration by soil. Himes (1998) estimated that it takes 833 kg N, 200 kg P and 143 kg S to sequester 10 tonnes of carbon in humus. Soil fertility is therefore an important aspect of carbon sequestration. Fertilization has been recommended and proved to be successful method of increasing carbon sequestration (Lal *et al*., 1999).

A few field experiments suggest that soil organic matter increases with elevated CO_2 (Schlesinger, 1977). Large accumulations of organic matter are expected where environmental factors (e.g. temperature) limit decomposers. Thus, increased delivery of labile organic matter to the soil could influence soil microbial communities and furthermore soil respiration rates. It is expected that soils with high organic matter and high root and microbial activities would vent more CO_2 than do soils with low organic matter (Bazzaz and Williams, 1991).

2.8.5 Effect of tillage on soil CO_2 efflux

Pretty *et al*. (2002) identified tillage as one of the major factors responsible for decreasing carbon in agricultural soils. The mould board plough and disc harrow are the biggest contributors to the loss of soil carbon through their destruction of soil aggregates and acceleration of decomposition by mixing of plant residues, oxygen and microbial biomass. Soil aggregates are vital for carbon sequestration (Six *et al.,* 1999), a process that is maximal at intermediate aggregate turnover

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(Plante and McGill, 2002). Of the organic matter fraction, the particulate organic matter is the most tillage sensitive (Hussain *et al*., 1999).

The flux of CO_2 from soil generated directly by tillage process may not always reflect the overall release of CO_2 and hence carbon storage of the system. This is illustrated by a comparison of conventional disc tillage and non-tillage in Central Texas (Franzleubbers *et al*., 1995) where, seasonal evolution of CO_2 was up to 12% greater in the non-till system. The study suggested that a change in the dynamics of carbon sequestration and mineralization have occurred under the non-till system. Similarly, Constantini *et al*., (1996) found that more CO_2 was released from zero-till or reduced –till compared to conventional tillage despite increased levels of soil carbon. This was ascribed to an increase in the microbial biomass.

2.9 Measurement of soil CO_2 efflux

Measuring soil CO_2 flux is important to accurately evaluate the effect of soil management practices on global warming and carbon cycling. Soil carbon modellers' generally consider soil efflux as a function of soil temperature or a combination of soil temperature and moisture (Raich and Schlesinger, 1992; Davidson *et al*., 1998; Epron *et al*., 1999; Treonis *et al*., 2002). However, there is no consensus in functional forms and parameterization in these models. The uncertainty is partly due to instrumentation and methods used to measure soil CO_2 production and efflux (Livingston and Hutchinson, 1995; Davidson *et al*., 2002). Although accurate prediction of soil CO_2 efflux is important for models of global carbon cycle, uncertainties in estimates exist due partly to methodological differences (Raich and Schlesinger, 1992) and variation in substrate availability, soil temperature and moisture (Davidson *et al*., 1998, 2006; Xu and Qi, 2001; Rodeghiero and Cescatti, 2008).

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Despite these challenges a variety of methods exist to measure soil CO_2 efflux (Sullivan et al., 2009). Widely used methods are static or dynamic chambers placed on the soil surface and soil diffusion gradients (Sullivan et al., 2009). Majority of these measurement methods are still being developed. Romell (1932) used the accumulation technique, in which a container is placed on the soil surface to trap the evolving gas. Carbon dioxide liberated from the soil accumulates in the container and a sample of the air in the container after a given period of time is compared with a sample taken at the beginning of the experiment. A modification of this method (Lieth and Quelette, 1962) is to place a carbon dioxide absorbant, commonly a solution of caustic alkali inside the container and then estimate the amount collected over the known period. Monteith et al., (1964) estimated the rate of evolution of CO_2 from soil by placing oven-dried (2 hours at 100 °C) soda lime granules in a petri dish placed under a glass tank which was inverted and pressed into the soil. After several days, the soda lime was oven-dried and weighed, and the weight increase per day divided by the cross-sectional area of the tanks was assumed to give the rate of evolution of CO_2.

Also an early method periodically extracts soil samples from different depths to study CO_2 profile and diffusion (De Jong and Schapper, 1972; Wagner and Buyanovsky, 1983; Burton and Beauchamp, 1994; Davidson and Trumbore, 1995). The gas extraction method can provide information of soil CO_2 production at several depths, but it cannot provide in situ, continuous and convenient data on CO_2 efflux. Furthermore, this method will disturb the soil environment.

Chamber-based methods whether static or dynamic allow us to measure soil CO_2 efflux by quantifying the increase of headspace gas concentration within the chamber headspace over a known time (Hutchinson and Mosier, 1981; Jensen *et al*,. 1996). Fixed (static) and portable chambers have evolved into automated systems for continuous and semi-continuous

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measurements (Goulden and Crill, 1997; Russel *et al*., 1998; Scott *et al.,* 1999; Drewitt *et al*., 2002; King and Harrison, 2002). Shortcomings with closed-chamber methods, however, still exist. Efflux readings may be biased by disturbing air pressure and altering CO_2 concentration in the soil (Livingston and Hutchinson, 1995; Healy *et al*., 1996; Davidson *et al*., 2002). By measuring accumulation of soil CO_2 productivity released from the soil surface, chambers are unable to provide information about soil profiles and individual contributions at certain soil depths, which is important for understanding soil carbon mechanisms.

Understory eddy covariance towers provide an alternative to measure soil CO_2 efflux continuously without disturbing the soil (Baldocchi and Meyers, 1991; Law *et al.,* 1999). With this technique, understory eddy covariance measurement may face difficulty in measuring respiration at night when turbulence is weak and intermittent and drainage flows dominate the transfer of CO_2 (Goulden *et al.,* 1996; Moncrieff *et al.,* 1997). Compared with overstory eddy covariance the low height of understory eddy covariance towers corresponds with small areas of foot print, which may induce errors when large areas of ecosystem are represented. Furthermore, understory eddy covariance data cannot separate a soil CO_2 efflux, bole respiration below sensors and overlying herbaceous vegetation when it is present.

The ideal system to measure soil CO_2 efflux has been described by Longdoz *et al*., (2000) to meet the following three requirements: (i) it must not disturb the vertical soil CO_2 concentration gradient at the soil-atmosphere boundary; (ii) it must not disturb the horizontal air velocity above the soil; and (iii) it must not disturb the vertical pressure gradient at the soil atmosphere boundary. These three requirements, however, do not consider such logistical factors facing investigators such as probability, cost or sensitivity to changes in soil temperatures or soil water content, two physical parameters often used to model soil CO_2 efflux. Non-automated

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chamber-based systems are often used to measure soil CO_2 efflux because their portability and low cost make them efficient at capturing spatial variation in soil CO_2 efflux. However, soil CO_2 efflux measurements using non-automated chambers are typically less than one hour in duration and diel measurements are less labour intensive; repeated measurements are needed to construct diel patterns.

Additionally, vegetation should not be present within the sampling areas of chambers, for the above ground component may photosynthesize and respire within the chamber and thus confound soil CO_2 efflux. Thus vegetation is usually removed or clipped from the sampling area before measuring soil CO_2 efflux, further altering soil CO_2 efflux (Grogan and Capin, 1999). Chamber methods may disrupt diffusion of CO_2 out of the soil, or may change the different pressures between the atmosphere and the headspace. As the CO_2 concentration increases within the chamber headspace of static chamber, the concentration of the soil decreases and the measured flux may be reduced (Davidson *et al*., 2002).

Conversely, dynamic chambers that are chemical CO_2 scrubbers may overestimate fluxes by increasing the diffusion gradient out of the soil when they reduce the CO_2 concentration in the chamber headspace below ambient levels (Davidson *et al*., 2002). Both static and dynamic chambers are susceptible to problems associated with pressure differences between the air inside and outside the chamber (Davidson *et al*., 2002; Dore *et al*., 2003). In unvented chambers, the increase in gas concentration can increase the pressure in the chamber above ambient levels, suppressing fluxes as gas diffuses laterally away from the zone of high concentration. However, Bain *et al*. (2005) have shown that vented chambers may overestimate fluxes as a result of the venturi effect (Conen and Smith, 1998) when even low wind speeds cause a mass flow of CO_2 out of the soil profile.

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Until one method is proven to best minimize artifacts when measuring soil CO_2 efflux, and is efficiently deployable, investigators will have to choose among a variety of available methods. Several studies have shown that different methods of measuring soil CO_2 efflux yield different results (Nay *et al*., 1994; Jensen *et al*., 1996; Bekku *et al*., 1997; King and Harrison, 2002; Liang *et al*., 2004; Pumpanen *et al*., 2004).

2.10 Bio-char

2.10.1 Origin and production

Bio-char is a name for charcoal when it is used for particular purposes, especially as soil amendment. Like all charcoal, bio-char is created by pyrolysis of biomass (Lean and Geoffrey, 2008). The Pre – Columbian Amazonians are believed to have used bio-char to enhance soil productivity. It is produced by smouldering agricultural waste (that is, covering burning biomass with soil) (Solomon *et al*., 2007) in pits or trenches (Lehmann, 2007).

Early studies of soils being enriched from what appears to be deliberate mixing of burned biomass in soils around human settlements generated more recent interest in bio-char. These deposits of enriched soils known as terra preta in the Amazon region of South America have a fascinating history of their own (Lehmann, 2004). The term “bio-char” was coined by Peter Read to describe charcoal used as soil improvement.

Bio-char is produced through an energy conversion process called pyrolysis, which is essentially the heating of biomass in the complete or near absence of oxygen. Pyrolysis of biomass produces char, oils and gases. During pyrolysis, the polymeric building blocks of biomass, namely cellulose, hemicelluloses and lignin undergo various processes such as cross-linking, depolymerisation and fragmentation at various temperatures. The yields of pyrolysis

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products depend on the type, nature and composition of the feedstock, particularly the lignin and ash contents, and process conditions such as temperature, pressure, vapour residence time, heating rates, particle size and heat integration (McLaughlin, 2010). Similarly, the composition, quality and characteristics of bio-char such as density, particle size distribution, ash content, moisture content and pH depend on the type, nature and origin of the feedstock, together with pyrolysis reaction conditions (Zhang *et al*., 2008).

Based on conditions, pyrolysis can be classified into three basic groups, namely, slow, intermediate and fast pyrolysis (Sohi *et al*., 2009; Brown, 2009; McLaughlin, 2010). As can be seen from Table 2.1, slow pyrolysis and intermediate pyrolysis both result in higher bio-char yields, while fast pyrolysis gives higher liquid (bio-oil) yields. Thus, to optimize the production of bio-char, slow pyrolysis and intermediate pyrolysis seem to be the most appropriate technology.

Table 2.1 The mean post-pyrolysis feedstock residues resulting from different temperatures and residence times (Verheijen, 2010).

Mode	Conditions	Liquid	Bio-char	Syngas
Fast pyrolysis	Moderate temp ~ 500℃, shot hot vapour residence time of ~ 1 second.	75 %	12 %	13 %
Intermediate pyrolysis	Moderate temp ~ 500℃, moderate hot residence time of 10 – 20 seconds.	50%	20%	30%
Slow pyrolysis (Carbonization)	Low temp ~ 400℃, very long solids residence time.	30 %	35 %	35 %
Gasification	High temp ~ 400℃, long vapour residence time.	5 %	10 %	85 %

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2.10.2 Carbon sequestration potential using bio-char

Carbon sequestration is the capture and subsequent storage of carbon to prevent it from being released to the atmosphere. Large amounts of carbon in bio-char may be sequestered in the soil for long periods estimated to be hundreds to thousands of years (Lehmann *et al*., 2006; Bracmork, 2010). The principle of using bio-char for carbon sequestration is related to the role of soils in the carbon (C) cycle. In the natural C-cycle, as plants pull carbon dioxide (CO_2) from the atmosphere, part of that carbon is built into the plants structure through the process of photosynthesis. When plants die, they sequester that embodied carbon into the soil, but most of the carbon is rather quickly released back into the atmosphere as CO_2 through plants respiration and soil microbiological activity. The relative amounts of CO_2 are more or less balanced and hence the process is said to be carbon neutral. Carbon neutral means that there is no net carbon added to the atmosphere other than what naturally occurs. Climate change is caused by net additions of carbon (carbon positive) to the atmosphere. These additions are primarily due to humans burning carbon-based fossil fuel stocks at an increasing rate over the past five hundred (500) years.

In the case of bio-char, the natural process of C-cycle is interrupted by capturing part of the biomass before it reaches the soil directly and using part (25%) for the production of bio energy and part for the production of bio-char. The biomass that is converted to energy (potentially in the form of heat, gas or liquid fuels) releases part of the carbon in the form of CO_2 back into the atmosphere in an assumed carbon-neutral process. The other part of the biomass is converted into bio-char and because of its stability sequesters all but 5% of the carbon in the soil and hence has the ability to provide a carbon negative source of energy. Carbon negative refers to the actual net reduction of carbon in the atmosphere.

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Large amounts of carbon in bio-char may be sequestered in the soil for long periods estimated to be hundreds to thousands of years (Lehmann *et al*., 2006; Ogawa *et al*., 2006; Woolf, 2008). This is because components of bio-char are proposed to be considerably more recalcitrant than soil organic matter (SOM). This means that bio-char allows carbon inputs into soil to be increased greatly compared to the carbon output through soil microbial respiration, and this is the basis behind bio-chars possible carbon negativity and hence its potential for climate change mitigation.

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CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Description of study area

3.1.1 Location

The field experiment was carried out at the University of Ghana, Soil and Irrigation Research Centre (SIREC) – Kpong which is located within the lower Volta basin of the Coastal Savannah agro-ecological zone of Ghana (Fig 1). The 1,024 ha (2,560 acres) Research Centre is located at latitude 6° 09 N, longitude 00° 04’ E and at an altitude of 22 m above mean sea level.

3.1.2 Climate

The rainfall distribution pattern within a year is bimodal with mean annual rainfall of 1136.4 mm (coefficient of variation = 24% and a standard deviation from the mean = 276 mm). About 60 % of the total amount (682 mm) occurs in the major season, and 30 % (342 mm) in the minor season. Thus, the remaining 10% (113 mm) occurs during the off-season. The major growing season begins from March to mid-July and the minor season from early September to mid-November. There is high intensity of rainfall during the months of May to June and October.

The mean air temperature is 27.2 °C with mean maximum and minimum temperatures of 33.3 °C and 22.1 °C respectively. The relative humidity for the night time to the early hours of the day ranges from 70 to 100 % throughout the year. The afternoon relative humidity ranges from 20 to 65 % during the year.

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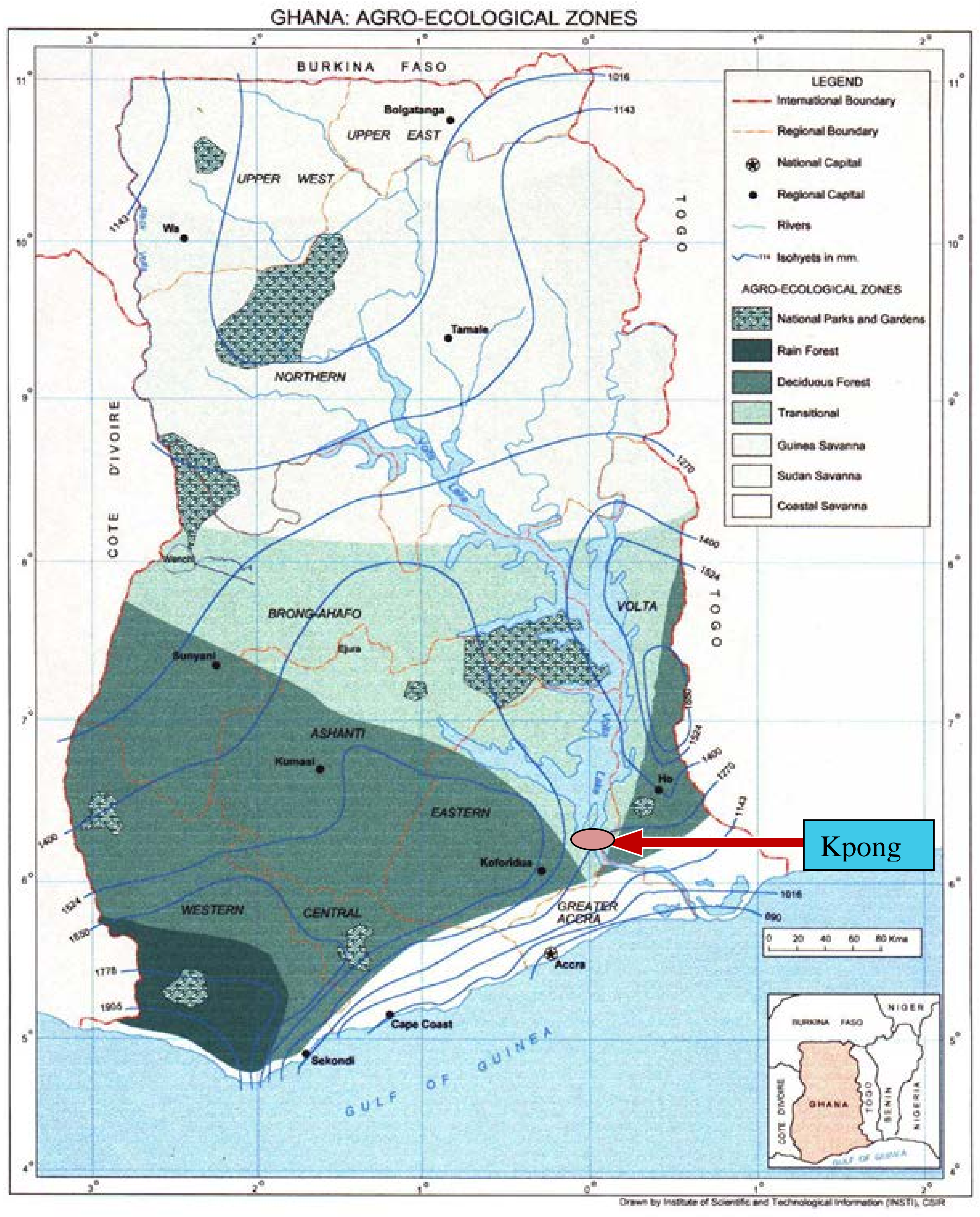

Fig. 3. 1 Map of Ghana showing study area: Soil and Irrigation Research Centre – Kpong.

3.1.3 Soils

The soil of the area is alluvial material derived from the weathering of garnetiferous hornblende gneiss. It is classified as Typic calciustert (Amatekpor *et al*., 1993). Locally, it is the Tropical Black Clay called Akuse Series (Amatekpor *et al*., 1993) which is categorized as Vertisols (Brammer, 1967). By definition, Vertisols are deep black soils that contain more than 30 % clay which is often dominated by smectite mineralogy (Soil Survey Staff, 2006). Generally, the clay content is very high in Vertisols and the dominant clay minerals are 2:1 type minerals (Smectite, montmorillonites). These clay minerals have the outstanding feature to expand (swell) when wet and shrink when dry. Therefore, pronounced changes in volumes with changes in soil moisture result in deep cracks in the dry season and very plastic, swelling and sticky soil consistency when wet. The chemical and physical properties of the soils are given in Table 4.1 and 4.2 respectively.

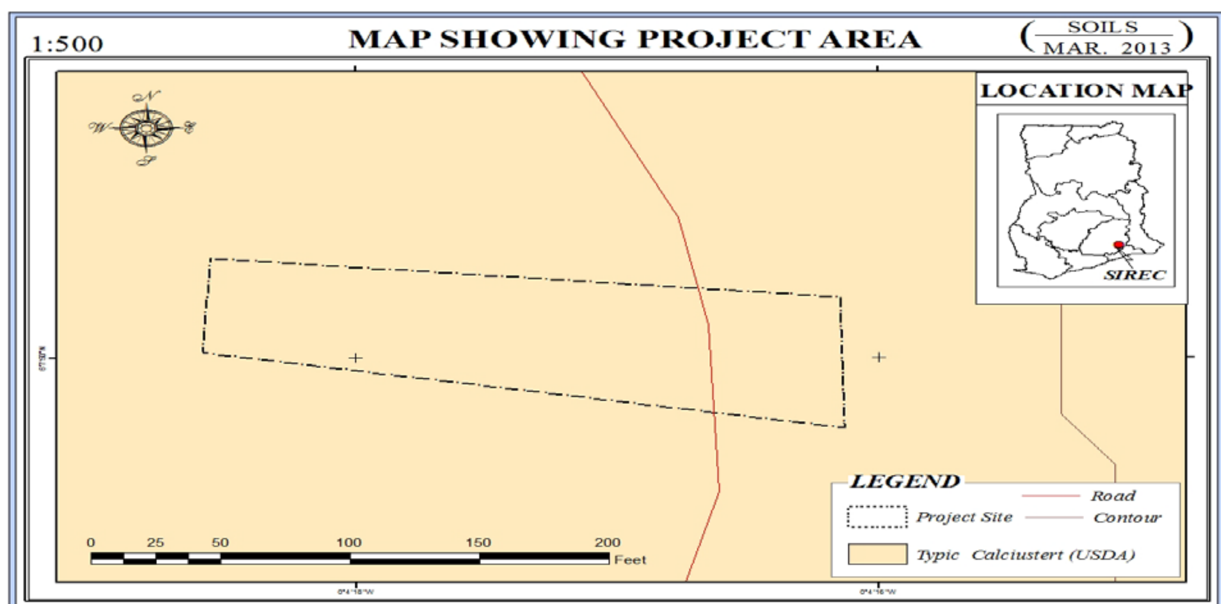

Fig. 3. 2 Map showing the location of the experimental field and the type of soil within the study area (Typic calciustert, Amatekpor et al., 1993)

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3.1.4 Vegetation

The natural vegetation types are, to a certain extent, limited by the soil properties such as high clay content, shrink-swell characteristics, and soil structure. Both climate and soil properties limit the vegetation types to grasses and slow growing, deep-rooting tree species. The main features of the natural vegetation in these soils are tolerance to drought, as well as development of deep roots to overcome root damage as a consequence of the annual cracking.

3.1.5 Topography

The general topography of the area is gently sloping with slopes ranging from 1 to about 5%.

3.2 Experimental procedure and treatment application

3.2.1 Field experiment

A paddy field basin labelled plot 16 was used for the experiment. The basin was 76 m × 14.5 m (1102 m^2). Preparation of the plot area involved removal of rice stubble, ploughing to a depth not exceeding 30 cm and construction of bunds in each plot after puddling to prevent contamination or flooding from adjacent plots.

3.2.1.1 Experimental design

The experiment was designed as a split plot with soil amendment as main plot treatment and levels of N fertilization as sub-plot treatment. The main plots were (control, bio-char rice husk and raw rice husk. The subplots treatments were 0, 45, 90 and 120 kg N ha^{-1} applied as urea. The subplots were replicated three times and completely randomized in each main plot. Each subplot measured 3.5×4.5 m. Details on treatment are indicated in Table 3.1.

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Table 3. 1 Description of treatment names for the field experiment

Soil amendment	Abbreviation	Fertilizer level (kg N ha^{-1})	Treatment name
Control	C	0	C0N
Bio-char rice husk	BRH	0	BRH0N
Rice husk	RH	0	RH0N
Control	C	45	C45N
Bio-char rice husk	BRH	45	BRH45N
Rice husk	RH	45	RH45N
Control	C	90	C90N
Bio-char rice husk	BRH	90	BRH90N
Rice husk	RH	90	RH90N
Control	C	120	C120N
Bio-char rice husk	BRH	120	BRH120N
Rice husk	RH	120	RH120N

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3.2.1.2 Application of soil amendments

The bio-char used in this study was produced from rice husk by the batch process or technology. This process simulates the traditional charcoal making process which uses pits, earth mound, brick and metal kiln as reactor types. A metal barrel which measured 0.85 m high and 0.6 m in diameter was used as the metal kiln (burning compartment). One end of the circular top part of the barrel was sealed while at the other end, a circular hole was made in the middle to accommodate firewood (source of heat). Few holes each about 0.01 m diameter were punched all around the barrel. The rice husk was gathered around the barrel and as the firewood burned, this acted as a shield against oxygen by insulating the carbonizing rice husk materials closer to the holes against excessive loss of heat. Thermal decomposition of the rice husk commenced when the firewood burned considerably. The charring husk was stirred thoroughly. The final product which is a dark-black material was evenly spread and sprinkled with water, after which it was dried.

Bio-char and rice husk were applied and incorporated in the top soil (0-20 cm). The amendments applied to the surface of the soil were mixed thoroughly to a depth of about 0 – 15 cm until a top soil homogenization was observed. Bio-char was applied at a rate of 10 t ha^{-1} and for the rice husk a 40 % loss in weight after charring was factored in calculating the weight. Reports from several studies indicate that bio-char is effective at application rates beyond 10 t ha^{-1}. The rice husk is normally used as a feed stock. It is readily available in rice growing communities and is regarded as nuisance and hence burnt to ashes.

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3.2.1.3 Seedling preparation and crop establishment

Rice (*Oryza sativa*) seeds were provided by the Soil and Irrigation Research Centre, Kpong. Clean seeds (KRC Baika variety) of uniform size and weight were selected. The seeds were soaked in water for 24 hours and sowed into nursery beds. The three weeks (21 days) old seedlings were transplanted to each subplot. Seedlings were transplanted at three per hill and thinned to 2 at a planting distance of 20 cm × 20 cm, giving a total density of 700 plants per plot. The field study was carried out in 2012 (Aug-Dec) and 2013 (Jan-May)

3.2.1.4 Fertilizer application

Four levels of urea (N-fertilizer) (0, 45, 90 and 120 kg/ha) were applied as split (50 %). The basal application was applied one week after transplanting of seedlings together with Triple Superphosphate (TSP) (30 kg P_2O_5 ha^{-1}) and muriate of potash (35 kg K_2O ha^{-1}) by broadcasting uniformly onto experimental plots. The second split (50 %) (Top dressing) was applied prior to panicle initiation (i.e. 6 weeks after transplanting).

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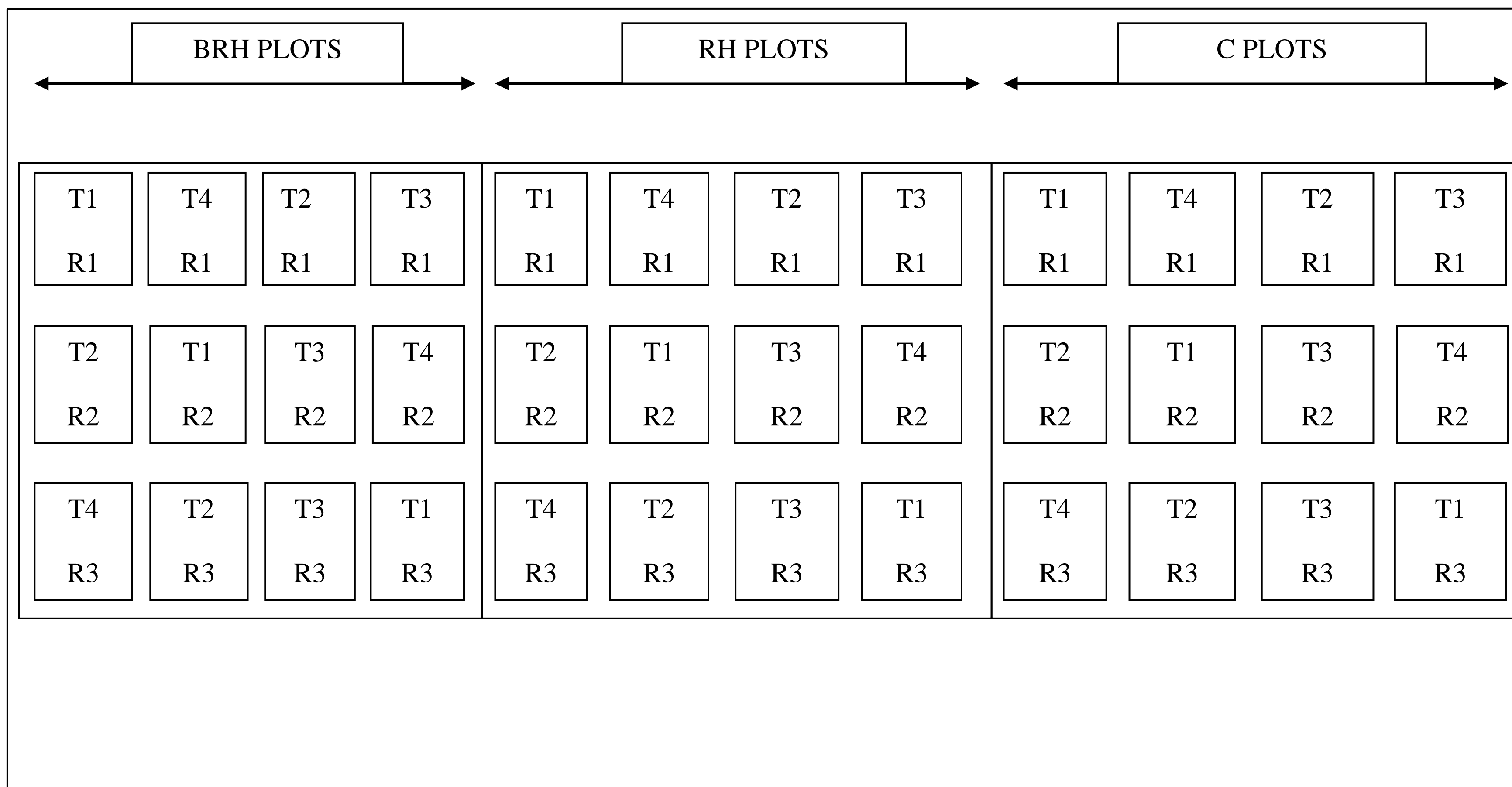

Fig. 3. 3 Layout of the experimental field (Split plot design)

T1 – TREATMENT 1 (0 kg N/ha), T2 – TREATMENT 2 (45 kg N/ha), T3 – TREATMENT 3 (90 kg N/ha), T4 – TREATMENT 4 (120 kg N/ha). R1 – REP 1, R2 – REP 2, R3 – REP 3. BRH – BIO-CHAR RICE HUSK, RH – RICE HUSK, C - CONTROL

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3.2.2 Pot experiment

Apart from the field studies pot experiment was conducted in a screen house of the Soil and Irrigation Research Centre, Kpong between January and May 2013. Plastic pots of height 33.8 cm and diameter 31.3 cm were used for the experiment (Plate 2). A total of thirty six pots were used. The bottoms of the pots were drilled to allow water to drain out.

3.2.2.1Soil sampling and filling of pots

The experimental soils were collected at 0 – 20 cm depth from an uncultivated field that has had no history of bio-char, rice husk and fertilizer use. The soils were subsequently air dried and roots and other plant materials removed, crushed and sieved through 2 mm size mesh to obtain fine earth fraction. Twenty kilograms (20 kg) of the soil was packed into the plastic pots to attain the field bulk density 1.4 M gm^{-3}.

3.2.2.2 Experimental layout and treatment application

The pot experiment was set out in a Completely Randomized Design (CRD) with three factors; 3 types of soil amendment, 2 fertilizer levels and 2 moisture regimes. Details of treatment are shown in Table 3.2. These treatments were replicated three (3) times to give thirty six (36) experimental pots.

The amendments applied to the surface of the soil were mixed thoroughly by hand to a depth of about 0 – 5 cm until a top soil homogenization was observed. Bio-char was applied at a rate of 10 t ha^{-1}. For the rice bio-char, a 40 % loss in weight after charring was factored in calculating the weight applied to the surface of soil.

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Rice seeds were provided by the Agricultural Research Institute, Soil and Irrigation Research Centre, Kpong. Clean KRC Baika variety which was uniform in size and weight was selected for seedling establishment. These seeds were sown on nursery beds and transplanted into experimental pots 21 days after emergence. Seedlings were transplanted at three per hill and thinned to two, giving a density of four plants per pot (770 cm^2).

Fertilizer levels consisted of 0 kg N/ha (N 0) and 90 kg N/ha (N 90) applied as urea. Urea was applied seven days after transplanting as split (50 %) for basal application. Urea was applied together with TSP (30 kg P_2O_5 ha^{-1}) and muriate of potash (35 kg K_2O ha^{-1}) by placement. Top dressing was applied twenty eight days after the basal application as split (50 %).

Two water treatments were also applied in the pot studies. In one of the water treatments W0, irrigation water was maintained with 0 cm head. For the second treatment W1, a 5 cm head of water was maintained after transplanting except during the water drainage period at maturity. Details on treatment are indicated in Table 3.2.

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Table 3. 2 Description of treatment names for the pot experiment.

Soil amendment	Abbreviation	Fertilizer level (kg N ha^{-1})	Water treatment	Treatment name
Control	C	0	W0	CN0W0
Bio-char rice husk	BRH	0	W0	BRHN0W0
Rice husk	RH	0	W0	RHN0W0
Control	C	0	W1	CN0W1
Bio-char rice husk	BRH	0	W1	BRHN0W1
Rice husk	RH	0	W1	RHN0W1
Control	C	90	W0	CN90W0
Bio-char rice husk	BRH	90	W0	BRHN90W0
Rice husk	RH	90	W0	RHN90W0
Control	C	90	W1	CN90W1
Bio-char rice husk	BRH	90	W1	BRHN90W1
Rice husk	RH	90	W1	RHN90W1

Plate 3. 1 The experimental set-up for the screen house study.

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3.2.3 Measurements and analyses

3.2.3.1 Measurement of soil CO_2 efflux

The gas entrapment method by Hutchinson and Mosier (1981) and Sullivan et al., (2008) was used in this study. For the field study transparent PVC chambers were inverted 2 cm into the soil (Plate 2). A 10 ml solution of 3 M NaOH was dispensed into a vial and placed under the plastic chamber to trap CO_2 evolved from the soil. Additional vials containing 10 ml of 3 M NaOH placed in the transparent PVC with their lids on to exclude CO_2 evolved from the soil served as controls to account for the CO_2 trapped from the atmosphere. The trapping solutions were changed every 4 hours during a day's measurement i.e. 6am – 10am, 10am – 2pm and 2pm – 6pm. After exposure of the alkali, the vials were removed, covered with lids (air tight seal) immediately and taken to the laboratory for analysis.

For the pot experiment, the trapping solutions were changed every 3 hours during a day's measurement i.e. 6 – 9 am, 10 – 1 pm and 2 – 5 pm. In each of the pot and field experiments the first soil CO_2 efflux measurement was taken immediately following transplanting without fertilization and at daily intervals for the first seven days following the application of the first and second doses of urea. Thereafter, measurements were taken weekly until maturity.

It should be noted that whereas it is desirous to also determine other GHGs such as CH_4 and N_2O emissions, the lack of the appropriate equipment such as Gas Chromatograph compelled this study to focus only on CO_2, for which a simple methodology could be applied.

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Plate 3. 2 Layout of field experiment showing chambers for trapping effluxed CO_2.

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Plate 3. 3 Layout of pot experiment showing chambers for trapping CO_2.

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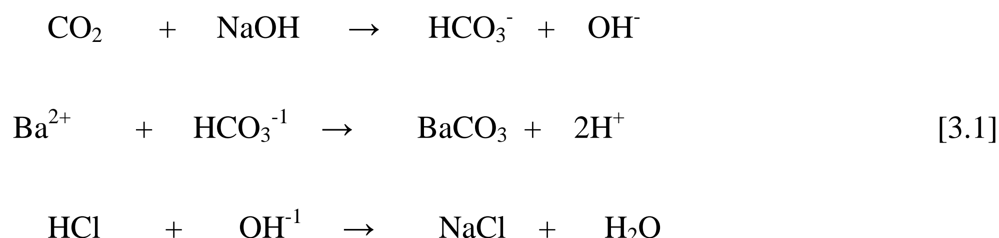
3.2.3.2 Laboratory analysis of soil CO_2 efflux

The alkali solutions from both the experimental plots and the controls were brought to the laboratory to determine the amount of CO_2 evolved from the soil. The evolved CO_2 was determined by back titration. Excess NaOH in solution was back titrated against 1.0 M HCl using phenolphthalein indicator after precipitating the carbonate/bicarbonate formed with 1.0 M $BaCl_2$.

3.2.3.3 Chemical reaction and computation of evolved soil CO_2

At the end of each reaction, the 1.0 M $BaCl_2$ added precipitated the carbonate as insoluble $BaCO_3$. Each mole of CO_2 dissolved in the NaOH leads to the production of 2 moles of H^+ after addition of the $BaCl_2$. Thus each mole of dissolved CO_2 results in the neutralization of 2 moles of OH^-. The reactions that occurred were:



$$CO_2 + NaOH \rightarrow HCO_3^- + OH^-$$

$$Ba^{2+} + HCO_3^{-1} \rightarrow BaCO_3 + 2H^+ \qquad [3.1]$$

$$HCl + OH^{-1} \rightarrow NaCl + H_2O$$

The HCl neutralizes the remaining OH^-, so that the amount of CO_2 trapped in the NaOH can be calculated as:

$$\text{mmol } CO_2 \text{ trapped in the NaOH} = 0.5 \times (\text{original mmol } OH^- - \text{ mmol HCl added}) \qquad [3.2]$$

Taking into account the amount of CO_2 trapped in the blank, this becomes:

$$\text{mmol } CO_2 \text{ trapped in the NaOH} = 0.5 \times (\text{ml HCl}_{blank} - \text{ml HCl}_{sample}) \times M_{HCl} \qquad [3.3]$$

where, M_{HCl} is the molarity of the HCl used in the titration.

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The amount of CO_2 trapped (mmol) in equation [3.3] is converted to mg by multiplying it by the molar mass of CO_2 (44 $gmol^{-1}$). This amount of CO_2 (mg CO_2) measured by the opened circular area of the chamber (0.1111m^2) in contact with the soil is expressed per m^2 per 4 hours of the sampling time (mg m^{-2}/4hrs).

3.2.3.4 Soil and flood-water measurements

Soil and floodwater pH and temperature were measured between 9-10 am, 1-2 pm and 5-6 pm for the first seven days following the application of the first and second doses of urea and thereafter weekly until maturity for the pot experiment. Temperature was measured with a pH/mV/℃ meter (RS232) with their probes placed in the floodwater and soil at 5 cm (Plate 3.4). Daily ambient air temperature and precipitation data were obtained from the weather data station at Soil and Irrigation Research Centre, Kpong.

Plate 3. 4 Measurement of soil and floodwater temperature using pH/°C/mV meter.

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3.2.3.5 Analysis of soil amendments (Bio-char and Rice husk)

The rice husk and bio-char used in the study as soil amendment were air-dried, crushed and passed through a 2 mm sieve. Composite sample of the sieved rice husk and bio-char was taken for laboratory analysis of pH, total P, K, available P and CEC. Total C and N values from literature were reported in Table 4.2.

3.2.3.6 Soil sampling and analysis

Prior to the application of soil amendments, soil samples were taken by auguring at depths of 0–0.15 m, 0.15–0.3 m and 0.3–0.45 m at four random positions in each of the main plots. Air-dried samples were crushed and sieved through a 2 mm sieve for characterization. The procedure was repeated before the beginning of the second season and at the end of the two cropping seasons. Samples of the soil used in the pot experiment were also taken for physical and chemical characterization.

3.2.4 Characterization of soil (Physical properties)

3.2.4.1 Particle size analysis

The particle size distribution was determined by the modified Bouyoucous hydrometer method described by Day (1965). Soils from 0 – 15 cm, 15 – 30 cm and 30 – 45 cm depth were analysed for clay, sand and silt content. Forty grams (40 g) of the 2 mm sieved soil was weighed into a beaker and 60 ml of 6 % H_2O_2 was added to oxidize the organic matter. The content was transferred into a dispersion cup and mixed with 100 ml of 5 % Calgon solution (Sodium hexametaphosphate). The suspension was shaken and transferred into a settling cylinder and was made up to the 1000 ml mark with distilled water. The suspension was agitated vigorously with a plunger and the time noted immediately shaking was stopped. The temperature of the suspension

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was recorded after equilibration. A hydrometer (ASTM 15 2H) was then placed into the suspension and the first and second readings noted after 5 minutes and 5 hours respectively. The suspension was then poured directly onto a 0.5 mm sieve and the particles retained on the sieve washed thoroughly with water and dried in an oven at 105 °C for 24 hours. The dried samples were then weighed to represent the sand fraction. The particle size distribution was then determined using the following formulae:

$$\text{i) Silt \%} + \text{Clay \%} = \frac{\text{5 minutes hydrometer reading}}{\text{weight of soil}} \times 100 \quad [3.5]$$

$$\text{ii) Clay \%} = \frac{\text{5 hour hydrometer reading}}{\text{weight of soil}} \times 100 \quad [3.5]$$

$$\text{iii) Silt \%} = \%\,(\text{silt} + \text{clay}) - \%\,(\text{clay}) \quad [3.6]$$

$$\text{iv) Sand \%} = \frac{\text{oven dry mass (g) of particles retained on the 0.5 mm sieve}}{\text{weight of soil}} \times 100 \quad [3.7]$$

The textural classes were determined using the texture triangle.

3.2.4.2 Determination of bulk density

Cylindrical soil cores (3 replicates) were taken from the depths 0 – 15 cm, 15 – 30 cm and 30 – 45 cm. The cores were weighed and dried in an oven at 105 °C for 24 hours. The oven dried mass (M_s) of the samples were also taken. The volume (V_t) of soil which is the product of cross-sectional area and height (h) of soil cores was determined by computation from measured internal diameter (d) and height (h) of the cylindrical core sampler. The bulk density (ρ_b) was calculated using the formula:

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$$\rho b = \frac{Ms}{Vt} \qquad [3.8]$$

3.2.5 Characterization of soil (Chemical properties)

3.2.5.1 Soil pH (1:1)

Twenty grams of the soil sample was weighed into a 50 ml beaker. An amount of 20 ml distilled water was then added to make the ratio (1:1). The soil suspension was then stirred for 30 minutes. The suspension was then allowed to stand for an hour to allow the entire suspended particles to settle. A glass electrode pH meter was standardized with two aqueous solutions of pH 4 and 7. The pH of the prepared suspension was then measured by the glass electrode.

3.2.5.2 Organic carbon

Organic carbon was determined by the wet combustion method of Walkley and Black (1934). This method involves the reduction of the $Cr_2O_7^{2-}$ ion by the organic matter and the un-reduced $Cr_2O_7^{2-}$ measured by titration with ammonium sulphate. The quantity of organic matter oxidized is calculated from the amount of $Cr_2O_7^{2-}$ reduced. A 10 ml of 1M potassium dichromate ($K_2Cr_2O_7$) solution and 20 ml of concentrated sulphuric acid (H_2SO_4) were added to 0.5 g of soil in a conical flask and digested for 2 hours. The K_2CrO_7 remaining in solution after the digestion was titrated against 0.2 M ferrous ammonium sulphate using barium diphenylamine sulphonate as the indicator to a green end point. The titre values were used to calculate the % C from the formula below:

$$\% \text{ C} = \frac{0.3[10\text{-}(XN)]1.33}{W} \qquad [3.9]$$

Where,

X = titre value of ferrous ammonium sulphate, N = molarity of ferrous ammonium sulphate,

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W = weight of soil, 0.3 is the milliequivalent weight of carbon and 1.33 is the correction factor for 25% un-oxidized total carbon due to the wet digestion procedure.

3.2.5.3 Total nitrogen

Two grams of air-dried soil sample was weighed into a 250 ml Kjeldahl flask followed by addition of digestion accelerator, selenium catalyst and 5 ml of concentrated sulphuric acid (H_2SO_4). The mixture was allowed to digest until the digest was clear. It was then allowed to cool and then transferred with distilled water into a 50 ml volumetric flask and made up to the volume. A 5 ml aliquot was pipetted from the digest into a distillation flask and 5 ml of 40 % sodium hydroxide (NaOH) was added with 100 ml distilled water. The sample was then distilled and collected in 5 ml of 2 % boric acid to which about 2 drops of methylene blue indicator had been added. The distillate was then titrated against 0.01 N HCl (Bremner, 1965) from green to a red end point.

The amount of N (%) was calculated as shown below:

$$\% \text{ N} = \frac{\text{Molarity of HCl} \times \text{titre volume} \times 0.014 \times \text{vol.of extractant}}{\text{Weight of soil sample} \times \text{volume of aliquot}} \times 100 \qquad [3.10]$$

3.2.5.4 Available phosphorus

Available P of the soil was determined using the method Watanabe and Olsen (1965). Ten grams of the fine earth fraction was weighed into extraction bottles. A 100 ml sodium bicarbonate (pH 8.5) was added to the samples in the extraction bottles; it was then capped and shaken for 30 min on a mechanical shaker. The extracts were filtered using Whatman's No. 125 filter paper to obtain clear solution. A 10 ml aliquot of the extract was taken into a test tube and then in drop-wise, 1 ml of 1.5 M H_2SO_4 was added to decolourise the solution by settling the

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organic matter in it. It was then left in a refrigerator to cool for few minutes. The extracts in the test tubes were centrifuged, and gently decanted for colour development and phosphorus analysis. The concentration of P in the extracts was then determined using the Murphy and Riley method (1962) described as followed. An aliquot of 1mL of the sample solution was pipetted into a 50 ml volumetric flask and a drop each of P-nitrophenol and ammonium hydroxide were added. Then, 8 ml of a solution containing concentrated sulphuric acid, ammonium molybdate, potassium antimony tartrate, and ascorbic acid were added. The content was topped up to the 50 ml mark with distilled water. The concentration of phosphorus was then determined on a Philips' UV spectrophotometer at a wavelength of 712 nm. Available phosphorus content of the soil was calculated as;

$$P\ (mg\ kg^{-1}) = \frac{\text{Spectrometer reading} \times \text{total volume of extract}}{\text{Volume of aliquot} \times \text{weight of soil sample}} \qquad [3.11]$$

3.2.5.5 Determination of cation exchange capacity (CEC)

Ten grams of soil sample was weighed into an extraction bottle and 100 ml of 1 M ammonium acetate solution added. The bottle with its content was shaken for 30 minutes on a mechanical shaker. The content was filtered through a No. 42 Whatman filter paper and the sample leached four times with 25 ml of methanol to wash off excess ammonium. Thereafter another 25 ml of 1 M acidified potassium chloride was used to leach the soil four times. A 5 ml aliquot of the leachate was taken into a Markham distillation apparatus and 5 ml of 40 % NaOH solution added to distil. The distillate was collected into 5 ml of 2 % boric acid to which three drops of methyl red and methylene blue indicators were added. The distillate was back titrated against 0.01 M

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HCl to purplish end point. The cation exchange capacity in soil was then calculated from the number of moles of HCl consumed in the back titration.

3.3 Characterization of rice husk and rice husk bio-char

3.3.1 Determination of bio-char bulk density

An amount of the air-dried bio-char sample was carefully packed amidst intermittent tapping on the laboratory bench (to ensure good packing) into a measuring cylinder to a pre-determined volume (Vt). The quantity of sample packed was then transferred into a moisture can and put in an oven at 105 °C to dry for 48 hours after which its mass (M_s) was recorded. The dry bulk density (ρ_b) of the bio-char was then calculated from the relation:

$$\rho b = \frac{Ms}{Vt} \qquad [3.12]$$

3.3.2 pH of bio-char and rice husk

The pH values of the bio-char and the rice husk were determined using a Pancitronic MV 88 pH glass electrometer. The pH of each of the materials was determined in distilled water at a 1:10 ratio (Rice husk or bio-char to water). Two grams each of air-dried material were weighed into a 100 ml beaker. Twenty millilitres of distilled water were added and the rice husk or bio-char-liquid suspension was stirred for 30 minutes and allowed to equilibrate at room temperature. The pH meter was then standardized using buffer solution at pH 4.0, 7.0 and 9. The electrode was placed in the suspension and recorded as pH in water.

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3.3.3 Organic carbon of bio-char

Organic carbon was determined by the wet combustion method of Walkley and Black (1934). Ten millilitres of 1 M potassium dichromate ($K_2Cr_2O_7$) solution and 20 ml of concentrated sulphuric acid (H_2SO_4) were added to 0.1 g of each of particle size less than 0.5 mm rice husk bio-char and the uncharred raw rice husk into a conical flask. The flask was swirled to ensure full contact of the materials with the liquids, after which the mixture was allowed to stand for 4 hours to ensure complete digestion.

The unreacted $K_2Cr_2O_7$ in solution after the oxidation of the oxidizable organic material in the bio-char or the rice husk was titrated against 0.2 M ferrous ammonium sulphate solution after adding 10 ml of orthophosphoric acid and 2 ml of barium diphenylamine sulphonate indicator from dirty brown colour to bright green end point. A standardization titration of the $K_2Cr_2O_7$ with the ferrous ammonium sulphate was done and the amount of organic carbon calculated by subtracting the number of moles of unreacted $K_2Cr_2O_7$ from the number of moles of $K_2Cr_2O_7$ present in the standardized titration.

The concentration of oxidizable carbon in each of the samples was calculated indirectly from the number of moles of unreduced dichromate consumed by the ammoinium ferrous sulphate.

3.3.4 Available phosphorus determination

One gram of air-dried rice husk bio-char was weighed into an extraction bottle and 100 ml sodium bicarbonate (pH, 8.5) was added. The bottle was then capped and shaken for 30 min on a mechanical shaker. The extracts were filtered using Whatman's No. 125 filter paper to obtain clear solution. A 10 ml aliquot was taken into a test tube and then 1 ml 1.5 M H_2SO_4 added drop-wise to decolourise the solution by settling the organic matter in it. It was then left in a refrigerator to cool for few minutes. The extracts in the test tubes were centrifuged, and gently

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decanted for colour development using the Murphy and Riley method (1962) as described in section 3.2.5.4.

3.3.5 Determination of Total Phosphorus.

Total phosphorous was determined by digesting 0.2 g of bio-char with 25 ml of a mixture of concentrated HNO_3 and 60 % $HClO_4$ in the ratio of 2:3. Distilled water was added to the digest, filtered and made up to volume in a 100 ml volumetric flask with distilled water. Phosphorus in the digest was then determined as described by Murphy and Riley (1962) method.

3.3.6 Determination of Total Nitrogen

Total nitrogen was determined by the modified Kjeldahl digestion method (Bremner, 1965). The nitrogen in the 0.2 g sample was converted to ammonium by digestion with concentrated sulphuric acid using the catalyst selenium. The ammonium formed was then determined by distilling the digest with a strong alkali (40 % NaOH) and titration with a standard acid as outlined in section 3.2.5.3.

3.3.7 Cation exchange capacity of the rice husk bio-char

Ten grams of rice husk bio-char were weighed into an extraction bottle and 100 ml of 1 M ammonium acetate solution added. The CEC of the material was then determined using the method outlined in section 3.2.5.5.

3.4 Statistical analysis

Microsoft Excel (version 2010, Microsoft Corporation) and Genstat statistical software (Genstat version 9.2) were used to analyze the experimental data. Microsoft excel was used for data entry,

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analyses and subsequent export into Genstat where appropriate. All analyses were performed on data collected on two field seasons and four months pot experiment. Analysis of variance was performed on soil CO_2 efflux rates on each sampling dates separately. Regression analysis was also used to determine the relationship between CO_2 effluxes rates and temperature. Statistical differences were considered significant at $p \leq 0.05$. In addition, the statistical package Statistix version 9.0 was used to test differences in means using Tukey range test procedure at a significance level of $p \leq 0.05$.

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CHAPTER FOUR

4.0 RESULTS

4.1 Initial soil characteristics

Tables 4.1 and 4.2 summarize information on the physical and chemical characteristics of the paddy field prior to commencement of the experiment. Clay content of the soil increased with depth. Bulk density increased with depth from 1.40 to 1.60 Mg m^{-3} for 0-0.15 to 0.3-0.45 m.

The soil pH can be described as neutral with pH values ranging from 6.6 in the top soil and increasing down the profile to 7.1 at the 0.3-0.45 m depth. Organic carbon content was relatively high in the top soil (8.14 g kg^{-1}) and decreased down the profile to 5.95 g kg^{-1} in the 0.3–0.45 m depth. This may be due to accumulation of rice straw overtime from previous season's cultivation. Total nitrogen values were low with highest value occurring on the top soil and decreasing down the profile. Total phosphorus was 24.1 % within the first 0.3 m but decreased to 20.65 % at 0.3-0.45 m depth. This trend was consistent with that of available phosphorus with low value of 4.53 mg kg^{-1} decreasing down the profile to 3.48 mg kg^{-1} at 0.3-0.45 m depth.

The Cation Exchange Capacity (CEC) values were very high with values of 37.1 and 38.1 cmol kg^{-1} for 0-0.15 and 0.15-0.3 m depths respectively and decreased with depth to 34.7 cmol kg^{-1} for 0.3-0.45 m soil depth.

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Table 4. 1 Soil chemical properties of study site before land preparation for 1st experiment.

Soil depth	pH (1:2)	S.O.C	Total N	Total P	Avail P	CEC
(m)	Soil: H_2O	------------------- %	------------------------		(mg kg^{-1})	---- cmol kg^{-1} ---
0 – 0.15	6.6	0.82	0.06	24.10	4.53	37.1
0.15 – 0.3	7.1	0.67	0.05	24.11	4.25	38.1
0.3 – 0.45	7.1	0.59	0.05	20.65	3.48	34.7

Table 4. 2 Soil physical properties of study site before land preparation for 1st experiment.

Soil depth	Bulk density	Sand	Silt	Clay	Textural class
(m)	(Mg m^{-3})	---------------- %	-----------------		
0 – 0.15	1.40	47.1	5.4	47.6	Sandy clay
0.15 – 0.3	1.60	40.7	2.6	56.7	Clay
0.3 – 0.45	1.60	38.5	2.6	58.9	Clay

S.O.C – Soil organic carbon; N - Nitrogen; P – Phosphorus; CEC- cation exchange capacity

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4.2 Characteristics of the soil amendments used (rice husk and rice husk bio-char)

Table 4.2 summarizes the physical, elemental composition and pH of the rice husk and bio-char used. In this study, the rice husk from which the bio-char was produced had organic carbon (OC) content of 216 g kg^{-1} (21.6 %). Its total nitrogen content was 0.5 g kg^{-1} (0.05 %) and this gave a carbon to nitrogen ratio (C:N) of 432. The pH of rice husk was 5.9 which could be described as slightly acid. Total P and K in rice husk were 0.3 and 3.8 g kg^{-1} respectively whiles its bulk density was 0.13 Mg m^{-3}.

The organic carbon (OC) content of bio-char was 56 g kg^{-1} (5.6 %). Total N in bio-char was 0.8 g kg^{-1} (0.08 %). Nguyen *et al.,* (2013) reported 464 g kg^{-1} (46.4 %) and 0.4 g kg^{-1} (0.04 %) as total C and N respectively of rice husk bio-char and this accounted for a C: N ratio of 1160.

The cation exchange capacity (CEC) of bio-char was 20.3 cmol kg^{-1}. Cation exchange variation in bio-char ranges from negligible to around 40 cmol kg^{-1} (Lehmann, 2007). Nguyen *et al*., (2013) reported a CEC of 5.7 cmol kg^{-1} for bio-char rice husk that was produced at temperatures of 350–500 °C by thermal pyrolysis. Bio-char pH values are relatively homogenous (Verheijen *et al*., 2010), largely neutral to basic. In this study, the pH of bio-char was 6.9 which could be described as neutral. Total P and K in bio-char was 0.6 and 5.3 g kg^{-1} respectively. The bio-char used in this study had a bulk density of 0.22 Mg m^{-3}.

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Table 4. 3 Chemical and physical properties of the soil amendments

	Soil amendments	
Property	Rice husk	Bio-char
BD (Mg m^{-3})	0.13	0.22
pH (H_2O)	5.9	6.9
O.C (g kg^{-1})	216	56 (464*)
Total N (g kg^{-1})	0.5	0.8 (0.4**)
C: N ratio	432	70 (1160***)
Total K (g kg^{-1})	0.38	0.53
Total P (g kg^{-1})	0.03	0.06
CEC (cmol kg^{-1})	-	20.3

BD- Bulk density,

*, **, ***: adapted from Nguyen *et al.* (2013).

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4.3 Rainfall and air temperature patterns during the field study period

The major environmental factors that influence the flux of CO_2 from the soil surface in the field are temperature and moisture (Sullivan *et al*., 2009). Figure 4.1 shows the maximum air temperature records and rainfall events at the study site during the experimental period. Rainfall for the first study period, Season I (Aug-Dec 2012) and Season II (Jan-Apr 2013) were 464.60 mm and 70.12 mm respectively. Most of the rainfall was concentrated in Season I than Season II.

The monthly maximum temperature during the season ranged from 29.98 to 33.50 °C in August-December 2012 and from 33.30 to 35.92 °C in January-April 2013. Higher temperatures were observed in Season II than Season I.

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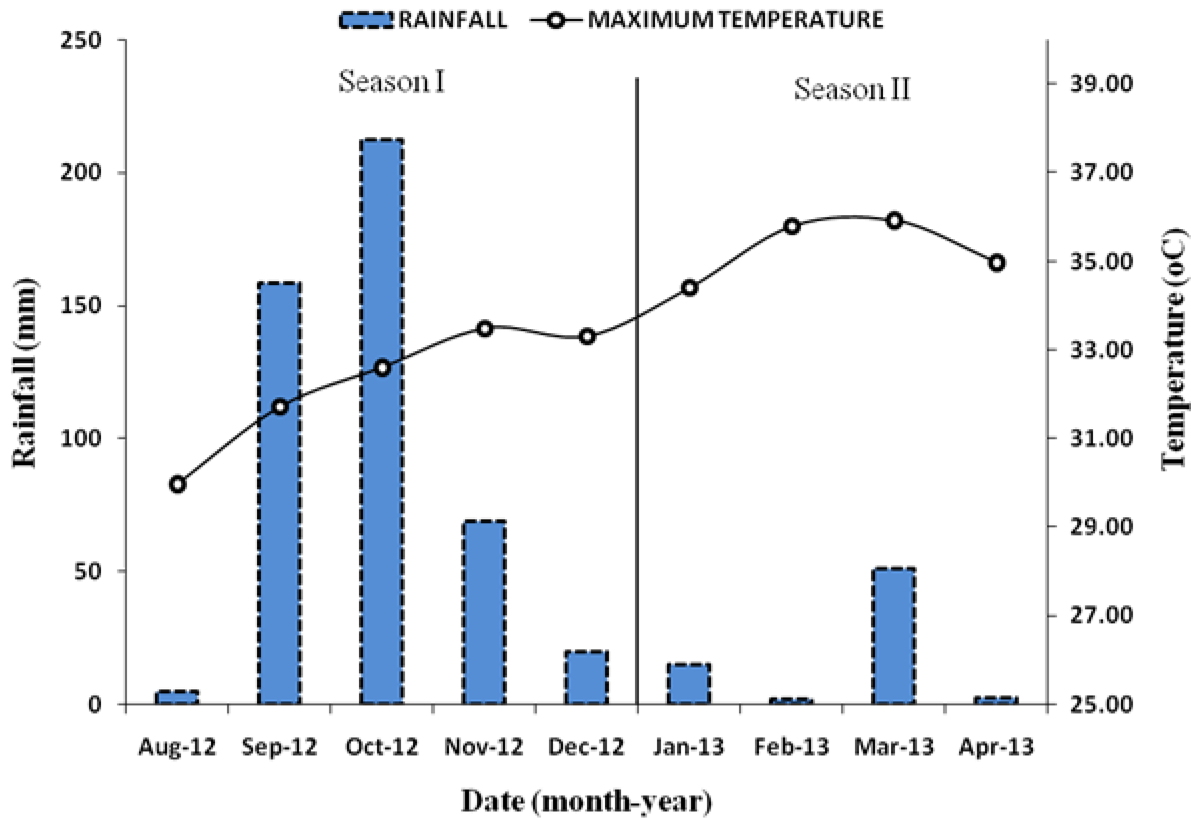

Fig. 4. 1 Monthly rainfall (mm) and temperature (°C) during the field study (vertical solid line separates the two seasons).

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4.4 Variations in soil CO_2 effluxes from the field studies

4.4.1 Season I

In the Season I, the initial CO_2 efflux after puddling was 500 mg $m^{-2}(4hrs)^{-1}$ with a peak efflux observed during mid-afternoon (10–2 pm) measuring 680 mg $m^{-2}(4hrs)^{-1}$ and lowest efflux occurring in the morning (6-10am) with soil CO_2 efflux of 380 mg $m^{-2}(4hrs)^{-1}$ (Fig 4.2).

Amendment of the paddy field initially decreased soil CO_2 efflux. Control (C) plots recorded low CO_2 emissions which were not statistically significant ($p<0.05$) from the CO_2 effluxes from bio-char (BRH) and rice husk (RH) amended plots (Fig 4.3). However, the slightly higher soil CO_2 effluxes recorded from bio-char and rice husk amended plots may be due some initial decomposition after incorporation (Fig 4.3).

One week after basal fertilizer application (two weeks after transplanting, WAT) clear seasonal variations were observed in weekly soil CO_2 effluxes from different treatments in Season I (Aug-Dec) (Fig 4.4). Soil CO_2 efflux increased in all the treatments. Soil CO_2 efflux decreased 4 WAT and this was sustained to the fifth week. Measurements taken during morning and late-afternoon showed significant differences ($P < 0.05$) among treatments. Rice husk amended plots fertilized with 120 kg N/ha urea showed the highest soil CO_2 efflux of 501.7 mg $m^{-2}(4hrs)^{-1}$ in the morning whilst the lowest soil CO_2 efflux of 36 mg $m^{-2}(4hrs)^{-1}$ was observed in late-afternoon from bio-char amended plot which received no fertilization (Fig 4.4).

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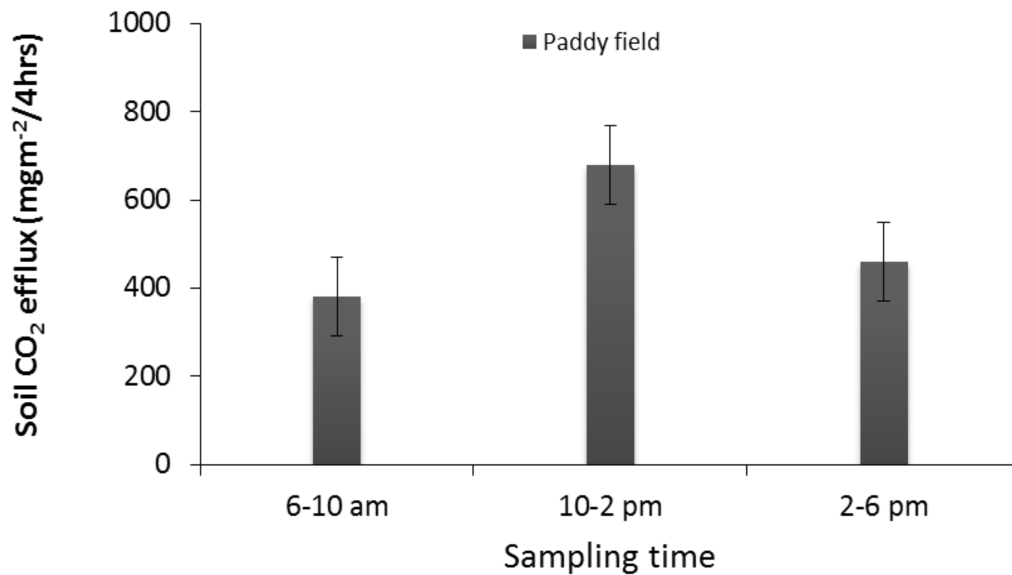

Fig. 4. 2 Initial soil CO_2 efflux from paddy fields two days after puddling the field during Season I. The vertical bars are standard errors of the means.

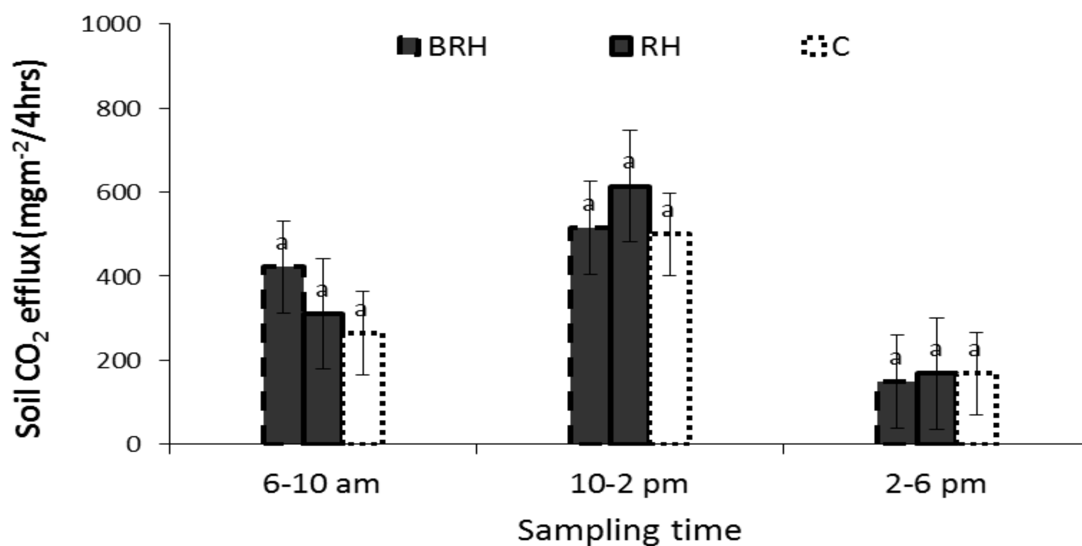

Fig. 4. 3 Soil CO_2 efflux one week after paddy plots were amended with rice husk bio-char (BRH) and rice husk (RH) as soil amendment in season I.

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Top dressing was applied during the 6th week after transplanting (WAT), the highest average soil CO_2 efflux of 390 mg $m^{-2}(4hrs)^{-1}$ was observed from RH90N followed by BRH120N with CO_2 efflux of 385.6 mg $m^{-2}(4hrs)^{-1}$ in late-afternoon. Bio-char decreased soil CO_2 efflux with the lowest soil CO_2 efflux average value of 108.4 mg $m^{-2}(4hrs)^{-1}$ measured for BRH0N. This was followed by BRH120N with 114.4 mg $m^{-2}(4hrs)^{-1}$ CO_2 efflux. A gradual increment in soil CO_2 efflux was observed 2 weeks after top dressing (7 WAT). Soil CO_2 efflux increased to 1000 mg $m^{-2}(4hrs)^{-1}$ from RH120N treatment in the morning. The lowest soil CO_2 efflux was also reported from BRH0N with a value of 30 mg $m^{-2}(4hrs)^{-1}$ in the morning. This trend of CO_2 efflux was maintained 12 WAT. Rice husk plot fertilized with 120 kg N/ha urea recorded the highest CO_2 efflux of 661.1 mg $m^{-2}(4hrs)^{-1}$, whereas BRH0N recorded the lowest soil CO_2 efflux of 117.2 mg $m^{-2}(4hrs)^{-1}$ for measurements taken throughout the day.

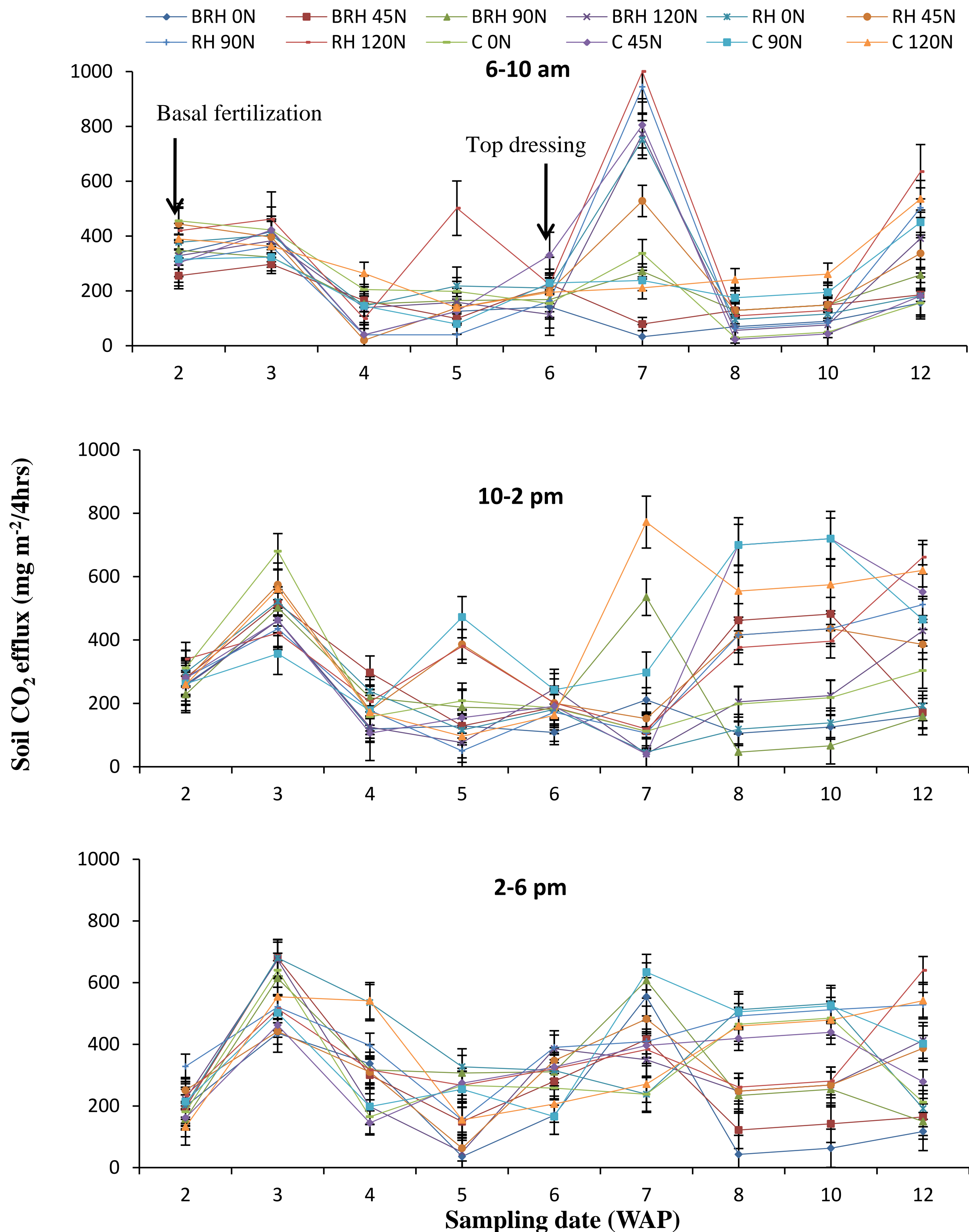

Fig. 4. 4 Changes in soil CO_2 effluxes from different treatments in Season I. (Vertical arrows denote fertilizer application dates. Treatment abbreviations are explained in Table 3.1 in chapter 3.

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4.4.2 Season II

Soil CO_2 effluxes measured in Season II (Fig 4.5), were generally low compared to Season I. One week after basal fertilization (2 WAT) averaged soil CO_2 effluxes based on 7 days daily measurement after basal fertilization also showed decreased CO_2 efflux compared to Season I. Rice husk amended plot which received 90 kg N/ha urea (RH90N) reported the highest CO_2 efflux of 436.6 mg $m^{-2}(4hrs)^{-1}$ followed by 399.8 mg $m^{-2}(4hrs)^{-1}$ from C120N. Low CO_2 effluxes were observed from treatments which received no fertilization particularly bio-char amended plots. The lowest CO_2 efflux (129.2 m $gm^{-2}(4hrs)^{-1}$) was observed from BRH0N.

Two weeks after basal fertilization (3 WAT) soil CO_2 efflux increased in the afternoon, RH90N recorded a maximum CO_2 efflux of 607.3 mg $m^{-2}(4hrs)^{-1}$ followed by 495.1 mg $m^{-2}(4hrs)^{-1}$ from BRH 45N. Treatments showed significant differences ($p<0.05$) in soil CO_2 efflux. Similar pattern of soil CO_2 efflux was observed during 4 and 5 WAT.

Top dressing was applied 6 WAT (Fig 4.5) as shown by the solid arrow. On the average, RH90N treatment recorded 414 mg $m^{-2}(4hrs)^{-1}$ as the highest soil CO_2 efflux. The lowest efflux (119.7 m $gm^{-2}(4hrs)^{-1}$) was observed from BRH 120N in the morning and this showed significant differences ($P < 0.05$) among treatments.

Two weeks after top dressing (7 WAT) C90N and RH90N showed sharp increases in soil CO_2 efflux in mid and late-afternoon respectively. Carbon dioxide efflux rose from 396 mg $m^{-2}(4hrs)^{-1}$ 6 WAT to 501.7 mg $m^{-2}(4hrs)^{-1}$ 7 WAT from C90N while soil CO_2 efflux from RH90N rose from 158.4 to 640 m $gm^{-2}(4hrs)^{-1}$. Low emissions were recorded in the morning from all the treatments particularly C45N recording the lowest soil CO_2 efflux of 66 mg $m^{-2}(4hrs)^{-1}$.

Soil CO_2 efflux increased gradually 10 WAT. Rice husk amended plots showed higher CO_2 efflux for all the treatments than control plots. Bio-char amended plots recorded lower CO_2 efflux

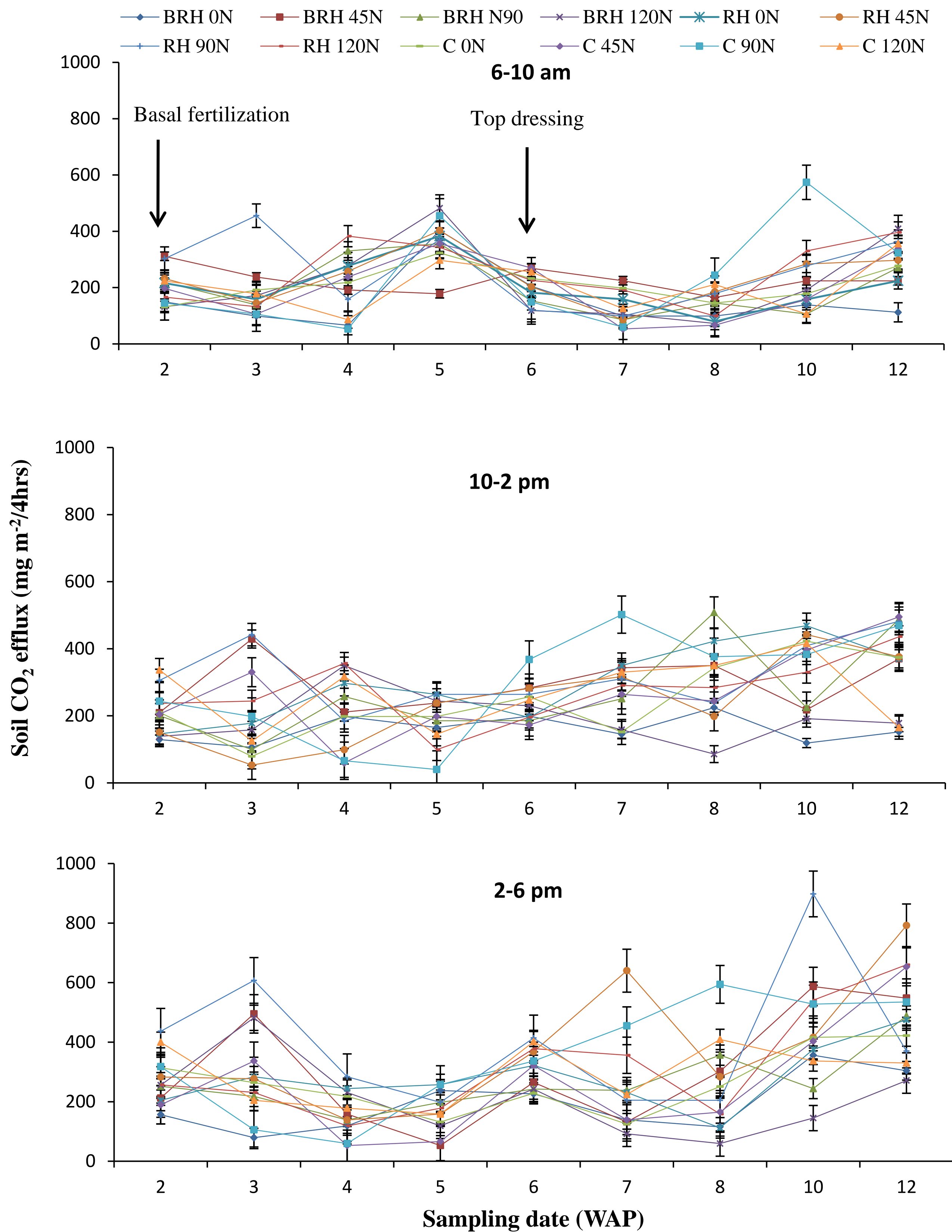

Fig. 4. 5 Changes in soil CO_2 effluxes from different treatments in Season II.

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compared with the other soil amended plots. In the late-afternoon, CO_2 efflux from RH90N peaked at 898 mg $m^{-2}(4hrs)^{-1}$. Bio-char amended plot with 120 kg N released 145 mg $m^{-2}(4hrs)^{-1}$ which was the lowest for 10 WAT. Bio-char amended plots recorded low CO_2 efflux 12 WAT. The highest soil CO_2 efflux (792 mg $m^{-2}(4hrs)^{-1}$) was observed from RH45N followed by RH120N with a CO_2 efflux of 660 mg $m^{-2}(4hrs)^{-1}$. The lowest CO_2 efflux of 112 mg $m^{-2}(4hrs)^{-1}$ was observed in the morning from BRH0N followed by BRH0N in mid-afternoon with 152 mg $m^{-2}(4hrs)^{-1}$ soil CO_2 efflux. Soil CO_2 efflux was significantly different ($p<0.05$) among treatments except measurements taken in the morning.

4.5 Dynamics of CO_2 effluxes from the pot experiment

The dynamics of average weekly CO_2 efflux over twelve weeks of the pot experimental study is summarized in Table 4.4. Average weekly soil CO_2 efflux ranged from 209.1 to 438.7 mg $m^{-2}(3hrs)^{-1}$ during the experimental study period. The results showed that soil CO_2 efflux was higher in fertilized soils than unfertilized soils. Lower CO_2 effluxes were also observed from bio-char amended soils than those amended with rice husk. The net effluxes of CO_2 from the pots were lower when the soil had 0.05 m water head than when it was kept at saturation. Generally, highest CO_2 effluxes were observed in the afternoons and lowest emissions in the mornings.

The highest CO_2 efflux (446.7 mg $m^{-2}(3hrs)^{-1}$) was observed for RHN90W0 during late-afternoon (2-5 pm). This was followed closely by 438.7 and 403 mg $m^{-2}(3hrs)^{-1}$ for CN90W0 and BRHN90W0 respectively during the same measurement duration. The bio-char amended treatment recorded the lowest CO_2 emission among the three and this was significantly different ($p<0.05$). The treatment CN90W1 recorded the lowest CO_2 emission of 290.1 mg $m^{-2}(3hrs)^{-1}$. Its corresponding

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unfertilized treatment, CN0W1 recorded 271.4 mg $m^{-2}(3hrs)^{-1}$ which was relatively higher but showed non-significant differences ($p<0.05$).

Comparing the influence of water head on CO_2 emission in unfertilized plots BRHN0W0 recorded the lowest emission of 311.7 mg $m^{-2}(3hrs)^{-1}$ in the morning. Its corresponding flooded treatment, BRHN0W1 showed 2 % decrease in CO_2 emission but did not decrease the emission significantly ($p<0.05$). For RHN0W0 and CN0W0, 3 % and 4 % respective reduction in CO_2 emission over their corresponding flooded treatments were observed but the emissions were non-significant ($p<0.05$). In mid-afternoon, BRHN0W1 and CN0W1 reduced CO_2 emission significantly ($p<0.05$) over BRHN0W0 and CN0W0 respectively. All the flooded treatments showed significant ($p<0.05$) reduction in CO_2 emission over their corresponding saturated treatments.

In the case of the fertilized pots, all the flooded treatments showed significant ($p<0.05$) reduction in CO_2 emission over their corresponding saturated treatments.

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Table 4. 4 Average weekly soil CO_2 efflux (mg m^{-2}/3hrs) for pot experiment

	Soil CO_2 efflux (6-9 am)			Soil CO_2 efflux (10-1 pm)			Soil CO_2 efflux (2-5 pm)		
Treatment	BRH	RH	C	BRH	RH	C	BRH	RH	C
N0W0	311.7cde	277.5bcd	354.7cde	383.4abc	385.3ab	384.9ab	359.6c	389.4b	381.7b
N0W1	296.6e	306.3de	299.6de	314.0d	352.8bcd	330.5d	255.5g	301.2de	271.4fg
N90W0	349.1b	376.1a	354.2b	395.0a	413.8a	395.8a	403.0b	446.7a	438.7a
N90W1	307.1de	324.9c	314.7cd	342.4cd	350.5bcd	345.2bcd	280.7ef	307.9d	290.1def
	L.S.D (0.05) = 17.90			L.S.D (0.05) = 41.02			L.S.D (0.05) = 21.80		

Soil CO_2 efflux values followed by common letters are not significantly different ($p<0.05$) according to Tukey's mean separation test.

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4.5.1 Soil temperature

The soil temperature measurements at 0.05 m depth (Fig 4.6) exhibited patterns similar to soil CO_2 effluxes i.e. increased soil temperature was observed with increased soil CO_2 efflux and vice versa. The soil temperature varied from 23.17 to 39.6 °C during the whole rice growing period from January to April, 2013. Lower temperatures were observed in the morning ranging from 23.17 to 35.7 °C. This was followed by higher temperatures in the late-afternoon that varied from 29.6 to 36.63 °C. Highest temperatures were observed in the mid-afternoon where temperatures ranged from 31.7 to 39.63 °C.

Two weeks after transplanting, treatment BRHN90W0 recorded the lowest temperature of 23.17 °C in the morning. This was followed by RHN0W1 with a temperature of 25.25 °C while CN90W1 reported the highest temperature of 28.55 °C. Temperature increased sharply from 2 to 3 WAT with BRHN0W0 showing the highest temperature of 35.7 °C while the lowest temperature of 33.3 °C was observed from RHN90W1 treatment. All the treatments further saw a steep decline in temperature 5 WAT. Soil temperature increased gradually 7 WAT and was maintained until 12 WAT in all the treatments. During 7 WAT, the treatment CN90W1 reported 33.95 °C as the highest temperature whiles RHN0W1 recorded the lowest temperature value of 30.25 °C. Twelve weeks after transplanting, RHN0W1 further declined to 29 °C recording the lowest temperature in the morning whereas CN90W0 increased gradually from 31 °C in 7 WAT to 31.6 °C as the highest soil temperature at the end of week twelve in the morning.

In the mid-afternoon (10-1pm) CN90W0 recorded the lowest temperature (33.7 °C) at 2 WAT and increased gradually to 39.37 °C at the end of the fourth week (Fig 4.7b). The highest soil temperature of 35.63 °C was reported by BRHN90W0 at 2 WAT which also increased gradually to 37.5 °C at the end of the fourth week. Soil temperature declined gradually from week four to week six and thereafter, increased to 7 WAT. Temperature dropped at 8 WAT and increased gradually at 11 WAT. Thereafter, all the treatments recorded a decreasing trend until maturity. At 7 WAT,

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BRHN0W0 recorded the highest soil temperature of 39.25 °C and this was followed by CN90W0 with a soil temperature of 38.4 C. At the end of maturity (12 WAT), CN90W0 showed the highest temperature of 35.8 °C whiles RHN0W1 recorded the lowest temperature (33.2 °C).

In late-afternoon (Fig. 4.6c), at low soil temperature of 30.95 °C was recorded by RHN90W0 at 2 WAT whereas RHN0W1 reported 33.3 °C as the maximum soil temperature. All the treatments reported a gradual increase in soil temperature trend until 4 WAT and thereafter, decreased in week five. The highest soil temperature of 36.37 °C during the late-afternoons measurement was recorded in 4 WAT for CN90W0. During this week, BRHN90W1 reported a minimum soil temperature of 33.07 °C. Soil temperature increased gently from 5 to 7 WAT. During week seven, soil temperature varied from 32.3°C to 35°C as observed from treatments RHN0W0 and RHN90W0 as the minimum and maximum soil temperature respectively. Generally, soil temperature decreased gently from 7 to 8 WAT and further increased gently to week twelve at maturity. Temperature peaked at 33.7°C for CN90W0 12 WAT whereas, RHN0W1 recorded a minimum soil temperature of 31.1 °C.

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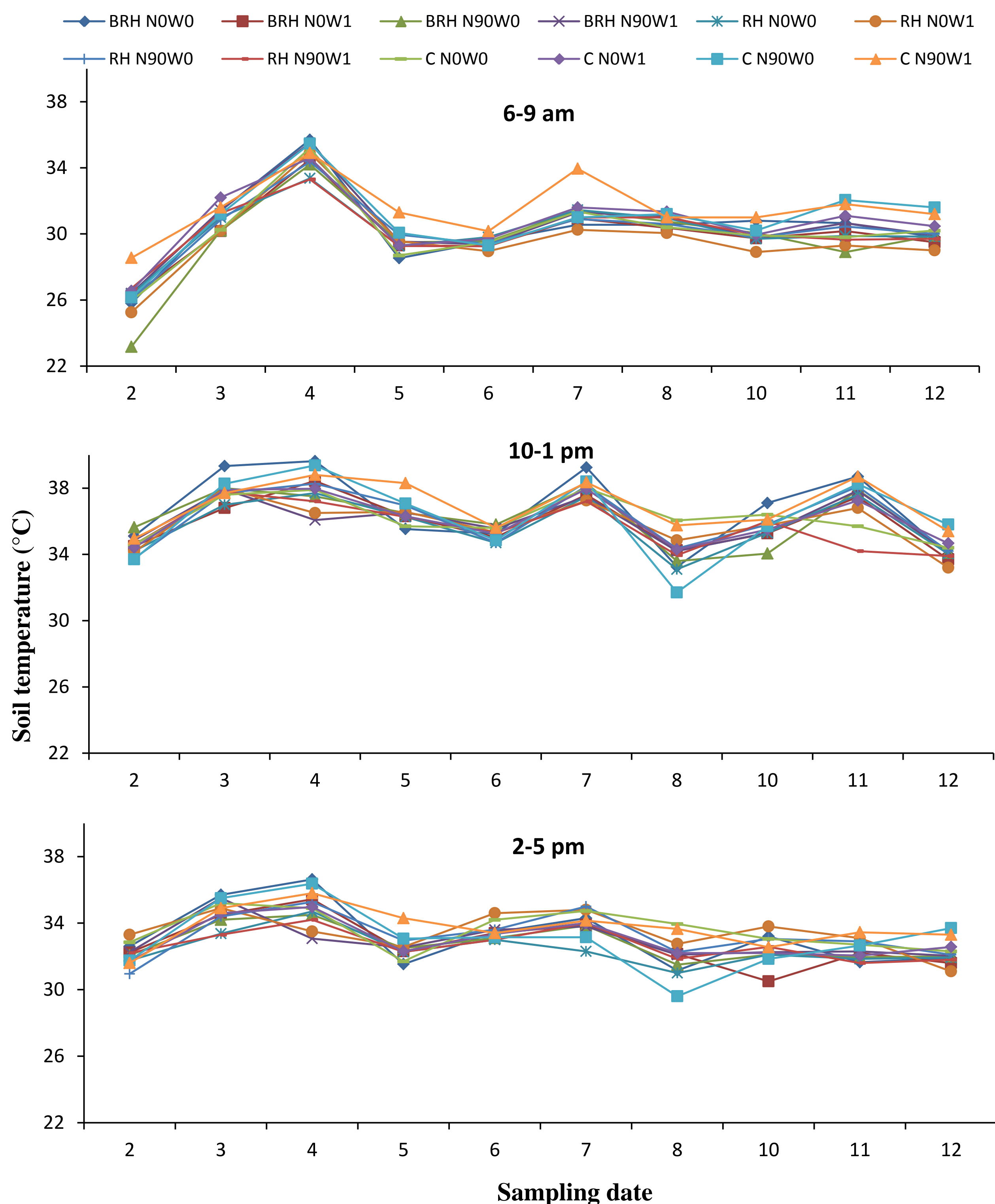

Fig. 4. 6 Variation of soil temperature in the pot experiment.

(BRH: Bio-char rice husk, RH: Rice husk, C: Control, N0 and N90 are N fertilizer levels at 0 and 90 kg N ha^{-1} respectively, W0 and W1 are moisture regimes at saturation and flooding respectively).

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CHAPTER FIVE

5.0 DISCUSSION

5.1 Characteristics of the initial soils

The clay content of Akuse series was high and increased with depth. The sand and silt content were low. The amount of sand and silt decreased with depth. Using the textural triangle, Akuse soil may be described as sandy clay for the top soil and clay beyond 0.15 m under the USDA system. This is consistent with Brammer (1967) classification as clay for Akuse. Bulk density was low on the top soil but increased with increased depth.

The pH of Akuse series is described as neutral with values ranging from 6.6 on the top soil and increased with depth to 7.1 for 0.03 to 0.45 m. The almost neutral pH (6.9) of the soil (Amatekpor *et al.,* 1993) might not have much influence on volatilisation of ammonia fertilizer applied to it (Brammer, 1967). This is because ammonia volatilisation has been shown to increase substantially as soil pH increases above 7 (Koelliker and Kissel 1988).

The organic carbon of Akuse soil is low (Quaye *et al*., 2009). This is a characteristic of most tropical savannah soils. The soil organic carbon decreased with depth. The relatively higher organic carbon observed for the top soil would be due to accumulation of rice straw over time from previous season's cultivation.

Total nitrogen content of Akuse soil was consequently low. The nitrogen content of Vertisols in the Accra plains is generally low (0.133 g kg^{-1}). The range of total nitrogen reported for this soil is between 0.07 g kg^{-1} and 0.193 g kg^{-1} (Abunyewa, 1997; Amatekpor *et al.,* 1993; Acquaye and Owusu-Bennoah, 1989). Vertisols elsewhere have also been found to have low nitrogen concentration

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(ICRISAT, 1984; Mohanty *et al.,* 2007). One would therefore expect positive crop response to nitrogen application on these soils provided there is adequate soil moisture.

The cation exchange capacity (CEC) of Akuse soil was high. This is consistent with the fact that it has high clay content which is mostly montmorillonitic. The CEC value of 28.47 cmol kg^{-1} for the Akuse soil was reported by Amatekpor *et al.,* (1993).

5.2 Characteristics of the soil amendments (Bio-char and rice husk)

The pH of bio-char was neutral whereas that of rice husk was described as slightly acid. Incorporation of bio-char is expected to increase the soil pH over time as bio-char is reported as a good liming material (Verheijen *et al*., 2010).

Bio-char composition is highly heterogeneous, containing both stable and labile components (Sohi *et al.,* 2009). The organic carbon (OC) content of bio-char was 56 g kg^{-1} (5.6 %). Chang and Xu (2009), reviewed the total carbon content of different feedstock's and found its carbon content to range between 172 to 905 g kg^{-1} (17.2 to 90.5 %), although OC accounts for < 500 g kg^{-1}. In a similar analysis, Nguyen *et al.* (2013) obtained 464 g kg^{-1} as the total carbon content of rice husk bio-char. The Walkley and Black (1934) procedure for determining OC was used in this study to determine OC content of the bio-char. This procedure only accounted for the oxidizable C and not the total C content of bio-char and was employed due to lack of facility. In general, bio-char may not support microbial activity due to the refractory nature of C and thus, can present a long-term C sink in soil (Kuhlbusch, 1998 and Rumpel *et al.,* 2006). However, some studies have shown that a small labile part of the bio-char is utilized by microbial processes (Zimmerman, 2010).

Nguyen *et al.,* (2013) reported a C:N ratio of 1160 for bio-char produced from rice husk. This high C:N ratio will impede the capacity of bio-char to release inorganic N when incorporated into the soil. In this study, the C:N ratio of bio-char was not reported because its total C was not accounted for.

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However the total C of bio-char is expected to be higher than the rice husk due to its lower oxidizable carbon.

The bulk density of bio-char was low (0.22 Mg m^{-3}). The bulk density of the 0-0.15 m of the paddy soil was 1.37 Mg m^{-3}. It is therefore expected that the incorporation of bio-char can reduce the overall bulk density of the soil (Verheijen *et al*., 2010).

Total exchangeable potassium was higher in bio-char than the raw rice husk used. The total phosphorus of bio-char was about 50 % higher than the rice husk. Cation exchange capacity variation in bio-char ranges from negligible to around 40 cmol kg^{-1} (Lehmann, 2007). Nguyen *et al*., (2013) reported a CEC of 5.7 cmol kg^{-1} for bio-char rice husk that was produced at temperatures of 350–500 °C by thermal pyrolysis. The CEC of bio-char used in this study was 20.3 cmol kg^{-1}.

5.3 Seasonal emissions of CO_2 in field studies

5.3.1 Amendment effects

The relatively higher soil CO_2 efflux observed after puddling compared to those before puddling may be due to increased surface tillage which opened up soil voids for CO_2 emission. Tillage systems may affect soil biological, chemical and physical properties and therefore influence the release of CO_2 (Robertson *et al*., 2000). Increased surface roughness and larger voids produced by soil disturbance has been shown to increase soil CO_2 efflux (Reicosky *et al*., 2007; Ball *et al*., 1999).

The average weekly soil CO_2 efflux from the bio-char, rice husk and control amended plots over Season I and Season II are summarized in Table 5.1. In Season I, CO_2 efflux from bio-char amended plots (BRH) were the least (204.8 mg $m^{-2}(4hr)^{-1}$) followed by control (C) plots (250.8 mg $m^{-2}(4hr)^{-1}$) with highest emissions from rice husk (RH) amended plots (309.9 mg $m^{-2}(4hr)^{-1}$). Soil amendments significantly ($p<0.05$) influenced CO_2 emission. In the mid-afternoon, soil CO_2 efflux from BRH plots increased by 13.3 % to 236.2 mg $m^{-2}(4hr)^{-1}$ but the CO_2 efflux change for RH in the mid-afternoon

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was marginal. In mid-afternoon, emissions from control plots increased by 30.8 % over measurements in the morning. Again, all reported soil CO_2 effluxes from all the soil amended plots were significantly different ($P<0.05$) from each other.

The results indicated that bio-char reduced soil CO_2 emission compared with rice husk and control plots. This point to the work done by Wang *et al*. (2011) who reported that CO_2 emissions in paddy soil following a second 30-day incubation period were significantly reduced by bio-char addition. This depression effect of CO_2 emission during the second incubation may have derived from bio-char carbon as suggested by Major *et al*. (2010). In a similar study, Liu *et al*., (2011) reported a 91 % reduction in CO_2 emissions when a paddy soil was amended with bio-char pyrolized at 600 °C.

Knoblauch *et al.* (2008) established a contrasting result in which charred rice residues had no effect on the carbon mineralization or CO_2 emissions of paddy soil over about two years of oxic incubation. Other studies have also reported that bio-char addition stimulated the initial evolution of soil CO_2 (Kuzyakov *et al.,* 2009; Smith *et al.*, 2010; Wang *et al.,* 2011). The results of this study however suggested that emission rates even from bio-char amended soils differed seasonally with Season I emitting larger amounts of CO_2 than Season II. Zimmerman *et al*., (2011) observed that after the addition of bio-char, either the organic C in the soil, the added labile organic matter or the bio-char C could be lost simultaneously (Lehmann and Sohi 2008; Luo *et al*., 2011) for a short time (Nguyen *et al*., 2010). The initial CO_2 emission increase could be attributed to some bio-char mineralization reported by Smith *et al*., (2010). Usually, the bio-char decomposition rate is observed to be initially high due to the water-soluble carbon (Nguyen and Lehmann 2009) and decreased to approximately 1 % of the initial decomposition rate and remained constant during the second and third years of laboratory incubation (Kuzyakov *et al*., 2009).

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Jones *et al.* (2011) and Wardle *et al.* (2008) also observed that the ''priming effect'' by enhanced decomposition of existing organic matter or humus by bio-char addition is responsible for the initial high CO_2 emission after addition of bio-char. Thus, initial CO_2 release by bio-char addition is due both to (i) mineralization to labile-C added through bio-char and (ii) stimulation of microbial activity. Consequently, higher initial decomposition of soil organic matter by bio-char addition is expected. Therefore, though bio-char addition tended to initially increase CO_2 evolution for a relatively short-term, it could serve as a long-term C sequestration acting as a recalcitrant C in the soil (Lehmann *et al*., 2011).

Rice husk amended soil emitted the largest amounts of CO_2. As shown in Table 4.2 rice husk had a high amount of oxidizable OC (216 g kg^{-1}) compared with bio-char (56 g kg^{-1}), much of which would have been in the labile pool. Labile C is generally considered not protected and is shown to be readily accessible to microbial mineralization. In effect high CO_2 emission would be expected from the rice husk amended plots as a result of microbial decomposition and this was confirmed in this study (Table 5.1).

The control treatments emitted CO_2 mainly from soil organic carbon (SOC). The field history indicated that the experimental plots had regularly been cultivated to paddy rice during the past 50 years before commencement of the field trials. Hence some accumulation of SOC would have occurred over time, especially in the upper soil layers. The quality of the SOC was not determined but would contain appreciable balance of both labile and recalcitrant portions. Hence the emission of CO_2 for control plots fell somewhat in-between that of BRH and RH. Zhang Xu-hui *et al*. (2006) suggested that C mineralization of paddy soils depends not only on the chemical lability of SOC (pool distribution), but also on the microbial metabolic activity and the soil N status. Although labile C may give significant contribution to the very actively mineralizable C, accumulation of younger or labile C does not necessarily enhance the carbon mineralization potential (Zhang Xu-hui *et al*., 2006). This

could be explained by the mutual interaction of C availability, accessibility to the protected labile C pools, and the metabolic activity of microbes which are influenced by soil nutrient and moisture regimes (Zhang Xu-hui *et al*., 2006).

Most of the rainfall that was recorded during the study period was concentrated in Season I and this influenced soil CO_2 efflux. An abrupt rise in near-surface soil moisture due to precipitation can cause an instantaneous soil respiration pulse (Maier *et al*., 2011; Lee *et al*., 2004). Soil respiration is shown to respond rapidly and instantaneously to the onset of rain and return to the pre-rain rate shortly after the rain stops (Lee *et al.,* 2004). The likely reason for this is that CO_2 is heavier than air and accumulates by gravitation within the air spaces of the soil. Replacement of this gaseous carbon by dilution will not occur without water and, unstirred by turbulent mixing, accumulation of CO_2 within the soil will increase. A sudden flooding might simply seal the soil pores, replace the captured CO_2 by water and release it back into the air (Chen *et al*., 2005). These occurrences, termed “Birch effect”, can have a marked influence on the ecosystem carbon balance (Birch, 1964; Unger *et al*., 2010).

5.3.2 Fertilizer application effects

Table 5.2 summarizes the soil CO_2 efflux pattern of the four different levels of N fertilizer during both seasons. In both seasons the 0 kg N ha^{-1} plots showed the least CO_2 emission of 219 and 189 mg $m^{-2}(4hrs)^{-1}$ for Seasons I and II respectively. This was followed by 45 kg N ha^{-1} fertilized plots with highest emissions from 120 kg N ha^{-1} fertilized plots in Season I. In Season II, highest emissions were observed for 90 than 120 kg N ha^{-1} fertilized plots.

The results indicated that N fertilization stimulated CO_2 emission. Van Zwieten *et al.* (2010) reported that CO_2 emission increased initially with application of N fertilizer probably due to CO_2 evolution from urea hydrolysis. Wilson and Al-Kaisi (2008) and Iqbal *et al.,* (2009) reported that

increase in CO_2 emissions were due to a positive effect of N fertilization on plant biomass. In a similar study, Dick (1992) suggested that soil microbial activity may increase because of N fertilization and thus increase CO_2 emission.

The results of this study also suggested that N fertilization, even at different levels had stimulative effect on CO_2 emission. Carbon dioxide emission increased with increasing levels of N. The trend of CO_2 emission was in the order $0 < 45 << 90 <<< 120$ kg N ha^{-1} for Season I and $0 < 45 << 120 <<< 90$ kg N ha^{-1} for Season II. Low CO_2 emission for recorded for 120 than 90 kg N ha^{-1} fertilized plots during Season II could be attributed to increased N fertilizer effect. Wilson and Al-Kaisi, (2007) reported that the high N rate of 270 kg N ha^{-1} depressed cumulative CO_2 emissions compared to the other two N rates (0 and 135 kg N ha^{-1}). They did not cite one specific mechanism for this depression in CO_2 emissions due to N fertilization, so it is relatively unclear as to why this happens. Fogg (1988) suggested that the ligninase enzyme, which is used to initiate lignin decomposition, is inhibited by increasing soil N concentrations. Fungal populations, which decompose lignin, and the activity of many other soil enzymes have been found to decreased with N additions (DeForest *et al.*, 2004). This hypothesis has been supported by work conducted by Burton *et al*. (2004) where they found that N fertilized plots averaged 15 % less soil CO_2 emissions than unfertilized plots. Similar to Fogg (1988) and DeForest *et al.*, (2004), Burton *et al*., (2004) suggest that there is some decrease in extracellular enzyme activity due to increase N application. In effect, nitrogen fertilization increased the respiration in soil-plant ecosystem by increasing the accumulation of biomass and improving the bioavailability of C to soil microbes (Table 5.2) which is in good agreement with previous results (Raich and Schlesinger 1992; Wang *et al*., 2012).

The interactive effect of BRH and N fertilizer suppressed CO_2 emission over RH and N fertilizer interactive effect in both seasons. In a study by Wang *et al.,* (2012) the addition of bio-char and N fertilizer did not increase the ecosystem respiration during either crop season in the upland soil, which

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was in accordance with Knoblauch *et al.,* (2008) who found that charred rice residues had no effect on the CO_2 emissions of paddy soil. In a similar study (Iqbal *et al*., 2009; Zhang *et al.,* 2011), the addition of bio-char and N fertilizer suppressed soil microbial respiration. The addition of N fertilizer stimulates residue decomposition (Wilson and Al-Kaisi (2007). However, the recalcitrance of C-organic in rice husk bio-char has been suggested by many researchers (Glasser *et al.,* 2002; Lehmann et al., 2003; Rondon *et al*., 2007). Rice husk as a residue material is expected to have a high C:N ratio (Table 4.3). The addition of N fertilizer will have a "priming effect" by enhanced decomposition of existing organic matter or soil humus by rice husk addition.

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Table 5. 1 Average Seasonal CO_2 efflux (mg m^{-2}/4hrs) under various amendments.

	Soil CO_2 efflux (6-10 am)				Soil CO_2 efflux (10-2 pm)				Soil CO_2 efflux (2-6 pm)			
Season	BRH	RH	C	LSD	BRH	RH	C	LSD	BRH	RH	C	LSD
I	205c	310a	351b	30.55	236c	293b	362a	25.92	294b	369a	349a	25.09
II	199b	237a	212bc	22.28	228c	287a	270b	5.63	264b	343a	293b	30.26

Table 5. 2 Average Seasonal CO_2 efflux (mg m^{-2}/4hrs) under different levels of N fertilizer.

	Soil CO_2 efflux (6-10 am)					Soil CO_2 efflux (10-2 pm)					Soil CO_2 efflux (2-6 pm)				
	0	45	90	120	LSD	0	45	90	120	LSD	0	45	90	120	LSD
I	219c	230bc	246b	327a	17.71	219b	328a	311a	331a	27.27	311b	317b	373a	347ab	41.79
II	189a	218a	231a	225a	31.96	237c	269b	294a	246c	16.40	257b	328a	340a	275b	16.17

Values in each column followed by common letters are not significantly different ($p<0.05$) according to Tukey's mean separation test.

BRH, RH and CONT are rice husk bio-char, rice husk and control respectively. 0, 45, 90 and 120 kg N ha^{-1} is N fertilizer levels.

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5.4 Soil CO_2 emission in pot experiments

5.4.1 Temperature effects

In many research studies, soil temperature was noted to be a strong and positive predictor of soil respiration, accounting for 43-75 % of the variation in soil CO_2 efflux rates (Weber, 1990; Toland and Zak, 1994; Londo *et al.* 1999; Ohashi *et al.* 2000; Bajracharya and Kimble, 2000; Tufekcioglu *et al.* 2001). The regression of soil temperature on soil CO_2 efflux showed a positive correlation with CO_2 evolution increasing as soil temperature increased (Fig 5.1).

Soil temperature explained 61 % of the total CO_2 efflux in the regression model. This strong relationship between soil efflux and temperature is expected since soil respiration rates reflect heterotrophic and autotrophic activities that are highly temperature dependent (Londo *et al.,* 1999). There was a close relationship between CO_2 emissions and soil temperatures, because the high emissions of CO_2 were detected at high soil temperatures (Fig. 4.6). The peaks of CO_2 emissions were observed at soil temperatures ranging from 38.3 to 38.5°C and 33.7 to 34.3°C (Fig 4.6). The highest soil CO_2 emission was measured at a soil temperature of 38.4°C (Fig 4.6). These observations are consistent with the result of Chang *et al.,* (2008), who found strong relationships between CO_2 efflux and soil temperature. Liu *et al.,* (2010), in a similar research, reported a significant ($p<0.01$) linear relationship between soil CO_2 efflux and soil temperature at a depth of 0.05 m.

The temperature sensitivity coefficient Q_{10} values are a convenient index for comparing the temperature sensitivity of soil CO_2 efflux. It is commonly used to express the relationship between soil biological activity and temperature, although Holland *et al.* (1995) has shown that estimates of global soil respiration are very sensitive to the selected Q_{10} value for various biomes. In this study, the Q_{10} was 1.91, implying that soil CO_2 efflux was almost doubled for a 10 °C increase in soil temperature which in effect indicates high sensitivity of soil CO_2 efflux to temperature. This is comparable to (1.4-

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4.29) reported for wetland soils (Zhang *et al.,* 2005) and (1.7-2.38) for subtropical paddy soils (Zhu *et al.,* 2005).

Drawing conclusions from variations in Q_{10} is difficult because our knowledge of the responses of microbial communities and plant roots to changes in soil temperature is limited (Boone *et al.,* 1998). However, the strong correlation between soil CO_2 efflux and soil temperature shows that soil temperature is an important factor controlling the major processes of C cycle (Knorr *et al*., 2005), which exhibits large amplitude in its effects on CO_2 fluxes.

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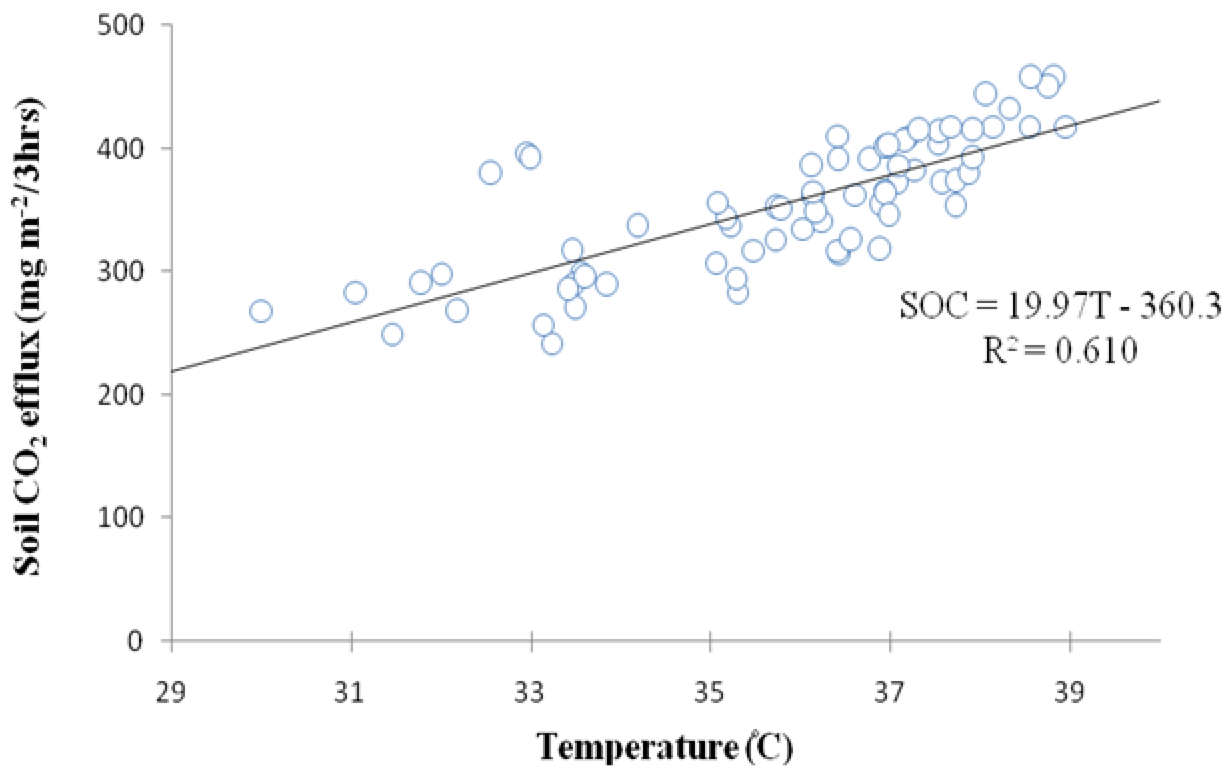

Fig. 5. 1 Relationship between soil temperature and soil CO_2 efflux (SCE: Soil CO_2 Efflux; T: temperature).

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5.4.2 Effect of water head

The effect of water head (flooding condition) on CO_2 evolution is not clear cut. The net effluxes of CO_2 from the paddy soil were lower when the soil was flooded than when it was kept under saturation condition (Table 4.1). A recent study by Liu *et al.,* (2013) reported that drainage during rice cultivation period significantly increase CO_2 emissions. Miyata *et al*, (2000) also found that water management on paddy soils had strong effects not only on CH_4 emissions but also on CO_2 emissions. Liu *et al.*, (2013) observed that during the submerged period of paddy rice cultivation, CO_2 production in the soil is severely restricted under flooding condition. This effect can be explained with two basic mechanisms (Maier *et al.*, 2011). First, flooding a field for subsequent rice cultivation cuts off the oxygen supply from the atmosphere and the microbial activities switch from aerobic (i.e. oxic condition) to facultative (i.e. anoxic condition) conditions (Kogel-Knabner *et al.*, 2010). As a result, biological activities reduce under anoxic condition, hence, inhibiting CO_2 production. Additionally, water replaces the gaseous phase in the soil pores. Since CO_2 diffusion rates in water are four orders of magnitude lower than those in air, part of the produced CO_2 is stored in the soil (Liu *et al.,* 2013). Hence, the soil CO_2 fluxes can be dramatically reduced by flooding during the paddy rice cultivation (Saito *et al.*, 2005; Miyata *et al.,* 2000; Campbell *et al.*, 2001 and Liu *et al.,* 2013).

Results from this present study provide indirect support for these conclusions, since the soil CO_2 efflux under flooding condition were lower than those observed under saturation condition. Even though keeping a head of water (flooding) reduces CO_2 evolution under paddy rice cultivation, research finding has revealed that CH_4 production increases (Miyata *et al.*, 2000).

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However, Cai *et al.,* 2001; Minamikawa and Sakai 2006; Liu *et al*., 2013 also reported that CH_4 emissions were higher under continuous flooding than intermittent draining practices.

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CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

Measurement of CO_2 efflux from the soil surface allows the understanding and accurate evaluation of soil management practices to reduce CO_2 emission from crop fields. Carbon dioxide effluxes were significantly influenced by soil amendments. The use of bio-char significantly reduced CO_2 emissions from irrigated fields. Bio-char therefore has the potential to reduce CO_2 efflux in irrigated rice cropping system. Application of N fertilizer resulted in increased CO_2 efflux. Emissions were also influenced by irrigation regime. The trend in data shows that higher emissions were observed few weeks after fertilizer application. High emissions were particularly observed from higher levels of application.

A 5 cm water head on the surface of the rice field yielded lower CO_2 efflux compared with keeping soil surface without a water head. We recommend for now, that, a head of water (0.05 m) should be kept on paddy soils to reduce CO_2 evolution. Temperature also correlated significantly with CO_2 efflux in this study and explained 61% of efflux with rate of efflux almost doubling (1.91) with 10^oC rise in temperature. This indicates that temperature is one of the factors that drive the processes of decomposition of organic matter.

Some suggestions to improve studies in this area include:

1. More improved methods of CO_2 measurement such as using gas chromatography have to be used where possible, to ensure more accurate measurement of CO_2 efflux as well as measuring other GHGs such as CH_4 and N_2O. This will be more resourceful in taking the

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decision on whether indeed keeping a 0.05 m head of water on paddy soils will enhance CH_4 production.

2. Measurement of CO_2 emission must be carried out on paddy fields especially with higher and varying application rates of bio-char for extended period of time.

3. Measurement of CO_2 emission from bio-char produced from other readily available sources of feedstock such as rice straw must be investigated.

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APPENDICES

Appendix A: Season one (I)

Appendix 1: A(a). Analysis of variance for soil CO_2 efflux, No amendment (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	26228.	13114.	1.85	
Treatment	2	6361.	3181.	0.45	0.668
Residual	4	28407.	7102.		
Total	8	60996.			

NS (Not significant)

Appendix 1: A(b). Analysis of variance for soil CO_2 efflux, No amendment (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	75570.	37785.	0.48	
Treatment	2	62499.	31250.	0.40	0.695
Residual	4	313870.	78467.		
Total	8	451939.			

NS

Appendix 1: A(c). Analysis of variance for soil CO_2 efflux, No amendment (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	9498.	4749.	0.25	
Treatment	2	11066.	5533.	0.29	0.761
Residual	4	75723.	18931.		
Total	8	96287.			

NS

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Appendix 2: A(a). Analysis of variance for soil CO_2 efflux, soil amendment (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	126088.	63044.	2.72	
Treatment	2	39822.	19911.	0.86	0.489
Residual	4	92714.	23179.		
Total	8	258624.			

NS

Appendix 2: A(b). Analysis of variance for soil CO_2 efflux, soil amendment (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	167653.	83826.	0.33	
Treatment	2	366065.	183033.	0.73	0.536
Residual	4	1001908.	250477.		
Total	8	1535626.			

NS

Appendix 2: A(c). Analysis of variance for soil CO_2 efflux, soil amendment (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	65876.	32938.	28.00	
Treatment	2	784.	392.	0.33	0.735
Residual	4	4705.	1176.		
Total	8	71366.			

NS

Appendix 3: A (a): Analysis of variance for soil CO_2 efflux, week 2 (6-10 am)

Source of variation	d.f .	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	647.3	323.7	0.45	
Main treatment	2	30436.4	15218.2	21.26	0.007
Residual	4	2863.1	715.8	1.06	
Fert level	3	29203.5	9734.5	14.42	< 001
Main treatment * Fert level	6	64607.6	10767.9	15.95	< 001
Residual	18	12153.0	675.2		
Total	35	139911.0			

l.s.d (5 %) = 44.44 (Main treatment * Fert level)

Appendix 3: A (b). Analysis of variance for soil CO_2 efflux, week 2 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	305.4	152.7	0.45	
Main treatment	2	10344.5	5172.2	15.09	0.014
Residual	4	1370.8	342.7	1.03	
Fert level	3	5544.5	1848.2	5.54	0.007
Main treatment * Fert level	6	11875.6	1979.3	5.93	0.001
Residual	18	6007.5	333.7		
Total	35	35448.3			

l.s.d (5 %) = 31.10 (Main treatment * Fert level)

Appendix 3: A (c). Analysis of variance for soil CO_2 efflux, week 3 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	4769.6	2384.8	2.34	
Main treatment	2	42920.5	21460.2	21.04	0.008
Residual	4	4079.5	1019.9	1.37	
Fert level	3	9072.3	3024.1	4.05	0.023
Main treatment * Fert level	6	33858.0	5643.0	7.56	< 001
Residual	18	13436.4	746.5		
Total	35	108136.2			

l.s.d (5 %) = 48.66 (Main treatment * Fert level)

Appendix 4: A (a). Analysis of variance for soil CO_2 efflux, week 3 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1242.	621.	0.08	
Main treatment	2	15750.	7875.	1.04	0.433
Residual	4	30259.	7565.	1.93	
Fert level	3	32274.	10758.	2.75	0.073
Main treatment * Fert level	6	30694.	5116.	1.31	0.304
Residual	18	70451.	3914.		
Total	35	180669.			

NS (Main treatment * Fert level)

Appendix 4: A (b). Analysis of variance for soil CO_2 efflux, week 3 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	10217.	5108.	2.59	
Main treatment	2	6361.	3181.	1.61	0.306
Residual	4	7886.	1971.	1.20	
Fert level	3	74285.	24762.	15.12	< 001
Main treatment * Fert level	6	150922.	25154.	15.36	< 001
Residual	18	29474.	1637.		
Total	35	279145.			

l.s.d (5 %) = 70.55 (Main treatment * Fert level)

Appendix 4: A (c). Analysis of variance for soil CO_2 efflux, week 3 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	27535.	13768.	0.30	
Main treatment	2	29823.	14911.	0.33	0.739
Residual	4	182510.	45627.	2.83	
Fert level	3	20695.	6898.	0.43	0.736
Main treatment * Fert level	6	239868.	39978.	2.48	0.063
Residual	18	290560.	16142.		
Total	35	790992.			

NS (Main treatment * Fert level)

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Appendix 5: A (a). Analysis of variance for soil CO_2 efflux, week 5 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	14726.	7363.	1.23	
Main treatment	2	62369.	31184.	5.22	0.077
Residual	4	23897.	5974.	2.04	
Fert level	3	156358.	52119.	17.83	< 001
Main treatment * Fert level	6	228932.	38155.	13.05	< 001
Residual	18	52609.	2923.		
Total	35	538892.			

l.s.d (5 %) = 104.84 (Main treatment * Fert level)

Appendix 5: A (b). Analysis of variance for soil CO_2 efflux, week 5 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	173600.	86800.	3.78	
Main treatment	2	83739.	41870.	1.82	0.274
Residual	4	91930.	22983.	0.72	
Fert level	3	41074.	13691.	0.43	0.734
Main treatment * Fert level	6	502958.	83826.	2.63	0.052
Residual	18	572821.	31823.		
Total	35	1466123.			

NS (Main treatment * Fert level)

Appendix 5: A (c). Analysis of variance for soil CO_2 efflux, week 5 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	37208.	18604.	1.94	
Main treatment	2	64983.	32491.	3.39	0.138
Residual	4	38341.	9585.	0.80	
Fert level	3	41684.	13895.	1.15	0.355
Main treatment * Fert level	6	250804.	41801.	3.47	0.019
Residual	18	216973.	12054.		
Total	35	649992.			

NS

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Appendix 6: A (a). Analysis of variance for soil CO_2 efflux, week 6 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1084.3	542.1	0.31	
Main treatment	2	25204.0	12602.0	7.18	0.047
Residual	4	7021.4	1755.4	4.55	
Fert level	3	38294.6	12764.9	33.12	< 001
Main treatment * Fert level	6	38331.1	6388.5	16.58	< 001
Residual	18	6937.5	385.4		
Total	35	116872.8			

l.s.d (5 %) = 49.06 (Main treatment * Fert level)

Appendix 6: A (b). Analysis of variance for soil CO_2 efflux, week 6 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	3253.7	1626.9	1.07	
Main treatment	2	1282.3	641.1	0.42	0.683
Residual	4	6093.6	1523.4	2.29	
Fert level	3	11325.5	3775.2	5.68	0.006
Main treatment * Fert level	6	29547.5	4924.6	7.41	<.001
Residual	18	11965.1	664.7		
Total	35	63467.6			

l.s.d (5 %) = 51.48 (Main treatment * Fert level) 51.48

Appendix 6: A (c). Analysis of variance for soil CO_2 efflux, week 6 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	27352.	13676.	7.06	
Main treatment	2	65355.	32678.	16.86	0.011
Residual	4	7752.	1938.	1.63	
Fert level	3	24845.	8282.	6.99	0.003
Main treatment * Fert level	6	101010.	16835.	14.20	<.001
Residual	18	21340.	1186.		
Total	35	247653.			

l.s.d (5 %) = 63.48 (Main treatment * Fert level)

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Appendix 7: A (a). Analysis of variance for soil CO_2 efflux, week 6 (6-10 am)

Source of variation	d.f.	s.s.	m.s	v.r.	F pr.
Rep stratum	2	17079.	8539.	4.99	
Main treatment	2	2246473.	1123236.	656.83	< .001
Residual	4	6840.	1710.	0.16	
Fert level	3	657540.	219180.	20.46	< .001
Main treatment * Fert level	6	1869995.	311666.	29.10	< .001
Residual	18	192792.	10711.		
Total	35	4990719.			

l.s.d (5 %) = 156.8 (Main treatment * Fert level)

Appendix 7: A (b). Analysis of variance for soil CO_2 efflux, week 6 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	675.	338.	0.11	
Main treatment	2	242090.	121045.	37.67	0.003
Residual	4	12853.	3213.	0.64	
Fert level	3	359040.	119680.	23.94	< .001
Main treatment * Fert level	6	1066020.	177670.	35.54	< .001
Residual	18	89991.	5000.		
Total	35	1770669.			

l.s.d (5 %) = 114.3 (Main treatment * Fert level)

Appendix 7: A (c). Analysis of variance for soil CO_2 efflux, week 7 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	5838.	2919.	0.35	
Main treatment	2	81321.	40661.	4.87	0.085
Residual	4	33374.	8343.	1.33	
Fert level	3	271303.	90434.	14.41	< .001
Main treatment * Fert level	6	241785.	40298.	6.42	< .001
Residual	18	112930.	6274.		
Total	35	746551.			

l.s.d (5 %) = 140.4 (Main treatment * Fert level)

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Appendix 8: A (a). Analysis of variance for soil CO_2 efflux, week 8 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	29278.	14639.	6.18	
Main treatment	2	3202.	1601.	0.68	0.559
Residual	4	9476.	2369.	0.76	
Fert level	3	26523.	8841.	2.83	0.067
Main treatment * Fert level	6	99184.	16531.	5.30	0.003
Residual	18	56138.	3119.		
Total	35	223802.			

l.s.d (5 %) = 91.69 (Main treatment * Fert level)

Appendix 8: A (b). Analysis of variance for soil CO_2 efflux, week 8 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	6339.	3170.	14.92	
Main treatment	2	679216.	339608.	1598.92	<.001
Residual	4	850.	212.	0.08	
Fert level	3	689553.	229851.	91.22	<.001
Main treatment * Fert level	6	302738.	50456.	20.02	<.001
Residual	18	45355.	2520.		
Total	35	1724050.			

l.s.d (5 %) = 75.34 (Main treatment * Fert level)

Appendix 8: A (a). Analysis of variance for soil CO_2 efflux, week 8 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	11524.	5762.	2.02	
Main treatment	2	576045.	288022.	100.93	<.001
Residual	4	11415.	2854.	0.70	
Fert level	3	99457.	33152.	8.15	0.001
Main treatment * Fert level	6	181268.	30211.	7.42	<.001
Residual	18	73261.	4070.		
Total	35	952969.			

l.s.d (5 %) = 103.9 (Main treatment * Fert level)

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Appendix 10: A (a). Analysis of variance for soil CO_2 efflux, week 10 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	29278.	14639.	6.18	
Main treatment	2	3202.	1601.	0.68	0.559
Residual	4	9476.	2369.	0.76	
Fert level	3	26523.	8841.	2.83	0.067
Main treatment * Fert level	6	99184.	16531.	5.30	0.003
Residual	18	56138.	3119.		
Total	35	223802.			

L.s.d (5 %) = 91.69 (Main treatment * Fert level)

Appendix 10: A (b). Analysis of variance for soil CO_2 efflux, week 10 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	6339.	3170.	14.92	
Main treatment	2	679216.	339608.	1598.92	<.001
Residual	4	850.	212.	0.08	
Fert level	3	689553.	229851.	91.22	<.001
Main treatment * Fert level	6	302738.	50456.	20.02	<.001
Residual	18	45355.	2520.		
Total	35	1724050.			

L.s.d (5 %) = 75.34 (Main treatment * Fert level)

Appendix 10: A (c). Analysis of variance for soil CO_2 efflux, week 10 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	11524.	5762.	2.02	
Main treatment	2	576045.	288022.	100.93	<.001
Residual	4	11415.	2854.	0.70	
Fert level	3	99457.	33152.	8.15	0.001
Main treatment * Fert level	6	181268.	30211.	7.42	<.001
Residual	18	73261.	4070.		
Total	35	952969.			

L.s.d (5 %) = 103.9 (Main treatment * Fert level)

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Appendix 12: A (a). Analysis of variance for soil CO_2 efflux, week 12 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	628.	314.	0.08	
Main treatment	2	166027.	83014.	21.41	0.007
Residual	4	15511.	3878.	2.72	
Fert level	3	703512.	234504.	164.47	<.001
Main treatment * Fert level	6	71813.	11969.	8.39	<.001
Residual	18	25664.	1426.		
Total	35	983156.			

L.s.d (5 %) = 79.11 (Main treatment * Fert level)

Appendix 12: A (b). Analysis of variance for soil CO_2 efflux, week 12 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1477.	739.	0.19	
Main treatment	2	439132.	219566.	55.22	0.001
Residual	4	15905.	3976.	2.28	
Fert level	3	557571.	185857.	106.54	<.001
Main treatment * Fert level	6	122854.	20476.	11.74	<.001
Residual	18	31402.	1745.		
Total	35	1168342.			

L.s.d (5 %) = 83.28 (Main treatment * Fert level)

Appendix 12: A (c). Analysis of variance for soil CO_2 efflux, week 12 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1298.	649.	0.62	
Main treatment	2	315242.	157621.	150.06	<.001
Residual	4	4201.	1050.	0.40	
Fert level	3	617388.	205796.	77.54	<.001
Main treatment * Fert level	6	74840.	12473.	4.70	0.005
Residual	18	47771.	2654.		
Total	35	1060741.			

L.s.d (5 %) = 80.51 (Main treatment * Fert level)

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Appendix 13: A (a): Analysis of variance for average weekly soil CO_2 efflux, Season I (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1024.8	512.4	0.71	
Main treatment	2	66673.5	33336.7	45.88	0.002
Residual	4	2906.7	726.7	2.27	
Fert level	3	65011.6	21670.5	67.80	<.001
Main treatment * Fert level	6	16640.1	2773.4	8.68	<.001
Residual	18	5753.3	319.6		
Total	35	158010.0			

L.s.d (5 %) = 30.6, 17.7, 35.6 for main, fertilizer and (Main treatment * Fert level) respectively.

Appendix 13: A (b). Analysis of variance for average weekly soil CO_2 efflux, Season I (10-2 pm)

Source of variationn	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	5810.9	2905.5	5.56	
Main treatment	2	95633.0	47816.5	91.45	<.001
Residual	4	2091.6	522.9	0.69	
Fert level	3	75629.7	25209.9	33.26	<.001
Main treatment * Fert level	6	23193.9	3865.7	5.10	0.003
Residual	18	13643.7	758.0		
Total	35	216002.7			

L.s.d (5 %) = 25.9, 27.3, 44.8 for main, fertilizer and (Main treatment * Fert level) respectively.

Appendix 13: A (c): Analysis of variance for average weekly soil CO_2 efflux, Season I (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	2297.	1148.	2.34	
Main treatment	2	36291.	18146.	37.04	0.003
Residual	4	1960.	490.	0.28	
Fert level	3	22440.	7480.	4.20	0.020
Main treatment * Fert level	6	27748.	4625.	2.60	0.054
Residual	18	32055.	1781.		
Total	35	122790			

L.s.d (5 %) = 25.09 and 41.79 for main and fertilizer treatment respectively.

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APPENDIX B: Season two (II)

Appendix 1: B (a). Analysis of variance for soil CO_2 efflux, week 2 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	2226.0	1113.0	0.35	
Main treatment	2	18110.8	9055.4	2.82	0.172
Residual	4	12853.2	3213.3	5.44	
Fert level	3	40944.7	13648.2	23.10	<.001
Main treatment * Fert level	6	64481.4	10746.9	18.19	<.001
Residual	18	10635.2	590.8		
Total	35	149251.4			

L.s.d (5 %) = 65.19 (Main treatment * Fert level)

Appendix 1: B (b). Analysis of variance for soil CO_2 efflux, week 2 10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1516.5	758.2	1.00	
Main treatment	2	37647.4	18823.7	24.73	0.006
Residual	4	3044.9	761.2	2.08	
Fert level	3	46104.7	15368.2	42.05	<.001
Main treatment * Fert level	6	55106.1	9184.3	25.13	<.001
Residual	18	6578.0	365.4		
Total	35	149997.6			

L.s.d (5 %) = 37.24 (Main treatment * Fert level)

Appendix 1: B (c). Analysis of variance for soil CO_2 efflux, week 2 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	22.	11.	0.01	
Main treatment	2	53379.	26690.	16.22	0.012
Residual	4	6580.	1645.	0.83	
Fert level	3	82395.	27465.	13.79	<.001
Main treatment Fert * level	6	93913.	15652.	7.86	<.001
Residual	18	35848.	1992.		
Total	35	272138.			

L.s.d (5 %) = 73.94 (Main treatment * Fert level)

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Appendix 2: B (a). Analysis of variance for soil CO_2 efflux, week 3 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1634.	817.	0.55	
Main treatment	2	40062.	20031.	13.47	0.017
Residual	4	5947.	1487.	0.71	
Fert level	3	39419.	13140.	6.24	0.004
Main treatment * Fert level	6	228235.	38039.	18.06	<.001
Residual	18	37905.	2106.		
Total	35	353201.			

L.s.d (5 %) = 74.80 (Main treatment * Fert level)

Appendix 2: B (b). Analysis of variance for soil CO_2 efflux, week 3 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	17275.	8638.	9.17	
Main treatment	2	13354.	6677.	7.09	0.048
Residual	4	3769.	942.	0.19	
Fert level	3	125173.	41724.	8.44	0.001
Main treatment * Fert level	6	440459.	73410.	14.85	<.001
Residual	18	89011.	4945.		
Total	35	689041.			

L.s.d (5 %) = 107.0 (Main treatment * Fert level)

Appendix 2: B (c). Analysis of variance for soil CO_2 efflux, week 3 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	11045.	5522.	1.37	
Main treatment	2	96527.	48263.	11.96	0.021
Residual	4	16142.	4036.	1.50	
Fert level	3	119324.	39775.	14.80	<.001
Main treatment * Fert level	6	612338.	102056.	37.99	<.001
Residual	18	48361.	2687.		
Total	35	903737.			

L.s.d (5 %) = 93.96 (Main treatment * Fert level)

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Appendix 3: B (a). Analysis of variance for soil CO_2 efflux, week 4 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	6078.	3039.	0.46	
Main treatment	2	87508.	43754.	6.63	0.054
Residual	4	26403.	6601.	2.73	
Fert level	3	29191.	9730.	4.02	0.024
Main treatment * Fert level	6	244051.	40675.	16.80	<.001
Residual	18	43591.	2422.		
Total	35	436821.			

L.s.d (5 %) = 103.17 (Main treatment * Fert level)

Appendix 3: B (b). Analysis of variance for soil CO_2 efflux, week 4 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1372.	686.	0.28	
Main treatment	2	59014.	29507.	12.04	0.020
Residual	4	9803.	2451.	0.72	
Fert level	3	239748.	79916.	23.49	<.001
Main treatment * Fert level	6	56531.	9422.	2.77	0.044
Residual	18	61236.	3402.		
Total	35	427704.			

L.s.d (5 %) = 95.26 (Main treatment * Fert level)

Appendix 3: B (c). Analysis of variance for soil CO_2 efflux, week 4 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	38428.	19214.	8.91	
Main treatment	2	28821.	14410.	6.68	0.053
Residual	4	8627.	2157.	0.77	
Fert level	3	29322.	9774.	3.49	0.037
Main treatment * Fert level	6	112952.	18825.	6.72	<.001
Residual	18	50453.	2803.		
Total	35	268602.			

L.s.d (5 %) = 87.04 (Main treatment * Fert level)

Appendix 4: B (a). Analysis of variance for soil CO_2 efflux, week 5 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	85242.	42621.	4.20	
Main treatment	2	2701.	1351.	0.13	0.879
Residual	4	40628.	10157.	0.78	
Fert level	3	33058.	11019.	0.84	0.488
Main treatment * Fert level	6	164603.	27434.	2.10	0.104
Residual	18	235141.	13063.		
Total	35	561373.			

NS (Main treatment * Fert level)

Appendix 4: B (b). Analysis of variance for soil CO_2 efflux, week 5 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	5947.	2974.	5.87	
Main treatment	2	36140.	18070.	35.68	0.003
Residual	4	2026.	506.	0.13	
Fert level	3	27220.	9073.	2.27	0.115
Main treatment *Fert level	6	92998.	15500.	3.87	0.012
Residual	18	72019.	4001.		
Total	35	236350.			

L.s.d (5 %) = 95.4 (Main treatment * Fert level)

Appendix 4: B (c). Analysis of variance for soil CO_2 efflux, week 5 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	3682.	1841.	0.50	
Main treatment	2	16491.	8245.	2.23	0.224
Residual	4	14813.	3703.	1.22	
Fert level	3	91266.	30422.	10.06	<.001
Main treatment * Fert level	6	43198.	7200.	2.38	0.072
Residual	18	54439.	3024.		
Total	35	223889.			

NS (Main treatment * Fert level)

Appendix 5: B (a). Analysis of variance for soil CO_2 efflux, week 6 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	646.0	323.0	0.67	
Main treatment	2	14399.5	7199.7	15.03	0.014
Residual	4	1916.1	479.0	0.66	
Fert level	3	52035.4	17345.1	23.94	<.001
Main treatment * Fert level	6	31575.4	5262.6	7.26	<.001
Residual	18	13042.6	724.6		
Total	35	113615.0			

L.s.d (5 %) = 43.61 (Main treatment * Fert level)

Appendix 5: B (b). Analysis of variance for soil CO_2 efflux, week 6 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1336.9	668.4	0.34	
Main treatment	2	8571.1	4285.5	2.21	0.226
Residual	4	7768.6	1942.1	2.74	
Fert level	3	16811.1	5603.7	7.90	0.001
Main treatment * Fert level	6	76683.3	12780.6	18.02	<.001
Residual	18	12767.9	709.3		
Total	35	123938.8			

L.s.d (5 %) = 55.91 (Main treatment * Fert level)

Appendix 5: B (c). Analysis of variance for soil CO_2 efflux, week 6 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	5549.7	2774.8	1.23	
Main treatment	2	97559.0	48779.5	21.61	0.007
Residual	4	9029.4	2257.4	3.00	
Fert level	3	36581.8	12193.9	16.20	<.001
Main treatment *F ert level	6	24840.9	4140.1	5.50	0.002
Residual	18	13552.1	752.9		
Total	35	187112.9			

L.s.d (5 %) = 59.23 (Main treatment * Fert level)

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Appendix 6: B (a). Analysis of variance for soil CO_2 efflux, week 7 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	13593.	6797.	4.84	
Main treatment	2	4117.	2059.	1.47	0.333
Residual	4	5620.	1405.	1.01	
Fert level	3	25913.	8638.	6.23	0.004
Main treatment * Fert level	6	75091.	12515.	9.02	<.001
Residual	18	24965.	1387.		
Total	35	149299.			

L.s.d (5 %) = 63.29 (Main treatment * Fert level)

Appendix 6: B (b). Analysis of variance for soil CO_2 efflux, week 7 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	5642.	2821.	0.49	
Main treatment	2	64852.	32426.	5.63	0.069
Residual	4	23048.	5762.	1.92	
Fert level	3	97017.	32339.	10.80	<.001
Main treatment * Fert level	6	177565.	29594.	9.88	<.001
Residual	18	53916.	2995.		
Total	35	422040.			

L.s.d (5 %) = 104.60 (Main treatment * Fert level)

Appendix 6: B (c). Analysis of variance for soil CO_2 efflux, week 7 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	9302.	4651.	0.73	
Main treatment	2	262023.	131011.	20.63	0.008
Residual	4	25401.	6350.	1.39	
Fert level	3	118453.	39484.	8.62	<.001
Main treatment * Fert level	6	483875.	80646.	17.61	<.001
Residual	18	82410.	4578.		
Total	35	981463.			

L.s.d (5 %) = 120.8 (Main treatment * Fert level) 120.8

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Appendix 7: B (a). Analysis of variance for soil CO_2 efflux, week 8 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	610.	305.	0.20	
Main treatment	2	13354.	6677.	4.27	0.102
Residual	4	6252.	1563.	0.97	
Fert level	3	32502.	10834.	6.74	0.003
Main treatment * Fert level	6	65593.	10932.	6.80	<.001
Residual	18	28951.	1608.		
Total	35	147263.			

L.s.d (5 %) = 67.77 (Main treatment * Fert level)

Appendix 7: B (b). Analysis of variance for soil CO_2 efflux, week 8 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	3202.	1601.	3.92	
Main treatment	2	12809.	6405.	15.68	0.013
Residual	4	1634.	408.	0.12	
Fert level	3	101548.	33849.	9.98	<.001
Main treatment * Fert level	6	306289.	51048.	15.05	<.001
Residual	18	61040.	3391.		
Total	35	486522.			

L.s.d (5 %) = 87.80 (Main treatment * Fert level)

Appendix 7: B (c). Analysis of variance for soil CO_2 efflux, week 8 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	65.	33.	0.04	
Main treatment	2	285920.	142960.	159.09	<.001
Residual	4	3594.	899.	0.27	
Fert level	3	222506.	74169.	22.01	<.001
Main treatment * Fert level	6	836151.	139358.	41.36	<.001
Residual	18	60648.	3369.		
Total	35	1408884.			

L.s.d (5 %) = 89.17 (Main treatment * Fert level)

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Appendix 8: B (a). Analysis of variance for soil CO_2 efflux, week 10 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	35639.	17820.	0.43	
Main treatment	2	69950.	34975.	0.83	0.498
Residual	4	167609.	41902.	1.47	
Fert level	3	121677.	40559.	1.42	0.269
Main treatment * Fert level	6	370269.	61712.	2.17	0.095
Residual	18	512761.	28487.		
Total	35	1277906.			

L.s.d (5 %) = 304.7 (Main treatment * Fert level)

Appendix 8: B (b). Analysis of variance for soil CO_2 efflux, week 10 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	3202.	1601.	0.61	
Main treatment	2	388655.	194328.	74.34	<.001
Residual	4	10457.	2614.	1.39	
Fert level	3	7352.	2451.	1.30	0.305
Main treatment * Fert level	6	49211.	8202.	4.35	0.007
Residual	18	33918.	1884.		
Total	35	492796.			

L.s.d (5 %) = 77.53 (Main treatment * Fert level)

Appendix 8: B (c). Analysis of variance for soil CO_2 efflux, week 10 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	457.	229.	0.15	
Main treatment	2	307095.	153547.	99.98	<.001
Residual	4	6143.	1536.	0.42	
Fert level	3	247155.	82385.	22.53	<.001
Main treatment * Fert level	6	641747.	106958.	29.25	<.001
Residual	18	65811.	3656.		
Total	35	1268408.			

L.s.d (5 %) = 94.8 (Main treatment * Fert level)

Appendix 9: B (a). Analysis of variance for soil CO_2 efflux, week 12 (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	17798.	8899.	2.03	
Main treatment	2	36685.	18342.	4.19	0.104
Residual	4	17493.	4373.	0.58	
Fert level	3	155366.	51789.	6.82	0.003
Main treatment * Fert level	6	42785.	7131.	0.94	0.492
Residual	18	136719.	7595.		
Total	35	406845.			

NS (Main treatment * Fert level)

Appendix 9: B (b). Analysis of variance for soil CO_2 efflux, week 12 (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1562551.	781276.	1.12	
Main treatment	2	6043363.	3021681.	4.34	0.099
Residual	4	2783959.	695990.	0.99	
Fert level	3	7519572.	2506524.	3.58	0.035
Main treatment * Fert level	6	18536586.	3089431.	4.41	0.007
Residual	18	12617089.	700949.		
Total	35	49063120.			

L.s.d (5 %) = 1418.9 (Main treatment * Fert level)

Appendix 9: B (c). Analysis of variance for soil CO_2 efflux, week 12 (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1067.	534.	0.06	
Main treatment	2	176802.	88401.	9.70	0.029
Residual	4	36445.	9111.	1.59	
Fert level	3	395648.	131883.	22.96	<.001
Main treatment * Fert level	6	267164.	44527.	7.75	<.001
Residual	18	103389.	5744.		
Total	35	980516.			

L.s.d (5 %) = 138.9 (Main treatment * Fert level)

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Appendix 10: A (a). Analysis of variance for soil CO_2 efflux, average Season II (6-10 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1005.	503.	1.30	
Main treatment	2	8834.	4417.	11.43	0.022
Residual	4	1546.	386.	0.37	
Fert level	3	9327.	3109.	2.99	0.059
Main treatment * Fert level	6	7924.	1321.	1.27	0.320
Residual	18	18741.	1041.		
Total	35	47377.			

L.s.d (5 %) = 22.3 (Main treatment)

Appendix 10: A (b). Analysis of variance for soil CO_2 efflux, average Season II (10-2 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	441.1	220.5	8.93	
Main treatment	2	21552.5	10776.2	436.34	<.001
Residual	4	98.8	24.7	0.09	
Fert level	3	17680.7	5893.6	21.50	<.001
Main treatment * Fert level	6	29124.3	4854.1	17.71	<.001
Residual	18	4933.8	274.1		
Total	35	73831.1			

L.s.d (5 %) = 5.6, 16.4, 24.9 for main, fertilizer and (Main treatment * Fert level) respectively.

Appendix 10: A (c). Analysis of variance for soil CO_2 efflux, average Season II (2-6 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	1137.7	568.9	0.80	
Main treatment	2	38603.8	19301.9	27.09	0.005
Residual	4	2850.4	712.6	2.67	
Fert level	3	44300.4	14766.8	55.40	<.001
Main treatment * Fert level	6	36855.6	6142.6	23.04	<.001
Residual	18	4798.2	266.6		
Total	35	128546.1			

L.s.d (5 %) = 30.3, 16.2, 34.0 for main, fertilizer and (Main treatment * Fert level) respectively

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Appendix C: Pot experiment

Appendix C: (a). Analysis of variance for average weekly soil CO_2 efflux (6-9 am)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Main treatment	11	19763.2	1796.7	15.92	<.001
Residual	24	2708.4	112.9		
Total	35	22471.7			

L.s.d (5 %) = 17.90

Appendix C: (b). Analysis of variance for average weekly soil CO_2 efflux (10-1 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Main treatment	11	31242.7	2840.2	4.79	<.001
Residual	24	14223.0	592.6		
Total	35	45465.7			

L.s.d (5 %) = 41.02

Appendix C: (c). Analysis of variance for average weekly soil CO_2 efflux (2-5 pm)

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Main treatment	11	149653.7	13604.9	81.27	<.001
Residual	24	4017.6	167.4		
Total	35	153671.3			

L.s.d (5 %) = 21.80