

# **Evaluation of soil chemical and physical characteristics in a complex agroecosystem in the Argentine Pampa.**

by

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## **Author's Declaration**

I hereby declare that I am the sole author of this thesis. This is a true copy of the thesis, including any required final revisions, as accepted by my examiners.

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## Abstract

The Argentine Pampa is a global producer of maize (*Zea mays* L.) and soybean [*Glycine max* L. (Merr.)], however agricultural practices have caused severe soil degradation and amplified greenhouse gas (GHG) production rates. This study presents the effects of maize-legume intercrops compared with maize and soybean sole crops on GHG production rates and soil physical properties over two field seasons. It also presents the results from a laboratory study in which GHGs were quantified from soils amended with maize and soybean crop residues. In the field study, soil organic carbon (SOC) concentrations were significantly greater ( $p < 0.05$ ) in the maize sole crop and intercrops, whereas soil bulk density was significantly lower in the intercrops and as a consequence soil infiltration was higher. Soil total nitrogen (TN) concentrations were not significantly different between treatments. Soil CO<sub>2</sub> production rates were significantly greater in the maize sole crop but did not differ significantly for N<sub>2</sub>O. However, over the two field seasons both trace gases showed a general trend of greater production rates in the maize sole crop followed by the soybean sole crop. Linear regression between soil GHGs and soil temperature or volumetric soil moisture accounted for up to 51% of the variability in soil CO<sub>2</sub> production rates and 60% of the soil N<sub>2</sub>O production rates. In the laboratory study, soil GHG production rates varied between treatments and between residue addition for both CO<sub>2</sub> and N<sub>2</sub>O but varied only narrowly between treatments and experiments for CH<sub>4</sub>. Results from this study provided further insight into the effect of agroecosystem management practices on GHG production rates and soil physical and chemical characteristics, and contributed to improving our understanding of optimal agroecosystem design.

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## Introduction

Soil, the foundation for most terrestrial life, is highly complex. It is a layer of minerals altered physically and chemically from the bedrock by geological weathering, nutrient cycling and biomass growth and decay (Uphoff, 2006). One gram of soil can contain billions of fungi and bacteria and thousands of plant and animal species (Uphoff, 2006). Soil is the principle medium for plant growth and thus the primary environmental resource that supports agriculture (Wright and Hons, 2004; Desjardins et al., 2005). According to Lal et al. 2006, improving soil quality will increase the soil organic carbon (SOC) stock while increasing the production of food grains by more than 30-40Tg/yr. This is especially important as several crop-simulation models and future climate scenarios have predicted that grain yield could be reduced by 30% by 2080 under warmer climate scenarios (IPCC, 2007b). According to the IPCC, this could place an additional 5, 26 and 85 million people at risk of hunger globally, by the years 2020, 2050 and 2080 respectively. Using a Global Climate Model (GCM), Magrin and Travasso (2002) predicted that a 3°C increase in temperature in the Argentine Pampa would decrease wheat yields by 4% and maize yields by 9%.

There is an escalating concern that modern agricultural practices have been trading short-term increases in food production for long-term losses and degradation of environmental resources (Matson et al., 1997; Foley et al., 2005). With predictions of decreased yields, landowners need to intensify cropping systems to maintain crop yields, thus, increasing the amount of greenhouse gases (GHGs) emitted and further degrading the soil. However, sustainable agricultural land management practices have been proposed (Olesen and Bindi, 2002; Smith et al., 2007) to improve soil quality and prevent further soil degradation (Niggli et al.,

2009), some of these include, sustainable soil and fertilizer management practices (Verchot et al., 2008) such as conservation tillage including no-till farming, agroforestry and intercropping.

Intercropping, where more than one crop is grown on the same land unit at the same time may be a more sustainable agricultural production system compared to conventional or sole crop systems. Yet, the majority of research on intercropping systems has focused on grain yield (Tsubo et al., 2003), resource use (Willey, 1990), tillage (Hernanz et al., 2009), soil quality (Drinkwater et al., 1998), N fixation (Ofori and Stern, 1987; Stern, 1993) and fertilizer requirement (Studdert and Echeverria, 2000). However, field and laboratory experiments to quantify GHG dynamics and soil physical and chemical characteristics in intercrops are lacking. Such information is crucial as it identifies other agroecosystem management practices effective in the mitigation of GHGs (Rochette et al., 1999) and further contributes to already existing global GHG databases (Smith et al., 2007; Verchot et al., 2008).

## **General Research Goals and Objectives**

The general objectives of this study were as follows:

- 1) To quantify GHG production rates from intercropping systems compared to sole crop systems.
- 2) To determine temporal changes in soil chemical and physical properties from an intercropping system compared to sole crop systems.
- 3) To determine GHG production rates from soils amended with soybean and maize crop residues.

# Chapter 1

## Literature Review

### 1.1 Land-use Change

Increasingly, land-use change is a force of global importance. Land-use changes such as deforestation for agricultural crop production or grazing land are motivated by an escalating need for food, fiber, water, and shelter for an exploding human population. Although land-use practices vary across the landscape, the outcome is typically the same: the extraction of natural resources for human needs to the detriment of our environment (Foley et al., 2005). Land-use change affects the exchange of greenhouse gases (GHGs) between the terrestrial ecosystem and the atmosphere (Watson et al., 2003); through changes in carbon (C) stocks (Guo and Gifford, 2002; Lal, 2004b). In the last four decades for instance, 500 million hectares (ha) of natural ecosystems have been converted to agricultural land (FAO, 2010). For instance in 2002; agricultural land accounted for 5020 Mha; 69% of which was under pasture while cropland accounted for 1% (FAO, 2003). Such land conversion has made agriculture one of the largest terrestrial systems, occupying approximately 40% of the total global area (Ramankutty and Foley, 1999; Asner et al., 2004).

This conversion has affected the amount of C and nitrogen (N) stored in vegetation biomass and soil and further affected soil aeration, water and temperature dynamics and soil aggregation (IPCC, 2007b). The result is that agriculture has exerted immense pressure on land

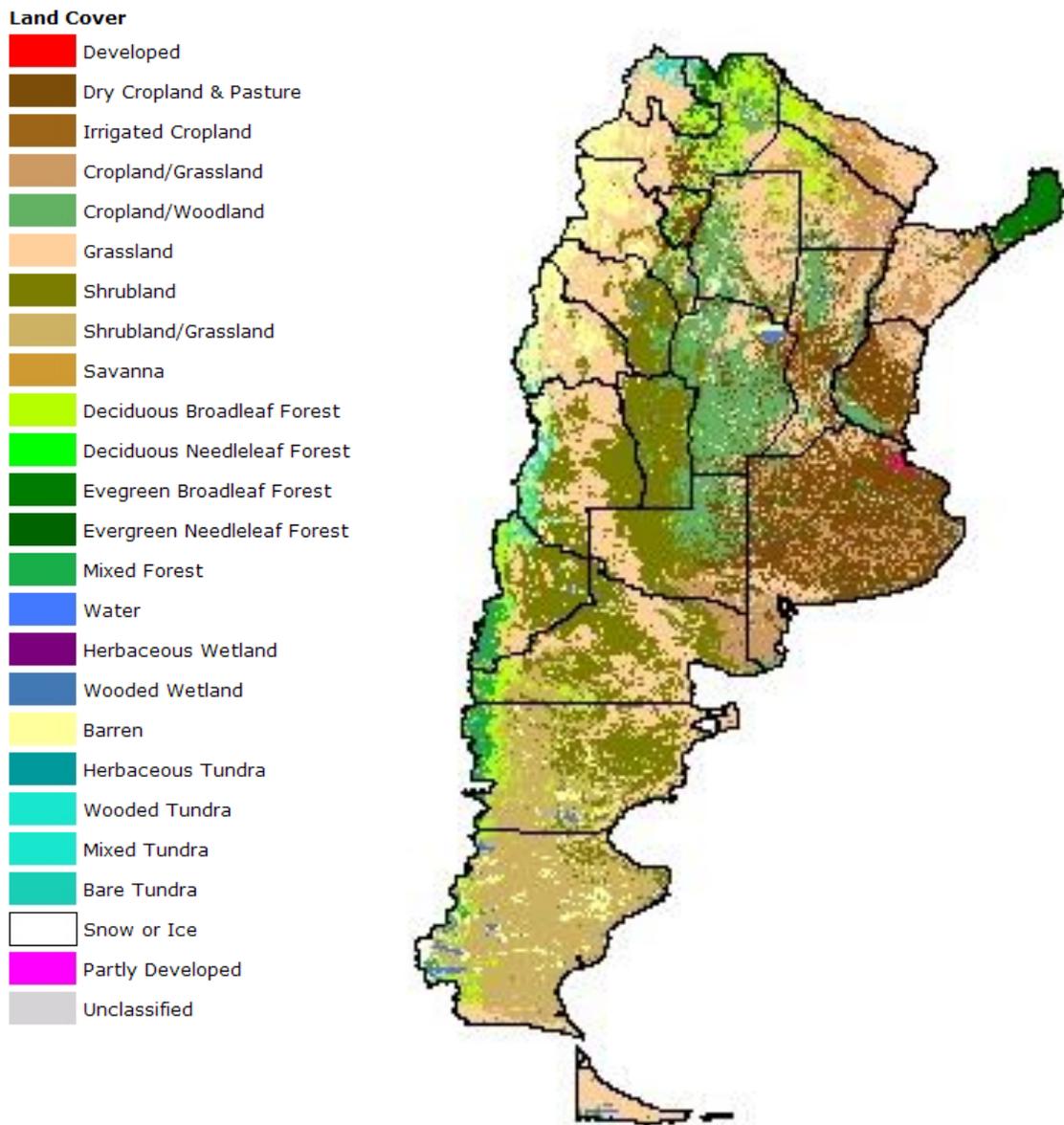
and water resources often leading to land degradation including soil erosion, salinisation and water pollution (Olesen and Bindi, 2002).

Several studies have reviewed land-use change and its effects on soil C including deforestation for pasture use (Neil and Davidson, 2000), crop cultivation (Mann, 1986; Davidson and Ackerman, 1993), biomass burning and removal of biomass (Fearnside, 2000), forest clearing (Allen, 1985) and tropical forest clearing (Detwiler, 1986). They have found that these land-use changes have dramatically depleted the storage of terrestrial C, emitting approximately  $136 \pm 55$  Pg (petagram) of carbon dioxide (CO<sub>2</sub>) from 1850 to 1998 (IPCC, 2007b). However, more recently, the Food and Agriculture Organization (FAO) predicted that by 2030 CO<sub>2</sub> production rates from land-use change could become stable or possibly decline due to the increased adoption of conservation practices such as agroforestry, intercropping and reduced tillage (FAO, 2003).

### **1.1.1 Land-use change in Latin America**

In the past, a lack of knowledge on the link between C sequestration and land management practices has led to conversion of C-rich forest and grassland to agriculture and pasture which store less C per unit area of land (Wood et al., 2000; Smith et al., 2007). This land-use change was observed on a global scale including Latin America. For example, during the 1930s, grassland in Latin America became rapidly converted to agriculture land until cattle production took over for most of the 1940s. However in the 1970s, a process known as '*agriculturización*' occurred where cereal production grew tremendously in contrast to previous

periods of agricultural stagnation and a predominance of cattle (Arroyo et al., 1985). This process was brought on by the burden of high debt, which triggered an increase in export of agricultural commodities, and a shift in policies intended at improving the trade balance (Smith et al., 2007). *Agriculturización* occurred predominantly in the Pampa region of Argentina (37°25'0''S, 67°0'0''W) and led to widespread land-use changes. For instance in 1974, cattle production covered 39,278 ha while in 1986 it fell by just under 8% to 36,196 ha. During the same period, area under wheat-soybean (*Glycine max* L. / *Triticum aestivum* L.) rotation increased from 165,000 ha to 990,000 ha (Arroyo et al., 1985). Presently, the majority of land in the Pampa is still centered in dry and irrigated croplands (Figure 1.1) and lands are most devoted to grain crops such as maize (*Zea mays* L.) and wheat that rotate with annual forage crops and perennial pastures (Diaz-Zorita et al., 2001). Land-use change in the region, brought on by *agriculturización* has greatly affected SOC stocks and other physical properties including soil fertility, structural porosity and infiltration rates (Noellemeyer et al., 2008).



**Figure 1.1 - Land-use systems in Argentina in 2009 (FAO, 2010)**

## **1.2 Soil and the Global Carbon and Nitrogen Cycle**

### **1.2.1 Soil Quality and Organic Matter**

Soil quality has been defined as the capacity of the soil to function within the boundary of an ecosystem while sustaining biological productivity, maintaining environmental quality, and promoting plant and animal health (Doran et al., 1994). The foundation of soil quality are the physical, chemical and biological properties of the soil, which vary as a function of the complex management practices applied, such as tillage, crop rotation and crop residues addition (Fuentes et al., 2009). However, enhancing soil quality can be a timely and challenging task for soil scientists; this involves increasing soil organic matter (SOM), which in turn improves aggregation, soil biodiversity and plant available water capacity. According to Lal et al. (2006), improving soil quality will increase the SOM stock while increasing the production of food grains by more than 30-40Tg/yr.

Soil organic matter plays an important role in maintaining soil physical properties and processes (Gregorich et al., 2001) such as soil structure, water-holding capacity cation-exchange capacity, nutrient supply to plants and the ability of a soil to recuperate after tillage and cropping (Matson et al., 1997; Studdert and Echeverria, 2000). A loss of SOM can lead to a decline in crop productivity (Lal and Bruce, 1999), a decrease in soil aggregate stability (Castro et al., 2002; Cosentino et al., 2006) and impede long-term sustainability of agriculture (Alvaro-Fuentes et al., 2008). However, SOM can be controlled by conservation management practices (Fuentes et al., 2009) including choice of crops and crop rotation (Larson et al., 1972), management of

crop residues, methods and intensity of tillage and green manure (Matson et al., 1997; Bhattacharyya et al., 2009). Several long-term studies have demonstrated the benefits of crop rotation on SOM and crop productivity (Ahmed and Rao, 1982; Li et al., 2001; Tsubo et al., 2003; Bhattacharyya et al., 2009). In a 10 year, no-till maize-wheat rotation study in Mexico resulted in stable yields, increased soil micro-flora and microbial biomass (Govaerts et al., 2008). In the Argentine Pampa, the incorporation of legumes on land where cattle graze has been shown to increase soil microbial activity, restore organic matter as well as increase soil fertility, and SOC and N contents (Diaz-Zorita et al., 2001).

### **1.2.2 Soil Organic Carbon**

Soil in its natural state contains a large soil organic carbon (SOC) stock that stratifies the surface horizons and decreases rapidly with depth (Fabrizzi et al., 2003). It is primarily composed of humus; a dark brown or black amorphous material comprised of plant and animal residue at various stages of decomposition and microbial by-products (Howarth, 2007). Compared with most plant residues, humus is extremely stable and is slowly broken down thus the organic C in humus can be over 1000 years old (Lal, 2004b). The SOC stock is comprised of two components: the inert or recalcitrant component, which is controlled by site parameters such as soil type, climate, and land-use history. The second component is the labile or active fraction, which is involved in the mineralization process (Lal, 2006).

Globally, losses of SOC from land-use change has been immense; several best estimates include 40 Pg by Houghton (2007), 55 Pg by the IPCC (2007a) and 60-100 Pg by Lal (2004b and

2006). Loss of SOC results in production rates of carbon dioxide (CO<sub>2</sub>) to the atmosphere however the SOC stock can be maintained by employing a variety of practices, including conservation tillage, residue management, use of agroforestry systems and incorporation of legumes in crop rotations (Post and Kwon, 2000; Studdert and Echeverria, 2000; Alvaro-Fuentes et al., 2009). Enhancement of SOC increases the availability of plant nutrients, improves the plant available water capacity (Lal, 2004b), decreases the risk of soil erosion and sedimentation and improves the supply of nutrients (Lal, 2006). Moreover, an increase in SOC can reduce the need for fertilizers while still maintaining crop yields (Studdert and Echeverria, 2000). For instance, in Latin America an increase in the SOC stock through improvement in soil quality could increase their crop yields substantially (Table 1.1). An increase in the SOC stock by 0.5 Mg ha<sup>-1</sup> yr<sup>-1</sup> could increase maize yields by between 100-150 kg ha<sup>-1</sup> while an increase of 1 Mg ha<sup>-1</sup> yr<sup>-1</sup> could increase yields be more than 200 kg ha<sup>-1</sup>.

**Table 1.1 - Potential for an increase in crop yield (kg ha<sup>-1</sup>) through improvement of soil quality while enhancing the soil organic carbon (SOC) stock in Latin America. Adapted from Lal, 2004.**

Crop	Area (ha)	Yield (kg ha <sup>-1</sup> )	Increase in SOC stock by 0.5 Mg ha <sup>-1</sup> yr <sup>-1</sup>		Increase in SOC stock by 1 Mg ha <sup>-1</sup> yr <sup>-1</sup>	
			Increase in Yield (kg ha <sup>-1</sup> )	Productivity increase (10 <sup>6</sup> Mg yr <sup>-1</sup> )	Increase in Yield (kg ha <sup>-1</sup> )	Productivity increase (10 <sup>6</sup> Mg yr <sup>-1</sup> )
Wheat	9.0	2515	25-35	0.225-0.315	50-70	0.45-0.63
Maize	22.6	3124	100-150	2.26-3.435	200-300	4.52-6.87
Rice	6.5	3585	15-25	0.10-0.615	30-50	0.20-0.33
Sorghum	4.1	3163	50-70	0.20-0.285	100-140	0.41-0.57
Soybean	24.0	2389	15-25	0.36-0.60	30-50	0.75-1.20

Management practices that can enhance the SOC stock may be vital in increasing crop productivity and sequestering C. Thus, the quality and rate of addition of SOC is dependent upon the plant species and/or the combination of several species (Rumpel and Chabbi, 2009). For instance, sustainable agricultural management practices, including complex systems such as intercropping and agroforestry, have been shown to reduce C and N losses (Mungai et al., 2006). Crops influence C cycling through the inputs of aboveground litter and rhizodeposits, however, the impact of this C is dependent upon the biochemical composition of inputs and their use of various components of the soil food web (Rumpel and Chabbi, 2009). Variation in plant species and their effects on the flow of C through the soil system can thus explain the potential for C accumulation in SOM and variation in GHG production rates (Rumpel and Chabbi, 2009). In a long-term study conducted by Studdert and Echeverría (2000) in Balcarce, Argentina, SOC concentrations ( $\text{g kg}^{-1}$ ) decreased dramatically between 4.1 to  $8.8 \text{ g kg}^{-1}$  after 11 years of a conventional wheat cropping system. However, in the same study, after 3 years with the inclusion of maize crops in the rotation, SOC increased by  $30.2 \pm 2.5 \text{ g kg}^{-1}$ . Several field experiments have also found a close linear relationship between rates of residue C return and the quantity of SOM levels found in temperate agriculture soils (Rasmussen et al., 1980; Domínguez et al., 2009; Hernanz et al., 2009). As such, these systems serve a dual purpose by maintaining crop productivity while simultaneously sequestering C and conserving biodiversity (Mungai et al., 2006).

### 1.2.3 Soil Nitrogen

Nitrogen (N) is crucial to plant growth, as large amounts are required to support photosynthesis and protein formation. The difficulty is it is essentially the only plant nutrient that is not released by the weathering of minerals in soils (Schulten and Schnitzer, 1998). The two main strategies for improving N availability in an agricultural system are to increase inputs or to reduce losses. The sole source of N in soils is synthetic fertilizer additions or dinitrogen ( $N_2$ ) through the symbiotic or non-symbiotic fixation of this nutrient by plants and bacteria (Uphoff, 2006). However, most organisms cannot use atmospheric  $N_2$  and therefore the N must originate from the highly competitive soil environment. Addition of soil N occurs through the fixation of  $N_2$  by a minority of microorganisms or a leguminous plant and from the return of ammonia ( $NH_4^+$ ) and nitrate ( $NO_3^-$ ) in rainwater (Uphoff, 2006). The challenge is that crops require large concentrations of N, but only trace amounts are accessible in mineral forms in the soil at any given time (Schulten and Schnitzer, 1998). Highly productive maize crops for instance, with a yield of 10 tons per ha of grain will need to extract  $260 \text{ kg N ha}^{-1}$  from the soil. This is equivalent to 5.2 tons of N over 20 years or 50% of the N stored in the native SOM (Uphoff, 2006).

In the temperate zone where agriculture is predominant, N is often the limiting nutrient and consequently restoring N loss is an imperative goal. Improving nutrient-use efficiency in agriculture thus requires organic and inorganic sources such as the use of synthetic or inorganic fertilizer. Field trials however, have shown that only one-third of fertilizer N applied to soils in the temperate region is immobilized and retained in organic forms after the first growing season

(Kelley and Stevenson, 1995). Of this, less than 15% will become available to plants during the following growing season.

The production of anthropogenic N was estimated at 15 Tg N yr<sup>-1</sup> in the 1860s but by 1990 it increased by approximately 10% to 140 Tg N yr<sup>-1</sup> (Martinelli et al., 2001; Galloway et al., 2004). The FAO reported that Latin America and the Caribbean consumed approximately five million tons of fertilizer N; this is equivalent to 6% of global consumption. Of this, Argentina consumed nearly 20% of the total N for the region (FAO, 2010). With the use of synthetic N fertilizer and increased fossil fuel production rates, the concentration of reactive nitrogen (Nr) and nitrous oxide (N<sub>2</sub>O) has dramatically increased in the past century (Snyder et al., 2009). This is because N applied to agricultural soils may be lost from the fields through surface erosion or leaching, the leached N then recycles through the soil and water systems and is eventually denitrified and converted to N<sub>2</sub> and N<sub>2</sub>O (Mosier, 1993). For example, in field (Sehy et al., 2003) and laboratory (Jarecki et al., 2008) studies, denitrification rates and/or N<sub>2</sub>O fluxes have increased following fertilizer application. Similarly, in a maize-wheat rotation in the North China Plain, Ding et al. (2007) found N<sub>2</sub>O production rates peaked following fertilizer application after which it remained consistently low throughout the growing season.

### 1.3 Anthropogenic Climate Change and Greenhouse Gases

One of the major, over-riding environmental concerns of our time is anthropogenic climate change. Since the Industrial Revolution, there has been a sharp increase in the concentration of GHGs in the atmosphere, increased surface temperatures, shifting weather patterns and unpredictable precipitation (IPCC, 2007a). The Intergovernmental Panel on Climate Change (IPCC) has concluded that with 95% certainty the main drivers of climate change have been anthropogenic increases in GHG production rates. They declared that, “warming of the climate system is unequivocal” (IPCC, 2007c). The three main naturally occurring radiatively active GHGs are CO<sub>2</sub>, methane (CH<sub>4</sub>), and nitrous oxide (N<sub>2</sub>O). These gases naturally trap the outgoing infrared radiation from the earth’s surface, which adds to the net energy input of the lower atmosphere and leads to regional and global changes in climatic parameters (Rastogi et al., 2002). However, the greenhouse effect has been heightened by anthropogenic activities, which have consequently increased the concentrations of naturally occurring GHGs (IPCC, 2007b; Barreto et al., 2009). The anthropogenic enrichment of GHGs in the atmosphere and their collective radiative forcing has led to a significant increase in global surface temperatures (Wang et al, 2010).

Globally, from 1970 to 2004, GHG production rates increased by 70% from 28.7 to 49 Gg CO<sub>2</sub>-eq. (IPCC, 2007c). This enrichment of GHGs in the atmosphere has initiated an increase in global surface temperatures from approximately 0.6 °C in the late 19<sup>th</sup> century to a current warming rate of 0.17 °C/decade (IPCC, 2007c). Since the 1990s surface temperatures have increased between 0.4 and 0.8 °C (0.6 ± 0.2 °C), while CO<sub>2</sub> has increased at a rate of 3.3 Pg C/yr

since 1999 (IPCC, 2007c). This observed rate of increase is beyond the threshold for which ecosystems can adjust (Lal, 2004b). The IPCC has reported that the majority of observed increases in mean global temperatures since the 20<sup>th</sup> century are likely due to the observed increase in anthropogenic GHG concentrations (IPCC, 2007a). What's more, these climatic changes have decreased the SOC stock and soil structural stability, increased the soils susceptibility to erosion and have disrupted biogeochemical cycles (Lal, 2004a).

### **1.3.1 Carbon Dioxide, Nitrous Oxide and Methane**

Carbon dioxide (CO<sub>2</sub>) accounts for 60% of global warming and has increased from 280 parts per million/volume (ppmv) at the beginning of the industrial revolution to 366 ppmv in 2007 (IPCC, 2007c). Anthropogenic perturbation such as land-use change and agricultural activities including tillage practices (Oorts et al., 2007), fallow periods (Houghton, 2007) and sole cropping (Smith et al., 2007; Guo et al., 2008) have intensified the production rates of CO<sub>2</sub> from soil. The effects of tillage and cropping systems on GHG fluxes have been well studied and documented. In two short-term tillage experiments, higher CO<sub>2</sub> production rates were measured in a moldboard plow treatment compared to no-till treatment in Minnesota (Reicosky and Lindstrom, 1993; Reicosky, 1997). Conversely, Jacinthe et al. (2002) found that CO<sub>2</sub> production rates were higher from a no-till treatment relative to moldboard plow due to greater quantity of mineralizable organic C in no-till soils.

In addition to CO<sub>2</sub>, CH<sub>4</sub> is an important contributor to global warming. Although it is reasonably short-lived and its concentration is only 0.5% that of CO<sub>2</sub>, its absorption of infrared

radiation is twenty times stronger and it has a global warmer potential twenty-three times greater than  $\text{CO}_2$  (IPCC, 2007a) . The anthropogenic sources of  $\text{CH}_4$  include coal mining, natural gas and petroleum extraction, rice paddies, farm and enteric fermentation and biomass burning (Rosenzweig and Hillel, 1998). Most soils however, have the ability to act as effective sinks of  $\text{CH}_4$ . The methane is predominantly used by bacteria in the soil (methanotrophs) which use the methane as a source of C in a process known as methane oxidation. In a study conducted in cooperation with Methane to Markets and Instituto Nacional de Tecnología Agropecuaria (INTA) (2006),  $\text{CH}_4$  production rates in Argentina from enteric fermentation accounted for 66% of GHG farming production rates in 2000; this is equivalent to 58 tons  $\text{CO}_2$ -eq. Several studies have also suggested that cultivation may reduce the  $\text{CH}_4$  oxidation capacity in soils (Hütsch, 1996; Mosier et al., 1997).

Unlike  $\text{CH}_4$ ,  $\text{N}_2\text{O}$  has a long residence time in the atmosphere of approximately 120 years and is a strong infrared absorber (Rosenzweig and Hillel, 1998).  $\text{N}_2\text{O}$  has 296 times the global warming potential of  $\text{CO}_2$  and has the ability to destroy the ozone layer.  $\text{N}_2\text{O}$  production rates from agriculture alone is estimated to be in the order of 4000 Pg-N/yr (Mosier et al., 1998) and since the Industrial Revolution, global  $\text{N}_2\text{O}$  production rates have increased to 319 ppb (IPCC, 2007c). Galloway et al. (2004) investigated N budgets in various regions and found that Latin America has the highest contribution of naturally occurring reactive nitrogen (Nr) from biological nitrogen fixation (BNF). Equivalent to 25% of the global Nr created from terrestrial ecosystems. Nr influences the biogeochemical cycle in the atmosphere, terrestrial, freshwater,

marine and aquatic ecosystems and can either enhance ecosystem productivity or decrease it through nutrient imbalances (Matson et al., 1997).

### **1.3.2 The Impact of Anthropogenic Climate Change in South America**

Countries in South America are experiencing serious impacts from anthropogenic climate change and these impacts are likely to become more severe under current trends. For instance, approximately 85% of all GHG production rates in Latin America are concentrated in just six countries. The majority is accountable to Brazil and Mexico, followed by Argentina, Colombia, Peru and finally Venezuela (de la Torre and Fajnzylber, 2009). Not surprisingly, land-use change is the largest contributor of CO<sub>2</sub> production rates in Latin America.

During the 20<sup>th</sup> century, temperatures in South America increased by approximately 1 °C while sea-level increased by 2-3 mm/yr since the late 1980s (de la Torre and Fajnzylber, 2009). Changes in precipitation patterns have also occurred in most parts of Latin America; for instance, Northeast Argentina has received an increase in annual rainfall while the southwest has seen a steady decline (IPCC, 2007a). The temperature in Argentina has increased between 1.8-3.5°C in the last century (IPCC, 2007b); however, ironically anthropogenic climate change is still not a major issue for mainstream policy implementation in South America (Smith et al., 2007). Although most governments in South America have not issued an umbrella policy for mitigating anthropogenic climate change, there have been small-scale initiatives and mitigation strategies in various countries. These include general land-use regulation and enforcement, forest management including reforestation and agroforestry, sustainable agricultural practices,

renewable energy including biofuel production, and public transportation infrastructure, waste reduction through recycling and composting; and C taxes on fossil fuels (IPCC, 2007c).

### **1.3.3 The Future of Climate Change**

By 2030, if mitigation policies and measures are not implemented, the IPCC forecasts that global GHG production rates will increase by approximately 90%; consequently global temperatures could then increase by as much as 1.7 °C by 2050 and 4.0 °C by 2100 (IPCC, 2007c). Assuming these predictions are correct there is likely to be a severe increase in polar ice melt, which will lead to sea level rise, an increase in natural hazards and increased desertification of land resulting in land becoming unfertile and thus unsuitable for agriculture (IPCC, 2007b). An increase in atmospheric temperature and associated decreases in soil available water will also lead to gradual replacement of tropical forest by savanna in the eastern Amazonia. It will also lead to a decline in crop and livestock productivity with serious consequences to food security (IPCC, 2007c). The projected changes in precipitation will also affect water availability for human consumption and agriculture. For example, central Argentina is expected to see a 12% decrease in precipitation during the summer months and a 5% decrease during the winter. The north of Argentina however, is projected to become exposed to intense rainfall with 60-80% of the high-flood-risk areas becoming affected (IPCC, 2007b).

The IPCC has reported that the institution of conservation agricultural management practices over the coming years will have a significant impact on the ability of our environment to stabilize atmospheric GHGs at lower levels. In order for GHG production rates to stabilize

they need to peak in the next 5 years by approximately 445-490 ppm; this would mean a 50-85% reduction on 2000 levels by 2015 (IPCC, 2007c). The majority of predictions however, indicate that GHGs will continue to rise. Agricultural N<sub>2</sub>O production rates for example, are expected to increase by 35-60% by 2030 because of an escalating need for mineral fertilizers and manure to maintain crop productivity (Smith et al., 2007). If GHG production rates continue unabated, the impacts will become more severe with each passing day, and the consequences will be devastating. While it is accepted that anthropogenic climate change needs a global reaction, leadership by the countries of Latin America could have a positive effect.

#### **1.4 Sustainable Agriculture**

The anticipated increase in GHG production rates and the consequences of anthropogenic climate change has caused increased interest in identifying mitigation strategies. For instance, sustainable agricultural practices have the potential to enhance the C sink in soils and reduce the need for fertilizer addition thus reducing N<sub>2</sub>O production rates (Wright and Hons, 2004; Desjardins et al., 2005). Although more recently agricultural expansion has slowed, crop yields have increased dramatically; this is largely due to the intensification of high-yielding crop varieties, chemical fertilizers (increase of eight-fold over the last 50 years), pesticides, irrigation and mechanization (Naylor, 1996). Presently, global agricultural GHG production rates account for approximately 10-12% of anthropogenic GHGs and although the net flux of CO<sub>2</sub> between the atmosphere and agricultural lands is relatively balanced, N<sub>2</sub>O production rates from agriculture

equals 60% of global anthropogenic production rates (Smith et al., 2007). There are also concerns that modern agricultural practices may be trading short-term increases in food production for long-term losses and degradation of environmental resources (Matson et al., 1997; Foley et al., 2005). However, researchers have found that sustainable agriculture can aid in reducing GHG production rates released from agricultural practices (Lal, 1997; Sey et al., 2008). Smith et al. (2007) examined GHG mitigation potentials for a wide range of agricultural practices including reduced biomass burning, bio-energy crops, water management, residue management, agroforestry, intercropping and land-use change. His study demonstrated that there is a potential of mitigating approximately 5,500-6,000 Mt CO<sub>2</sub>-eq. yr<sup>-1</sup> for all GHGs from agricultural GHG mitigation strategies (Smith et al., 2007). This is because changes in agricultural practices often influence both the quantity and quality of SOM (Wright and Hons, 2004; Bhattacharyya et al., 2009).

Sustainable agriculture practices are often dependent upon land-use and mitigation strategies that enhance and maintain high levels of SOC (Lal, 2006). For instance, several studies have shown that reduced-tillage practices can result in greater aggregation and increased preservation of SOM (Six et al., 2000; Alvaro-Fuentes et al., 2008; Bhattacharyya et al., 2009). In Argentina, agricultural soils have been intensively used for conventional crop production, however recently, a small number of farmers have implemented conservation tillage practices (Studdert and Echeverria, 2000). Mungai and Motavalli (2006) suggested that sustainable agricultural management practices, including complex systems such as intercropping and agroforestry, might help to reduce soil C and N losses. As such, these systems could serve a dual

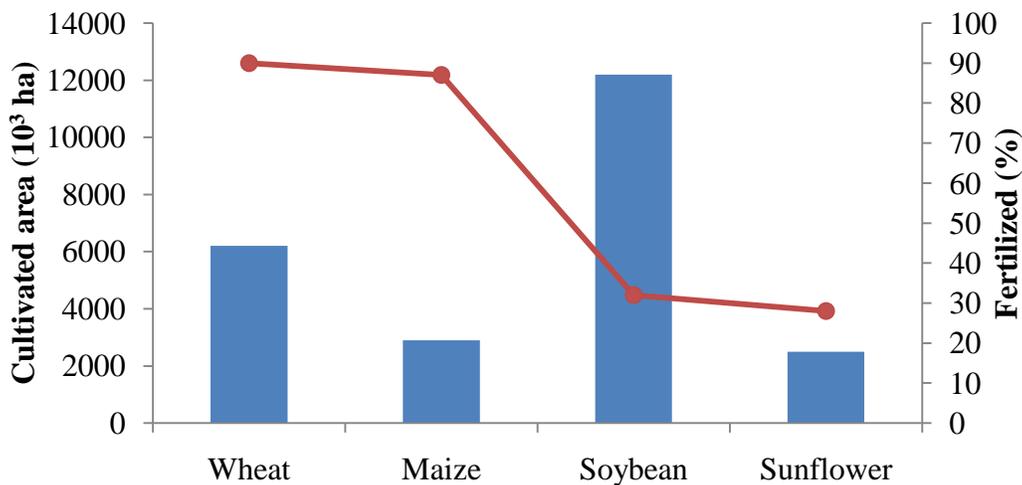
purpose by maintaining crop productivity while simultaneously sequestering C and conserving biodiversity (Mungai et al., 2006). Analysis of agroecosystem dynamics however, remains to be difficult due to the intrinsically complex interactions between components, which often present nonlinear behavior and intricate feedback mechanisms (Tornquist et al., 2009). Moreover, information gathered from field trials is rarely extrapolated to other soils and climates, therefore, simulation models have gained importance in scientific practice to overcome limitations (Tornquist et al., 2009).

#### **1.4.1 Agriculture in South America**

During the beginning of the 20<sup>th</sup> century, the majority of land in Latin America was converted from grassland to agriculture (cereal crops and oil seeds) and cattle production. With the loss of grasslands, heavy cultivation, and the lengthy fallow period, the land has been severely degraded by extreme erosion and loss of SOC. According to Robertson and Paul (2000) after conversion of natural ecosystems to agriculture, SOC will only stabilize to 40-60% of the original pre-cultivation values; moreover with intensive cropping the soil will become severely degraded.

In Argentina, loss of SOM and soil C due to intensive cultivation from agriculture has left the soils heavily eroded and nutrient deficient (Studdert and Echeverria, 2000). Noellemeyer et al. (2008) reported a 55% loss of SOC due to tillage of virgin soils after only 5 years of cultivation. Agricultural land under conservation tillage in Argentina for instance has increased from 25,000 ha in 1988 to more than 7 million ha in 2010 (FAO, 2010). Moreover, Argentina

continues to intensify agriculture and fertilizer use because of a growing concern for increased crop production in order to meet the growing need for food and to maintain a competitive presence in global agricultural markets (Niggli et al., 2009). Although most crops in the Pampa region of Argentina have high fertilizer use (Figure 1.2), the main crop, soybean, uses relatively low fertilizer. This is due to the contribution of BNF linked with its symbiosis with the bacteria *Rhizobium* (Austin et al., 2006). In spite of this, the amount of N gained through BNF or through fertilizer application is inadequate to balance the losses associated with the current agricultural practices that are used in this region (Austin et al., 2006). In the next 50 years, it is estimated that agricultural intensification will continue to accelerate in Argentina; thus, sustainable agricultural practices will be vital in sequestering C (Martinelli et al., 2001).



**Figure 1.2 - Cultivated area of primary cereal crops and percentage of fertilizer use in the Pampas, Argentina (2002-2003) (Austin, 2006). The bars represent cultivated area per crop (10<sup>3</sup> ha) and the line represents percent fertilizer per crop.**

### 1.4.2 The Future of Agriculture

By 2050, arable land per capita will decrease to approximately one-third of present levels; farmers will therefore need to increase their production considerably from the available arable land area (Wild, 2003). Although land productivity should increase due to technological progress and increasing fertilizer and irrigation (Matson et al., 1997), it will likely continue to decline due to greater use of marginal lands with lower productivity. Crop yields are also expected to decline as a consequence of soil degradation and will be amplified by climate change (Lal, 2006). This is disconcerting as Wild (2003) has estimated that in developing countries, food production of  $1,223 \text{ Mt/yr}^{-1}$  will have to increase at a rate of  $2.5\%/yr^{-1}$  between 2000-2025 in order to meet growing food demands. Changes in regional patterns of production and consumption are also expected to rise resulting in greater use of energy for transportation and thus an expected increase in  $\text{CO}_2$  production rates.

However, with improved management practices and technological innovations a reduction of production rates/unit of food produced is achievable through, improved crop varieties and use of legumes to improve crop health and thus decrease the need for fertilizers (Smith et al., 2007). For example, management practices, such as intercropping can maintain and often even increase productivity and can improve regional and presumably global food security (Lal, 2004b). These management practices are expected to become increasingly important as demand for food security increases in the future (de la Torre and Fajnzylber, 2009).

## 1.5 Intercropping

Intercropping is not a new concept, especially within tropical regions; however, it is beginning to gain attention in the temperate region (Austin, 2006). Intercropping is defined as the planting of two or more crops on the same land area at the same time (Sullivan, 2003). It is especially useful as plants have a more effective use of resources compared to sole crops (Li et al., 2001). Implementing an intercropping system also allows for integration of crops using space and labor more efficiently and it is ecologically sustainable. For example, when crops differ in the way they utilize resources than when grown in tandem they can complement each other making better use of resources including water, light and nutrients (Willey, 1990; Malézieux et al., 2009). Intercropping also has the benefit of improving surface moisture since the soil is not as exposed to direct sunlight and thus evaporative loss is diminished (Fukai and Trenbath, 1993). It also reduces runoff and erosion and increase infiltration as the crop canopy provides protection over the soil surface. The vast root system also protects the soil from erosion and aids in water retention (Ofori and Stern, 1987). The classic example of intercropping is that of the ‘three sisters’, maize, beans (*Phaseolus vulgaris* L.) and cucurbits (*Cucurbitaceae* Juss.). These plants were important components of Native American agriculture and were all seeded in the same hole simultaneously. The idea is that the maize will provide the stalk for the beans to climb on, the beans are nutrient-rich to offset what nutrients are taken up by the maize, and the cucurbits grow on the ground and thus keep weeds down and prevent soil water evaporation (Wang et al., 2010). This system provides reciprocal benefits and optimal crop and soil productivity, which allows biomass to be returned and C to be sequestered in the soil (Wang et al., 2010).

More recently, one of the most common intercrop combinations is a cereal-legume intercrop; this is because the inclusion of a legume in crop rotation increases productivity due to the complementary use of N resources. Legumes can also help improve soil physical properties, increase soil microbial activity and restore organic matter as well as increase soil fertility; crop yield and N supply (Ghosh et al., 2007). For instance, the integration of leguminous crop residue, which have low C/N ratios, have been shown to increase C and N retention in temperate agroecosystems (Drinkwater et al., 1998) thus reducing CO<sub>2</sub> and N<sub>2</sub>O production rates. Legumes also have the ability to regulate the internal N cycle via dinitrogen-fixation (N<sub>2</sub>-fixation) (Schipanski et al., 2010) and decrease the need for fertilizer application. Moreover, legume-based intercropping can reduce C and N losses because of the incorporation of crop residue with low carbon/nitrogen ratios (C/N) (Drinkwater et al., 1998). These beneficial effects of legumes in both intercropping and agroforestry system have been well documented (Ofori and Stern, 1987; Drinkwater et al., 1998; Gregorich et al., 2001; Ghosh et al., 2007; Ellert and Janzen, 2008). Studies have shown that agroecosystems that moderate bare fallow periods, increase temporal plant diversity and which rely predominantly on N<sub>2</sub> fixation for N inputs have increased crop yields while N losses are reduced (Drinkwater et al., 1998; Gregorich et al., 2001). Practices such as this have the potential to reduce N<sub>2</sub>O production rates improve N-use efficiency and reduce energy use from fertilizer manufacturing thus avoiding N pollution affects on water and air quality (Mosier et al., 1998). Additionally, a cereal-legume agroecosystem utilizes soil and water resources more efficiently than sole crop systems; this is due to the combination of the tall cereal

with an adventitious root system and the short legume with a deep tap root system and its N<sub>2</sub>-fixing abilities (Tsubo et al., 2003; Prasad and Brook, 2005).

## **1.6 Carbon Sequestration**

In recent years, the potential for reducing the accumulation of GHGs in the atmosphere by sequestering C in the soil has received widespread attention. World soils, if sustainably managed, can globally sequester approximately 1 Pg C/yr, which can offset 0.47 ppm/year of CO<sub>2</sub> in the atmosphere (Lal, 2007). Carbon sequestration is the removal and storage of atmospheric CO<sub>2</sub> through biological and chemical processes (Jose, 2009). In soils, CO<sub>2</sub> is removed from the atmosphere through plant photosynthesis. However, the amount of CO<sub>2</sub> that can be sequestered depends on the type of photosynthetic pathway (C<sub>3</sub>, C<sub>4</sub> or crassulacean acid metabolism) the plant uses and the abiotic factors of the soil environment including variations in light, soil moisture, temperature, and nutrient availability (Uphoff et al., 2006). Variations in photosynthetic capacity have a direct impact on the amount of fixed C that reaches the soil and becomes available for use by heterotrophic soil organisms (Bender, 1971). The process of photosynthesis results in the growth of plant roots and shoots and increased microbial biomass in the soil. Plants then release a portion of their stored C back into the atmosphere through respiration. As plants shed their leaves, and as their roots die, their organic matter decays and some of it can become protected physically and chemically as inert organic matter in the form of humus (Uphoff et al., 2006). The rate of crop residue decomposition is a function of the quality of plant residue and its accessibility to soil organisms (Blanco-Canqui and Lal, 2004). Thus in

order for C to be sequestered in the soil it needs to be protected from microbial degradation within stable microaggregates (<250  $\mu\text{m}$ ), absorbed on the inner surface of clay, or be chemically protected in organic mineral complexes (Lal, 1997).

The ability of soils to sequester C provides a beneficial situation; improving soil quality will not only increase the SOC stock but it can then increase the production of food grains by more than 30-40  $\text{Tg/yr}^{-1}$  (Lal, 2006). For instance, researchers estimate that soils have the ability to sequester approximately 1,900  $\text{Tg/yr}^{-1}$  of C from the atmosphere under various management practices (Barreto et al., 2009). Some conservation management practices include; conservation tillage (Lal, 1997), crop rotation and intercropping (Ahmed and Rao, 1982; Prasad and Brook, 2005), use of cover crops (Buckles et al., 1998), reduced fertilizer use (Studdert and Echeverria, 2000; Aita and Giacomini, 2007), crop residue management (Adiku et al., 2008), reforestation, and agroforestry (Jose, 2009).

Within a given soil and climate, a linear relationship exists between biomass C inputs and SOC (Larson et al., 1972; Rasmussen et al., 1980; Huggins et al., 2007). For instance, the type of crop species plays an important role in C sequestration because residues vary in quantity and quality, which affects their turnover rates in soil (Stewart et al., 2009). Leguminous crops for example, have high-lignin content and are rich in the labile organic fraction, which increases soil aggregation and SOC concentration. Given that the labile fraction is easily degraded, combining these crops with crops of a more complex chemical structure such as maize can be advantageous to the long-term decomposition and cycling of nutrients ( Hobbie, 1992; Blanco-Canqui and Lal, 2004).

## 1.7 Objectives

Increasingly, there is a need to improve our quantitative understanding of C retention by soil under diverse arrangements of soil types, climate and sustainable management practices. Sustainable agricultural management practice including agroforestry, conservation tillage and intercropping have the ability to improve soil physical and chemical qualities and sequester C. However even though intercropping may have the ability to reduce GHG production rates and sequester C, most research has focused on grain yield, resource use, tillage and fertilizer requirement (Prasad and Brook, 2005), and there are currently no known studies on the effect of maize-legume intercrops on GHG production rates, except those in agroforestry (Verchot et al., 2008). In spite the potential for maize-soybean systems to improve N-use efficiency, soil physical properties and sequester C, little research has focused on understanding these characteristics.

**Research Question:** What are the effects of a complex agroecosystem on GHG production rates and soil chemical and physical properties in the Argentine Pampa?

The objectives of this study were therefore:

- 1) To quantify GHG production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) from maize-soybean intercrops compared to maize and soybean sole crops over two field seasons.
- 2) To determine temporal changes in soil chemical and physical properties and field and laboratory GHG production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) from maize-soybean intercropping system, a soybean sole crop system and a maize sole crop system over two field seasons.
- 3) To quantify GHG production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) from soils incubated with  $\text{C}_3$  crop residues (soybean) or  $\text{C}_4$  crop residues (maize).

## 1.8 Hypothesis and Null Hypothesis

- 1) That GHG production rates will be significantly lower in the intercrop treatments compared to the sole crop treatments.

*H<sub>0</sub>*: There will be no significant differences in GHG production rates between treatments.

- 2) Significant temporal changes in soil chemical and physical properties as well as field and laboratory GHG production rates are evident between the intercrop treatments and the sole crop treatments.

*H<sub>0</sub>*: There will be no significant differences in soil chemical and physical properties or field and laboratory GHG production rates from any of the treatments.

- 3) Soils incubated with crop residues will show significantly greater CO<sub>2</sub> and N<sub>2</sub>O production rates than soils incubated with bare soil.

*H<sub>0</sub>*: There will be no significant differences in CO<sub>2</sub> or N<sub>2</sub>O production rates between soils amended with crop residues and bare soil.

## 1.9 Thesis Outline

This thesis is arranged into six chapters, starting with a general introduction and review of the literature. Chapter 2 gives site-specific information including historical context, landscape, climate, soil features and study design. Chapters 3-5 are split into experiments and all include a general introduction and literature review around the specific topic, as well as the statistical and experimental design, the results, discussion and conclusion. The last chapter provides a conclusion that draws from the results from all of the experiments, limitations and ideas for future research.

**Chapter 1:** Introduces the reader to the literature involving all aspects of this thesis including effects of intercropping on soil chemical and physical properties and GHG production rates.

**Chapter 2:** Gives the reader site-specific information including a description of the study site, including the history of the site, landscape features, climate trends in the region and soil features. The chapter concludes with a section on the study design including intercropping design.

**Chapter 3:** Presents the results from the soil chemical and physical properties including bulk density, soil carbon and total nitrogen, the soil carbon/nitrogen and the soil infiltration rate. This chapter examines the correlation between these properties and the significant differences between these properties with depth and with treatment.

**Chapter 4:** Presents the results from the soil carbon dioxide and nitrous oxide production rates taken from the field during 2008-2009 and 2009-2010 from November-February. This chapter then examines the correlation between these production rates and soil temperature and moisture.

**Chapter 5:** Presents the results from the greenhouse gas production rates from a 92 day incubation in which soils were amended with soybean and maize crop residues. This chapter then examines the statistical differences between these production rates and the type of soil used.

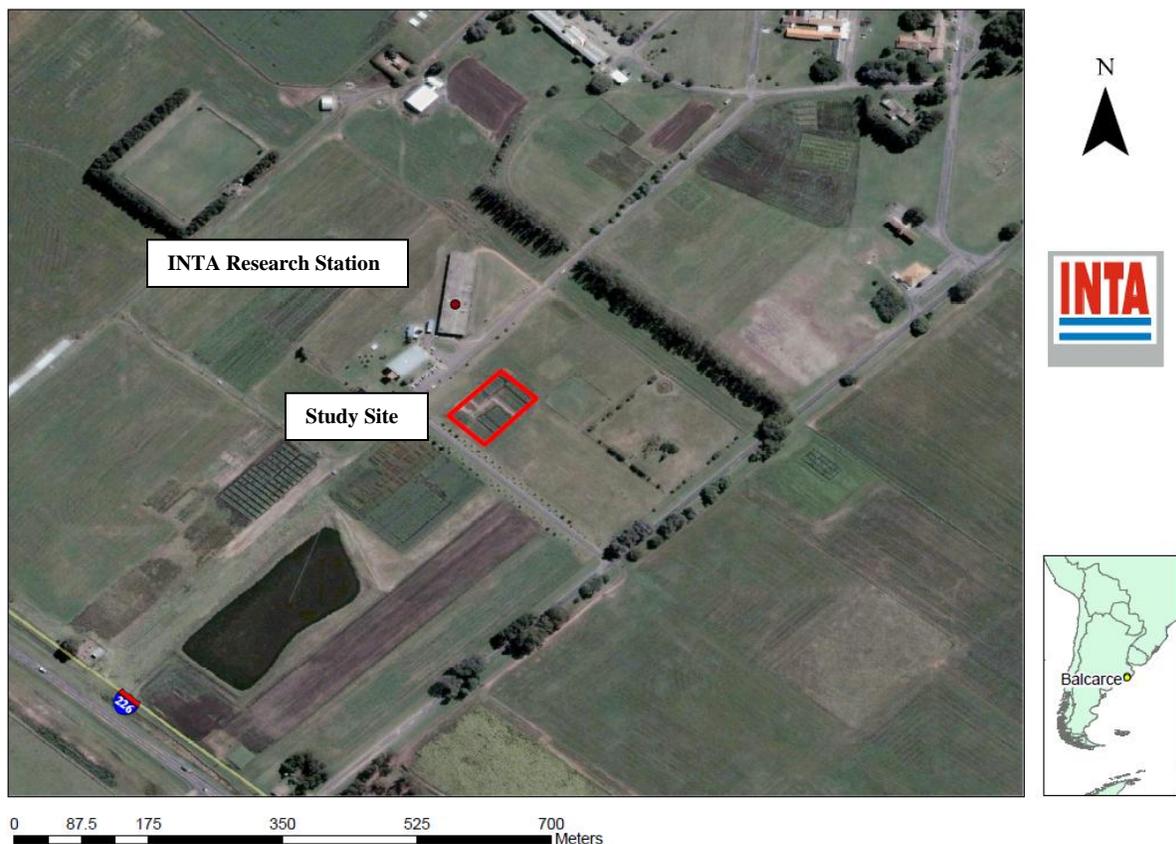
**Chapter 6:** Provides the reader with a conclusion that draws from the results of all of the experiments presented in chapters 3-5. The chapter then examines the limitations to the experiments and suggests further research.

## Chapter 2

### Study Site - Instituto Nacional de Tecnología Agropecuaria (INTA), Balcarce, Argentina

#### 2.0 Introduction

The research site was located at (37°45'55''S, 58°18'11''W) the Instituto Nacional de Tecnología Agropecuaria (INTA) located 15 km northwest of the city of Balcarce, Argentina, in the rolling Pampas (Figure 2.1).



**Figure 2.1 - Location of the field study site of a Randomized Complete Block Design (RCBD) of intercropped maize/soybean and maize and soybean sole crops at the Instituto Nacional de Tecnología Agropecuaria (INTA) located 15 km northwest of the city of Balcarce, Argentina, in the rolling Pampas.**

## 2.1 Climate and Soil Properties

The Rolling Pampa of Argentina are a relatively flat area of 10 million ha that comprises the most productive land in the country. The area is 130 m above sea level and is characterized by a temperate humid (Köpen classification) (Domínguez et al., 2009) with the growing season lasting normally lasting between October and March. The 30-year mean (February 1980 – February 2010) annual precipitation was 860 mm and the mean annual temperature was 13.9 °C (Andrade, 1995) with approximately 80% of the precipitation occurring in the spring and summer (October-March) (Table 2.1). The area surrounding the research station is characterized by low average temperatures during the growing season, with approximately 2409 hours of sunshine per year (Andrade, 1995).

**Table 2.1- 30 year mean (Feb 1980 - Feb 2010) monthly temperature and precipitation at the Instituto Nacional de Tecnología Agropecuaria (INTA, 2010)**

Month	Mean Precipitation (mm)	Mean Temperature (°C)
January	129.4	24.2
February	121.5	22.2
March	113.4	19.0
April	70.2	14.8
May	31.9	10.2
June	20.7	7.9
July	11.7	7.6
August	10.0	8.7
September	40.1	9.9
October	83.6	12.2
November	107.9	15.9
December	119.8	18.5

The soil at the experimental site is developed from eolic sediments from the quaternary period (Fabrizzi et al., 2003). They developed from loess materials under graminaceous vegetation in a temperate climate (Soriano et al., 1992). Over the past 40 years, the graminaceous vegetation was replaced by maize (*Zea Mays* L.) in rotation with wheat (*Triticum aestivum* L.) and soybean [*Glycine max* L. (Merr.)] under conventional tillage (Alvarez et al. 1998). The soils are a mixture of moderately well drained Luvic Phaeozems from the *Mar del Plata* series (FAO) (Studdert and Echeverria, 2000) and fine, mixed thermic Chernozemic Loam (FAO) from the *Balcarce* Series (Domínguez et al., 2009). The texture is 41.1% sand, 35.8% silt and 23.1% clay with an organic matter content of 5.6 g kg<sup>-1</sup> (Andrade, 1995; Domínguez et al., 2009). Soils in this region have high structural stability, low in available phosphorus (P) (Puricelli, 1985); with calcium carbonate content varying in the region between 14-64 g kg<sup>-1</sup> (Prieto et al., 2004).

## **2.2 Regional Context**

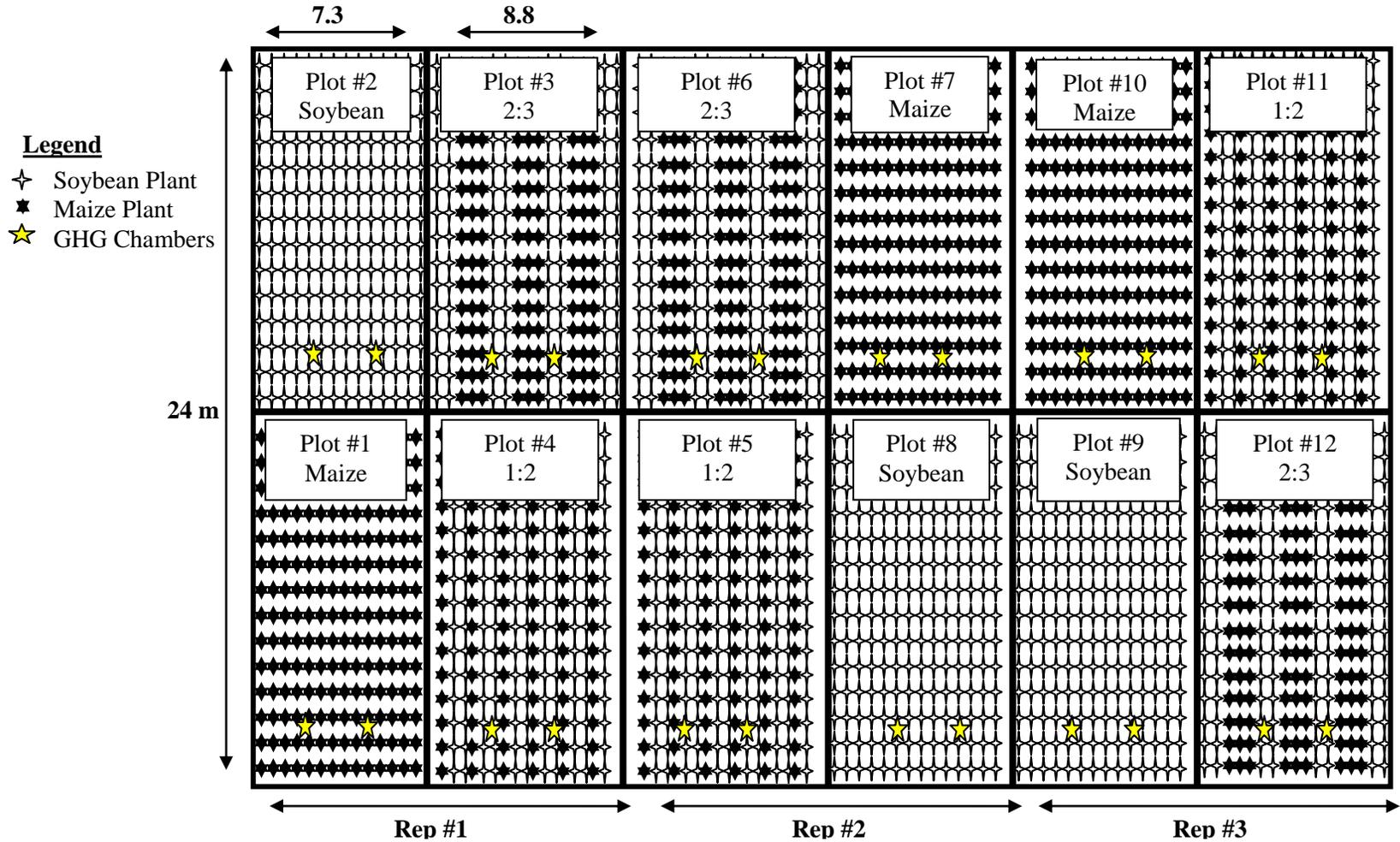
The Argentine Pampa is esteemed as some of the most fertile land for grain production in the world and as such, agricultural land under conservation tillage has increased from 25,000 ha in 1988 to more than 7 million ha in 2010 (FAO, 2010). Moreover, they continue to intensify agriculture and fertilizer use because of a growing concern for increased crop production in order to meet the growing need for food and to maintain a competitive presence in global agricultural markets (Niggli et al., 2009). Although most crops in the Pampa region of Argentina have high fertilizer use (Figure 1.2), the main crop, soybean, uses relatively low fertilizer. This is due to the contribution of biological nitrogen fixation linked with its symbiosis with the bacteria *Rhizobium* (Austin et al., 2006). In spite of this, the amount of N gained through BNF or through fertilizer

application is inadequate to balance the losses associated with the current agricultural practices that are used in this region (Austin et al., 2006). More recently, introduction of glyphosate, genetically modified soybean and the development of planters and drills that have allowed the sustained increase of area under no-till has allowed the region to remain one of the world's largest agricultural hubs (Fabrizzi et al., 2003). However, in the next 50 years, it is estimated that agricultural intensification will continue to accelerate in Argentina and thus, sustainable agricultural practices will be vital (Martinelli et al., 2001).

### **2.3 Study Design**

The study design was a randomized complete block design (RCBD) with four treatments and three replications per treatment. The treatments were a maize sole crop, a soybean sole crop, 1:2 intercrop and 2:3 intercrop (Appendix A). All crops were sown in the same plot since intercrop establishment in 2006. The 1:2 intercrop consisted of one row of maize and two rows of soybean, whereas the 2:3 intercrop consisted of two rows of maize and three rows of soybean (Figure 2.2 & 2.3). The two different intercrop configurations were implemented to evaluate optimum plant density, grain yield, and plant interception of photosynthetically active radiation. Moreover, these configurations are the most commonly used by local growers and producers. The maize sole crop and the maize in the intercrops were sown on October of both study years and were harvested in March of both study years. The soybean sole crop and the soybean in the intercrops were sown in December 2008 and November 2009 and were harvested in May 2009 and April 2010. (Appendix A)

Figure 2.2 - A randomized complete block design (RCBD) of a maize- soybean intercropping system (1:2 & 2:3) and a maize and soybean sole crop system replicated three times at the Instituto Nacional de Tecnología Agropecuaria (INTA) in Balcarce, Argentina. Stars represent location of greenhouse gas (GHG) sampling chambers.





**Figure 2.3 - Field site at INTA, Balcarce, Argentina. A) South-east facing in the 1:2 intercrop (January 4, 2009); B) South-west facing in the 2:3 intercrop (February 8, 2010)**

## **Chapter 3**

### **Soil Chemical and Physical Properties of a Complex Agroecosystem in the Argentine Pampa**

#### **3.0 Introduction**

Soil, the foundation for most terrestrial life, has unparalleled complexity. It is a layer of minerals altered physically and chemically from the bedrock by geological weathering, nutrient cycling and biomass growth and decay. One gram of soil can contain billions of fungi and bacteria and thousands of plant and animal species (Uphoff, 2006). Soil is the principal medium for plant growth and thus the primary environmental resource that supports agriculture. Since the emergence of agriculture however, soil management has been a challenge. Nearly a century ago, farmers used long-fallow periods to repair soil quality (both physical and chemical properties) lost by cultivation. However, with increasing demand for food security along with an ever-growing population settled agriculture evolved and farmers developed techniques to attempt to improve soil quality (Wood et al., 2000).

One of the single most important measures of soil quality is soil organic matter (SOM). SOM is vital as it facilitates the aggregation of soil particles thus reducing erosion. It also provides a source of carbon (C) and energy for microbes and stores and supplies nutrients such as nitrogen (N), phosphorus (P) and sulphur (S) (Bhattacharyya et al., 2009; Wood et al., 2000). There are two major ways to foster SOM in agriculture; the first is to increase soil C using crop residue addition, cover crops, compost and manure addition; the second is to reduce the loss of

soil C by slowing decomposition. Any management practice that increases the photosynthetic input of C or regulates respiration will essentially sequester C.

Lal (2004) has suggested that regions that have lost the most soil organic carbon (SOC) have the highest potential for C sequestration through agricultural management practices. These regions are characterized by soils that have been severely degraded from intensive farming practices. For instance, researchers believe that in the Argentine Pampa, improved management has high potential of sequestering C (Noellemeyer et al., 2008). However, the efficiency of C sequestration by vegetation and management practices in diverse systems varies significantly due to the physiological characteristics, growth rates, biomass accumulation and other environmental factors. Therefore, it is important to optimize ecosystems to efficiently and effectively sequester C from the atmosphere (Wang et al., 2010).

Intercropping, where more than one crop is grown on the same land unit at the same time may be a more sustainable agricultural production system compared to conventional or sole crop systems (Sullivan, 2003). One of the first known examples of intercropping is that of the ‘three sisters’, maize (*Zea mays* L.), beans (*Phaseolus vulgaris* L.) and cucurbits (*Cucurbitaceae* Juss.). These plants were important components of Native American agriculture and were all seeded in the same hole simultaneously. The idea is that the maize will provide the stalk for the beans to climb on, the beans are nutrient-rich to offset what nutrients the maize takes up, and the cucurbits grow on the ground, thus keeping weeds down and preventing soil water evaporation. This system provides reciprocal benefits, optimal crop and soil productivity, which allows for biomass to be returned and C to be sequestered in the soil (Wang et al., 2010). Intercropping systems also

have the ability to increase levels of SOC and N resulting in an increase in crop biomass production and ultimately in an increase in the amount of crop residue returned to the soil (Kong et al., 2005). Legume-based intercrops for instance, can reduce losses of C and N because they increase the overall system complexity and allow for the complementary use of resources in time and space (Prasad and Brook, 2005).

In the Argentine Pampa, a region that is presently a global exporter of maize and soybean (*Glycine max* L.), landowners are adopting maize-legume intercrops to minimize soil degradation and losses of SOC. The addition of a legume with maize in the same system regulates the internal N cycle via dinitrogen fixation (N<sub>2</sub>) (Schipanski et al., 2010), and thus decreases the need for N fertilizer (Inal et al., 2007). In cereal-legume systems, cereal crops such as maize form higher canopy structures and have root systems that grow to greater depths, thus differing spatially and temporally in their use of resources (Willey, 1990). Moreover, the greater canopy cover can protect the soil from the impact of rain and Aeolian erosion (Willey, 1990). In a study conducted by Alegre and Rao (1996) in Peru, contoured hedgerows of Guaba (*Inga edulis* L.) intercropped with rice or cowpea sole crops conserved on average, 287 mm of water and 73 t ha<sup>-1</sup> of soil annually. Other studies noted that water-use efficiency (WUE) of a maize-bean intercrop was equivalent or higher than a maize sole crop and higher in WUE than bean sole crops (Tsubo et al., 2003). Legume-based intercrops also have the ability to sequester C (Wang et al., 2010). The addition of crop residues with both a low C/N ratio from the maize and a high C/N ratio from the soybean allows for a significant increase in the retention of C and N in

the soil (Drinkwater et al., 1998; Vandermeer et al., 1998; Gregorich et al., 2001; Mungai and Motavalli, 2006).

Despite the potential for maize-soybean systems to improve N-use efficiency and soil physical properties and sequester C, little research has focused on understanding these soil characteristics. Presently, the majority of research on intercrop systems has focused on grain yield (Tsubo et al., 2003), resource use (Willey, 1990), tillage (Hernanz et al., 2009), soil quality (Drinkwater et al., 1998), N fixation (Ofori and Stern, 1987; Stern, 1993) and fertilizer requirement (Studdert and Echeverria, 2000). Thus, a critical first step in assessing the potential of a soil to mitigate GHGs is to evaluate initial C and N levels in the soil as well as the contribution of C and N from plant residue (Ordonez et al., 2008). Therefore, a field experiment was executed to compare soil chemical and physical characteristics in a maize-soybean intercrop compared to maize and soybean sole crops. Such information is crucial, as there is still little understanding of the multidimensional processes involved in how soil characteristics change both temporally and spatially in complex agroecosystems.

The objectives of this study were therefore:

- 1) Determine temporal changes in soil chemical and physical properties of a maize-soybean intercropping system at various ratios and a soybean sole crop system and a maize sole crop system over a two field seasons.
- 2) Quantify differences in SOC and TN of a maize-soybean intercropping system at various ratios compared to a soybean sole crop system and a maize sole crop system.
- 3) To examine the correlation between soil chemical and physical properties of a maize-soybean intercropping systems at various ratios and a soybean sole crop system and a maize sole crop system.

## **3.1 Materials and Methods**

### **3.1.1 Soil Chemical and Physical Characteristics**

Soil was sampled using a soil corer with a 5 cm inner-diameter in February 2009 (Y1) and 2010 (Y2) at depths of 0-10, 10-20, 20-40 and 40-80 cm. Two random samples per replicate were extracted, weighed and then composited from each block using the cone and quarter technique (Schumacher et al., 1990), totaling 3 samples per depth per treatment. Soils were then transported back to the soil laboratory where a sub-sample of 20 grams was oven-dried at 105°C for 48 hours to determine bulk density.

The remaining soil was air-dried and passed through a 2 mm sieve to remove the coarse mineral fraction and large plant residue fragments, it was then labeled, packaged and transported to the Soil Ecosystem Dynamics Laboratory in Waterloo, Ontario, Canada. Soil carbonates were

removed by treating 2 grams of soil with 50 mL of 0.5 M hydrochloric acid (HCl). The mixture was then shaken 3 times over a 24-h period for 10 minutes using a laboratory shaker. After the soil settled, the acid-solution was removed by using a 5-mL Pipette which was replaced with ultra-pure water that was removed and replaced once daily for 4 days; each time the water solution was removed using the pipette. The soils were then dried in an oven at 40 °C for 2 days (Midwood and Boutton, 1998). After carbonate removal, the soils were ground to a fine powder (<250µm) using a Retsch ball mill (MM 200 PA; Haan, Germany), weighed into tin capsules and analyzed for SOC and total nitrogen (TN) concentrations using a Costech Elemental Analyzer (Model 4010; Cernusco, Italy). Values obtained from the bulk density calculations were used to convert the SOC and N concentrations ( $\text{g kg}^{-1}$ ) into mass per area ( $\text{g m}^{-2}$ ) for each depth.

### **3.1.2 Soil Water Infiltration**

Soil water infiltration was measured on January 19<sup>th</sup>, 2009 using a tension infiltrometer (Model 2028 D20; Goleta, CA, U.S.A). Two infiltrometers were set up per replicate plot; four meters from the plot edge to curtail any border effects. Hydraulic contact between the infiltrometer disk and the soil was established by removing any plant debris and then leveling the soil with 500 g of (0.01 m thick) fine sand. A nylon guard cloth (53 µm equivalent pore size) was placed between the soil and the disk to prevent slumping of the soil and to ensure contact. The infiltration disk was then soaked in water for 5 minutes and then placed on top of the sand. Measurements were then taken for 180 minutes to ensure measurement of unsteady infiltration rate and steady-state infiltration rate at a 3 cm tension. Infiltration rates (IR) were considered steady when the rate of fall of the water level in the reservoir was constant for approximately 10

minutes (Reynolds, 1993) and when the same  $R^2$  value was obtained over 4-5 consecutive measurements (Reynolds and Elrick, 1991).

### **3.1.3 Statistical Analysis**

All data was examined for homogeneity of variance using the Levene Test and goodness of fit using the Kolmogorov-Smirnov (K-S) Test and were found to have a normal distribution. A one-way analysis of variance (ANOVA) was run in SPSS (SPSS Science Inc., v. 15.0, 2006) and was used to compare differences between treatments and years. Any significant differences were further analyzed using Tukey's least significant difference multiple comparison test (Steel et al., 1997; Zar, 2007). For all statistical analyses, the threshold probability level was  $P < 0.05$ . Infiltration rates and steady-IR values along with average (0-80cm) bulk density, SOC, soil TN, C/N ratio, C stock and the N, from all treatments were used from year 1 (Y1) (2008-2009) to calculate the correlation coefficient matrix. A correlation coefficient matrix was only completed for Y1 as soil water infiltration was only measured during this year.

## 3.2 Results

### 3.2.1 Soil Chemical and Physical Characteristics

Bulk density ( $\text{g cm}^{-3}$ ) significantly differed with depth in all treatments below 20 cm in depth in both years (Table 3.1 & 3.2). Similarly, SOC concentration ( $\text{g kg}^{-1}$ ), soil total N (TN) concentration ( $\text{g kg}^{-1}$ ), C/N Ratio, SOC stock ( $\text{g m}^{-2}$ ) and the soil TN stock ( $\text{g m}^{-2}$ ) were all significantly different between depths in all treatments, from the 40 to 80 cm in both years of study. No significant differences in these soil characteristics were observed at a depth of 0-20 cm.

In Y1, SOC concentration ( $\text{g kg}^{-1}$ ) and stock ( $\text{g m}^{-2}$ ) differed significantly between all depths and between treatments but generally, the top 20 cm were significantly different from that at a depth of 40 cm with the SOC concentration always decreasing with depth. In year 2 (Y2) (Table 3.2) and the mean of both years (Table 3.3) there were significant differences in SOC concentration between the sole crops and intercrops at a depth of 40-80 cm. Regardless of treatment, SOC concentration decreased with depth except in Y2 when there was a slight increase in the 10-20 cm layer in all treatments. In 0-10 and 10-20 cm depths, the soybean sole crop had the lowest SOC content ( $\text{g kg}^{-1}$ ) in Y2 (2009-2010). The C/N varied greatly depending on the treatment with a general trend of significant differences between the 0-10 cm and 40-80 cm vs. the 10-20 cm and 20-40 cm depths. All other soil physical properties (Soil TN, SOC stock and Soil TN stock) showed statistical differences at 20 to 80 cm depth.

SOC concentration differed statistically in Y1 between treatments, within years (denoted by uppercase letters) at all depths except 20-40 cm. However, SOC did not significantly differ in Y2 except in the 40-80 cm where SOC concentrations were higher in the sole crops by an average of  $0.8 \text{ g kg}^{-1}$ . In both soil TN concentration and the C/N, there were no statistical differences between treatments. However, the SOC stock and soil TN stock had similar trends with significant differences between treatments in Y1 in the top 10 cm and in Y2 significant differences between the sole crop and the intercrops in the 20-40 cm depth.

When comparing differences between years within treatments there were only a few statistical differences; in the 0-10 cm depth soybean differed in SOC, TN, SOC stock and C/N with a decrease in concentration from Y1 to Y2 in all soil properties except soil TN concentration where Y2 was slightly greater. The top 10 cm of the maize sole crop increased from Y1 to Y2 by  $50 \text{ g kg}^{-1}$  in the N stock. While in the 10-20 cm depth the 1:2 intercrop also decreased in Y2 in both SOC concentration and TN stock.

**Table 3.1 - Soil chemical and physical properties (0-80 cm) in maize and soybean sole crops and in 1:2 and 2:3 intercropping systems in Balcarce, Argentina collected during the first growing season (Year 1 - 2008-2009). Standard errors are given in parentheses**

Treatment	Depth (cm)	Bulk Density (g cm <sup>-3</sup> )	Soil Organic C (SOC) (g kg <sup>-1</sup> )	Soil Total N (TN) (g kg <sup>-1</sup> )	C/N Ratio	SOC stock (g m <sup>-2</sup> )	Soil TN stock (g m <sup>-2</sup> )
Soybean	0-10	1.19 (0.1) <sup>A, a</sup>	24.2 (0.2) <sup>A, a</sup>	2.2 (0.1) <sup>A, a</sup>	11.3 (0.4) <sup>A, ab †</sup>	2979 (15) <sup>A, a †</sup>	263 (7) <sup>A, a</sup>
	10-20	1.22 (0.1) <sup>A, ab</sup>	23.4 (0.2) <sup>A, a</sup>	1.9 (0.1) <sup>A, a</sup>	12.2 (0.5) <sup>A, b</sup>	2870 (18) <sup>A, a</sup>	235 (9) <sup>A, a</sup>
	20-40	1.23 (0.1) <sup>A, b</sup>	18.7 (0.3) <sup>A, b</sup>	1.5(0.1) <sup>A, b</sup>	12.8 (0.3) <sup>A, b</sup>	4551 (62) <sup>A, b</sup>	369 (14) <sup>A, b</sup>
	40-80	1.28 (0.1) <sup>A, c</sup>	8.3 (0.2) <sup>AB, c</sup>	0.8 (0.1) <sup>A, c</sup>	10.7 (0.4) <sup>A, a</sup>	2048 (24) <sup>A, c</sup>	192 (6) <sup>A, c</sup>
	<b>Grand Mean</b>	<b>1.20 (0.1)<sup>A</sup></b>	<b>18.6 (1.9)<sup>A</sup></b>	<b>1.6 (0.2)<sup>A</sup></b>	<b>11.7 (0.3)<sup>A</sup></b>	<b>3112 (273)<sup>A</sup></b>	<b>265 (20)<sup>A</sup></b>
Maize	0-10	1.19 (0.1) <sup>A, a</sup>	25.0 (0.1) <sup>B, a †</sup>	2.1 (0.1) <sup>A, a †</sup>	11.3 (0.1) <sup>A, ab</sup>	2892 (42) <sup>B, a</sup>	255 (6) <sup>A, a †</sup>
	10-20	1.23 (0.1) <sup>A, b</sup>	23.5 (0.2) <sup>A, b</sup>	1.9 (0.1) <sup>A, b</sup>	12.5 (0.6) <sup>A, ac</sup>	2874 (24) <sup>A, a</sup>	232 (12) <sup>A, ab</sup>
	20-40	1.25 (0.1) <sup>A, b</sup>	18.3 (0.2) <sup>A, c</sup>	1.4 (0.1) <sup>A, c</sup>	13.1 (0.4) <sup>A, c</sup>	4674 (62) <sup>A, b</sup>	356 (5) <sup>A, c</sup>
	40-80	1.28 (0.1) <sup>A, c</sup>	8.0 (0.1) <sup>AB, d</sup>	0.8 (0.1) <sup>A, d</sup>	10.6 (0.1) <sup>A, b</sup>	2125 (73) <sup>A, c</sup>	201 (7) <sup>A, b</sup>
	<b>Grand Mean</b>	<b>1.24 (0.1)<sup>A</sup></b>	<b>18.6 (1.9)<sup>A</sup></b>	<b>1.5 (0.1)<sup>A</sup></b>	<b>11.9 (0.3)<sup>A</sup></b>	<b>3141 (284)<sup>A</sup></b>	<b>261 (18)<sup>A</sup></b>
1:2	0-10	1.16 (0.1) <sup>B, a</sup>	25.2 (0.1) <sup>AB, a</sup>	2.2 (0.1) <sup>A, a</sup>	11.5 (0.6) <sup>A, a</sup>	2928 (4) <sup>AB, a</sup>	256 (13) <sup>A, a</sup>
	10-20	1.19 (0.1) <sup>B, ab</sup>	24.6 (0.2) <sup>B, a †</sup>	2.1 (0.1) <sup>A, a</sup>	11.3 (0.3) <sup>A, a</sup>	2912 (20) <sup>A, a</sup>	258 (6) <sup>A, a †</sup>
	20-40	1.19 (0.1) <sup>B, ab</sup>	18.9 (0.2) <sup>A, b</sup>	1.5 (0.1) <sup>A, b</sup>	12.6 (0.1) <sup>A, b</sup>	5000 (61) <sup>A, b</sup>	358 (2) <sup>A, b</sup>
	40-80	1.21 (0.1) <sup>B, b</sup>	8.9 (0.1) <sup>B, c</sup>	0.7 (0.1) <sup>A, c</sup>	11.8 (0.5) <sup>A, ab</sup>	2162 (40) <sup>A, c</sup>	184 (6) <sup>A, c</sup>
	<b>Grand Mean</b>	<b>1.18 (0.1)<sup>B</sup></b>	<b>19.4 (2.0)<sup>B</sup></b>	<b>1.7 (0.2)<sup>A</sup></b>	<b>11.8 (0.2)<sup>A</sup></b>	<b>3125 (257)<sup>A</sup></b>	<b>264 (19)<sup>A</sup></b>
2:3	0-10	1.17 (0.1) <sup>B, a</sup>	25.5 (0.4) <sup>B, a</sup>	2.2 (0.1) <sup>A, a</sup>	11.5 (0.3) <sup>A, a</sup>	2979 (39) <sup>AB, a</sup>	259 (8) <sup>A, a</sup>
	10-20	1.18 (0.1) <sup>B, a</sup>	24.3 (0.3) <sup>AB, a</sup>	2.1 (0.1) <sup>A, a</sup>	11.4 (0.7) <sup>A, a</sup>	2858 (30) <sup>A, a</sup>	253 (18) <sup>A, a</sup>
	20-40	1.19 (0.1) <sup>B, a</sup>	19.1 (0.4) <sup>A, b</sup>	1.5 (0.1) <sup>A, b</sup>	12.8 (0.3) <sup>A, b</sup>	4567 (92) <sup>A, b</sup>	357 (2) <sup>A, b</sup>
	40-80	1.22 (0.1) <sup>B, b</sup>	8.9 (0.1) <sup>B, c</sup>	0.8 (0.1) <sup>A, c</sup>	11.0 (0.1) <sup>A, a</sup>	2171 (20) <sup>A, c</sup>	197 (1) <sup>A, c</sup>
	<b>Grand Mean</b>	<b>1.19 (0.1)<sup>B</sup></b>	<b>19.5 (2.0)<sup>B</sup></b>	<b>1.7 (0.2)<sup>A</sup></b>	<b>11.7 (0.3)<sup>A</sup></b>	<b>3145 (265)<sup>A</sup></b>	<b>267 (18)<sup>A</sup></b>

Means followed by different uppercase letters are significantly different ( $p < 0.05$ ) between treatments, within depths.

Means followed by different lowercase letters are significantly different ( $p < 0.05$ ) between depths, within treatments.

Means followed by † are significantly different ( $p < 0.05$ ) between years, within treatments.

**Table 3.2 - Soil chemical and physical properties (0-80 cm) in maize and soybean sole crops and in 1:2 and 2:3 intercropping systems in Balcarce, Argentina collected during the second growing season (Year 2 – 2009-2010). Standard errors are given in parentheses.**

Treatment	Depth (cm)	Bulk Density (g cm <sup>-3</sup> )	Soil Organic C (SOC) (g kg <sup>-1</sup> )	Soil Total N (TN) (g kg <sup>-1</sup> )	C/N Ratio	SOC stock (g m <sup>-2</sup> )	Soil TN stock (g m <sup>-2</sup> )
Soybean	0-10	1.20 (0.1) <sup>A, a</sup>	23.3 (0.2) <sup>A, a †</sup>	2.4 (0.1) <sup>A, a</sup>	9.81 (0.1) <sup>A, a †</sup>	2796 (31) <sup>A, a †</sup>	284.8 (4) <sup>A, a</sup>
	10-20	1.23 (0.1) <sup>A, b</sup>	24.0 (0.3) <sup>A, a</sup>	1.8 (0.1) <sup>A, b</sup>	13.1 (0.3) <sup>A, b</sup>	2960 (39) <sup>A, a</sup>	226.2 (3) <sup>A, b</sup>
	20-40	1.25 (0.1) <sup>A, c</sup>	18.2 (0.1) <sup>A, b</sup>	1.5 (0.1) <sup>A, c</sup>	12.5 (0.1) <sup>A, b</sup>	4568 (35) <sup>A, b</sup>	365.1 (1) <sup>A, c</sup>
	40-80	1.28 (0.1) <sup>A, d</sup>	8.3 (0.2) <sup>A, c</sup>	0.8 (0.1) <sup>A, d</sup>	10.7 (0.7) <sup>A, a</sup>	2130 (52) <sup>A, c</sup>	201 (11) <sup>A, b</sup>
	<b>Grand Mean</b>	<b>1.24 (0.1)<sup>A</sup></b>	<b>18.4 (1.9)<sup>A</sup></b>	<b>1.6 (0.2)<sup>A</sup></b>	<b>11.5 (0.4)<sup>A</sup></b>	<b>3114 (270)<sup>A</sup></b>	<b>269 (19)<sup>A</sup></b>
Maize	0-10	1.20 (0.1) <sup>A, a</sup>	24.1 (0.3) <sup>A, a</sup>	2.4 (0.1) <sup>A, a †</sup>	10.3 (0.2) <sup>A, a</sup>	2896 (46) <sup>A, a</sup>	283 (2) <sup>A, ab †</sup>
	10-20	1.22 (0.1) <sup>AB, ab</sup>	24.4 (0.5) <sup>A, a</sup>	1.9 (0.1) <sup>A, b</sup>	13.1 (0.2) <sup>A, b</sup>	2977 (68) <sup>A, a</sup>	227 (3) <sup>A, b</sup>
	20-40	1.24 (0.1) <sup>A, b</sup>	18.3 (0.3) <sup>A, b</sup>	1.5 (0.1) <sup>A, c</sup>	13.0 (0.6) <sup>A, b</sup>	4549 (79) <sup>A, b</sup>	348 (12) <sup>A, c</sup>
	40-80	1.28 (0.1) <sup>A, c</sup>	8.4 (0.4) <sup>A, c</sup>	0.7 (0.1) <sup>A, d</sup>	10.2 (0.8) <sup>A, a</sup>	1898 (92) <sup>A, c</sup>	186 (5) <sup>A, d</sup>
	<b>Grand Mean</b>	<b>1.24 (0.1)<sup>A</sup></b>	<b>18.5 (2.0)<sup>A</sup></b>	<b>1.6 (0.2)<sup>A</sup></b>	<b>11.6 (0.5)<sup>A</sup></b>	<b>3080 (288)<sup>A</sup></b>	<b>262 (19)<sup>A</sup></b>
1:2	0-10	1.17 (0.1) <sup>B, a</sup>	23.7 (0.1) <sup>A, a</sup>	2.4 (0.1) <sup>A, a</sup>	10.16 (0.3) <sup>A, a</sup>	2788 (38) <sup>A, a</sup>	275 (6) <sup>A, a</sup>
	10-20	1.19 (0.1) <sup>BC, a</sup>	24.9 (0.3) <sup>A, a †</sup>	1.9 (0.1) <sup>A, b</sup>	13.1 (0.1) <sup>A, b</sup>	2970 (47) <sup>A, a</sup>	226 (4) <sup>A, b †</sup>
	20-40	1.19 (0.1) <sup>B, a</sup>	17.7 (0.2) <sup>A, b</sup>	1.4 (0.1) <sup>A, c</sup>	12.5 (0.6) <sup>A, ab</sup>	4222 (63) <sup>B, b</sup>	339 (16) <sup>B, c</sup>
	40-80	1.23 (0.1) <sup>A, b</sup>	8.8 (0.3) <sup>B, c</sup>	0.8 (0.1) <sup>A, d</sup>	10.7 (0.7) <sup>A, a</sup>	2173 (117) <sup>A, c</sup>	204 (5) <sup>A, b</sup>
	<b>Grand Mean</b>	<b>1.20 (0.1)<sup>B</sup></b>	<b>18.8 (1.9)<sup>A</sup></b>	<b>1.6 (0.2)<sup>A</sup></b>	<b>11.6 (0.4)<sup>A</sup></b>	<b>3038 (227)<sup>A</sup></b>	<b>261 (16)<sup>A</sup></b>
2:3	0-10	1.17 (0.1) <sup>B, a</sup>	23.3 (0.3) <sup>A, a</sup>	2.3 (0.1) <sup>A, a</sup>	9.84 (0.3) <sup>A, a</sup>	2730 (28) <sup>A, a</sup>	278 (7) <sup>A, a</sup>
	10-20	1.18 (0.1) <sup>C, a</sup>	24.0 (0.7) <sup>A, a</sup>	1.8 (0.1) <sup>A, b</sup>	13.0 (0.6) <sup>A, b</sup>	2804 (96) <sup>A, a</sup>	216 (5) <sup>A, b</sup>
	20-40	1.19 (0.1) <sup>B, a</sup>	17.8 (0.2) <sup>A, b</sup>	1.5 (0.1) <sup>A, c</sup>	11.7 (0.4) <sup>A, a</sup>	4261 (32) <sup>B, b</sup>	363 (9) <sup>B, c</sup>
	40-80	1.21 (0.1) <sup>A, b</sup>	8.9 (0.3) <sup>B, c</sup>	0.8 (0.1) <sup>A, d</sup>	10.8 (0.7) <sup>A, a</sup>	2170 (119) <sup>A, c</sup>	201 (12) <sup>A, b</sup>
	<b>Grand Mean</b>	<b>1.19 (0.1)<sup>B</sup></b>	<b>18.4 (1.8)<sup>A</sup></b>	<b>1.6 (0.2)<sup>A</sup></b>	<b>11.3 (0.4)<sup>A</sup></b>	<b>2991 (235)<sup>A</sup></b>	<b>264.6 (20)<sup>A</sup></b>

Means followed by different uppercase letters are significantly different (p<0.05) between treatments, within depths.

Means followed by different lowercase letters are significantly different (p<0.05) between depths, within treatments.

Means followed by † are significantly different (p<0.05) between years, within treatments.

**Table 3.3 - Soil chemical and physical properties (0-80 cm) in maize and soybean sole crops and in 1:2 and 2:3 intercropping systems in Balcarce, Argentina. Values are grand means of both year 1 and year 2 (2008-2010). Standard errors are given in parentheses.**

Treatment	Depth (cm)	Bulk Density (g cm <sup>-3</sup> )	Soil Organic C (SOC) (g kg <sup>-1</sup> )	Soil Total N (TN) (g kg <sup>-1</sup> )	C/N Ratio	SOC stock (g m <sup>-2</sup> )	Soil TN stock (g m <sup>-2</sup> )
Soybean	0-10	1.20 (0.1) <sup>A, a</sup>	23.7 (0.2) <sup>A, a</sup>	2.3 (0.1) <sup>A, a</sup>	10.6 (0.4) <sup>A, a</sup>	2888 (44) <sup>A, a</sup>	274 (6) <sup>A, a</sup>
	10-20	1.20 (0.1) <sup>A, b</sup>	23.7 (0.2) <sup>A, a</sup>	1.9 (0.1) <sup>A, b</sup>	12.6 (0.3) <sup>A, b</sup>	2915 (28) <sup>A, a</sup>	231 (5) <sup>A, b</sup>
	20-40	1.24 (0.1) <sup>A, c</sup>	18.4 (0.1) <sup>A, b</sup>	1.5 (0.1) <sup>A, c</sup>	12.4 (0.1) <sup>A, b</sup>	4559 (32) <sup>A, b</sup>	367 (6) <sup>A, c</sup>
	40-80	1.28 (0.1) <sup>A, d</sup>	8.3 (0.1) <sup>A, c</sup>	0.8 (0.1) <sup>A, d</sup>	10.7 (0.4) <sup>A, a</sup>	2089 (32) <sup>A, c</sup>	196 (6) <sup>A, d</sup>
	<b>Grand Mean</b>	1.24 (0.1) <sup>A</sup>	18.5 (1.3) <sup>A</sup>	1.6 (0.1) <sup>A</sup>	11.6 (0.2) <sup>A</sup>	3113 (188) <sup>A</sup>	267 (14) <sup>A</sup>
Maize	0-10	1.20 (0.1) <sup>A, a</sup>	24.5 (0.1) <sup>B, a</sup>	2.2 (0.1) <sup>A, a</sup>	10.8 (0.3) <sup>A, a</sup>	2894 (28) <sup>A, a</sup>	269 (9) <sup>A, a</sup>
	10-20	1.20 (0.1) <sup>AB, b</sup>	23.9 (0.2) <sup>B, a</sup>	1.9 (0.1) <sup>A, b</sup>	12.8 (0.3) <sup>A, b</sup>	2925 (40) <sup>A, a</sup>	230 (5) <sup>A, b</sup>
	20-40	1.24 (0.1) <sup>A, c</sup>	18.3 (0.2) <sup>A, b</sup>	1.4 (0.1) <sup>A, c</sup>	13.1 (0.3) <sup>A, b</sup>	4612 (53) <sup>A, b</sup>	353 (6) <sup>A, c</sup>
	40-80	1.28 (0.1) <sup>A, d</sup>	8.2 (0.3) <sup>A, c</sup>	0.7 (0.1) <sup>A, d</sup>	10.4 (0.3) <sup>A, a</sup>	2012 (73) <sup>A, c</sup>	194 (5) <sup>A, d</sup>
	<b>Grand Mean</b>	1.24 (0.1) <sup>A</sup>	18.0 (1.3) <sup>A</sup>	1.6 (0.1) <sup>A</sup>	11.9 (0.3) <sup>A</sup>	3111 (198) <sup>A</sup>	261 (13) <sup>B</sup>
1:2	0-10	1.17 (0.1) <sup>B, a</sup>	24.5 (0.3) <sup>B, a</sup>	2.3 (0.1) <sup>A, a</sup>	10.8 (0.4) <sup>A, a</sup>	2858 (36) <sup>A, a</sup>	266 (8) <sup>A, a</sup>
	10-20	1.19 (0.1) <sup>BC, ab</sup>	24.7 (0.2) <sup>B, a</sup>	2.0 (0.1) <sup>A, b</sup>	12.2 (0.4) <sup>A, b</sup>	2941 (26) <sup>A, a</sup>	242 (8) <sup>A, a</sup>
	20-40	1.19 (0.1) <sup>B, ab</sup>	18.3 (0.3) <sup>A, b</sup>	1.5 (0.1) <sup>A, c</sup>	12.5 (0.3) <sup>A, b</sup>	4361 (74) <sup>B, b</sup>	349 (8) <sup>A, b</sup>
	40-80	1.22 (0.1) <sup>B, b</sup>	8.8 (0.1) <sup>B, c</sup>	0.8 (0.1) <sup>A, d</sup>	11.2 (0.5) <sup>A, ab</sup>	2168 (55) <sup>A, c</sup>	194 (5) <sup>A, c</sup>
	<b>Grand Mean</b>	1.19 (0.1) <sup>B</sup>	19.0 (1.3) <sup>B</sup>	1.6 (0.1) <sup>A</sup>	11.7 (0.2) <sup>A</sup>	3082 (168) <sup>B</sup>	263 (12) <sup>B</sup>
2:3	0-10	1.17 (0.1) <sup>B, a</sup>	24.4 (0.5) <sup>B, a</sup>	2.3 (0.1) <sup>A, a</sup>	10.7 (0.4) <sup>A, a</sup>	2854 (60) <sup>A, a</sup>	268 (6) <sup>A, a</sup>
	10-20	1.18 (0.1) <sup>C, a</sup>	24.0 (0.4) <sup>AB, a</sup>	2.0 (0.1) <sup>A, b</sup>	12.2 (0.5) <sup>A, b</sup>	2831 (46) <sup>A, a</sup>	235 (12) <sup>A, b</sup>
	20-40	1.19 (0.1) <sup>B, ab</sup>	18.5 (0.4) <sup>A, b</sup>	1.5 (0.1) <sup>A, c</sup>	12.3 (0.3) <sup>A, b</sup>	4414 (81) <sup>A, b</sup>	360 (4) <sup>A, c</sup>
	40-80	1.22 (0.1) <sup>B, b</sup>	8.9 (0.2) <sup>B, c</sup>	0.8 (0.1) <sup>A, d</sup>	10.9 (0.3) <sup>A, a</sup>	2173 (54) <sup>A, c</sup>	199 (6) <sup>A, d</sup>
	<b>Grand Mean</b>	1.19 (0.1) <sup>B</sup>	18.9 (1.3) <sup>B</sup>	1.6 (0.1) <sup>A</sup>	11.5 (0.2) <sup>A</sup>	3068 (174) <sup>B</sup>	266 (13) <sup>A</sup>

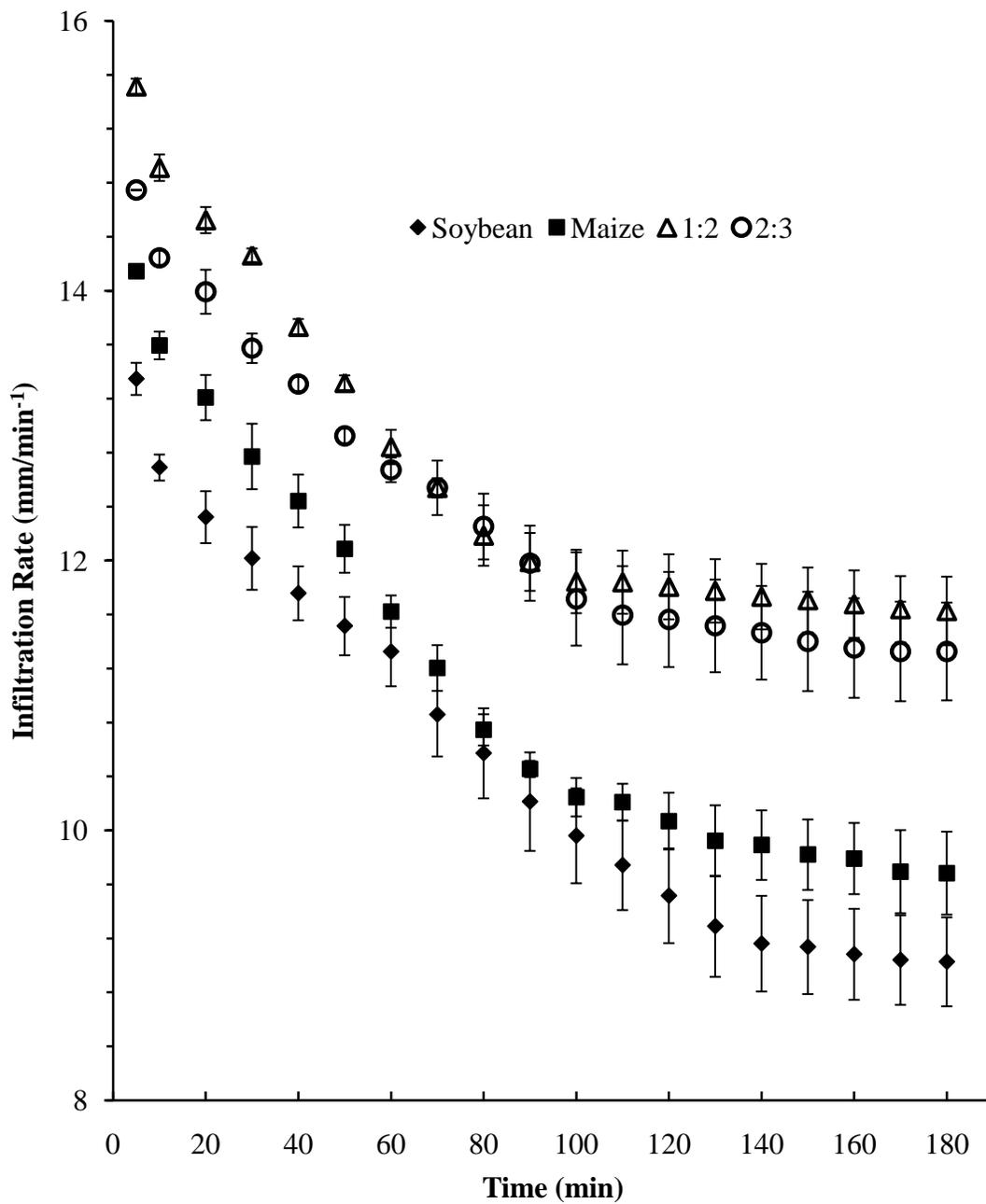
Means followed by different uppercase letters are significantly different ( $p < 0.05$ ) between treatments, within depths.

Means followed by different lowercase letters are significantly different ( $p < 0.05$ ) between depths, within treatments.

### 3.2.2 Soil Water Infiltration

The highest infiltration rate occurred in the 1:2 intercrop (12.70 mm/min<sup>-1</sup>), followed by the 2:3 intercrop (12.39 mm/min<sup>-1</sup>), the maize sole crop; (11.13 mm/min<sup>-1</sup>) and the soybean sole crop (10.55 mm/min<sup>-1</sup>) (Figure 3.1). This corresponded to a steady state IR of 11.76 mm/min<sup>-1</sup> (1:2 intercrop), 11.52 mm/min<sup>-1</sup> (2:3 intercrop), 9.97 mm/min<sup>-1</sup> (maize sole crop) and 9.41 mm/min<sup>-1</sup> (soybean sole crop).

Correlation analysis of the eight soil physical and chemical characteristics (Table 3.4) resulted in a significant correlation in approximately half of the soil physical properties in Y1. Among the negative correlations were bulk density and SOC, soil TN, IR and steady IR as well as C/N ratio and soil TN and N stock and C/N ratio. Significantly positive correlations were found between SOC and both IR and Steady IR.



**Figure 3.1 - Mean soil water infiltration rate as a function of time ( $\text{mm}/\text{min}^{-1}$ ) for both the 1:2 and 2:3 maize/soybean intercrops and the maize and soybean sole crops ( $n=6$ ) using a tension infiltrometer at -3 cm tension for 180 min.**

**Table 3.4 - Correlation coefficient matrix of soil characteristics from Y1 (December 2009-January 2010) (n=12). All characteristics excluding the infiltration rate and the steady-state infiltration rate are an average of 0 - 80 cm depths.**

	<b>Bulk Density</b>	<b>SOC</b>	<b>Soil TN</b>	<b>C/N Ratio</b>	<b>C stock</b>	<b>N stock</b>	<b>IR</b>	<b>Steady IR</b>
<b>Bulk Density</b>	1.000							
<b>SOC</b>	-.885**	1.000						
<b>Soil TN</b>	-.639*	.712**	1.000					
<b>C/N Ratio</b>	.124	-.142	-.762**	1.000				
<b>C Stock</b>	-.004	.458	.365	-.090	1.000			
<b>N Stock</b>	-.169	.336	.850**	-.918**	.444	1.000		
<b>IR</b>	-.833**	.862**	.452	.168	.264	.013	1.000	
<b>Steady IR</b>	-.814**	.837**	.434	.176	.246	-.004	.994**	1.000

Values followed by \* and \*\* are significantly correlated at  $p < 0.05$  and  $p < 0.01$  respectively.

### 3.3 Discussion

#### 3.3.1 Soil Chemical and Physical Characteristics

The results from this study on soil physical and chemical properties from all treatments correspond to other studies from the Buenos Aires Province, Argentina (Bermejo and Suero, 1981; Studdert and Echeverria, 2000; Fabrizzi et al., 2003; Aparicio and Costa, 2007; Domínguez et al., 2009). Bermejo and Suero (1981) reported bulk density values that ranged from 1.22 to 1.26 g/cm<sup>-3</sup> for plowed soils at INTA Balcarce, Argentina. However, Ferreas et al. (2000) reported values of 1.44-1.52 g/cm<sup>-3</sup> in the top 0-10 cm of plowed soils at INTA with continuous cropping. Although these higher bulk density values could be related to a difference in cropping systems most likely they are a product of soil composition. It is likely that the soil samples from this field site at INTA could have a higher clay content than the field site sampled by Ferreas et al. (2000). Although bulk density is a product of soil composition it is also highly correlated to tillage practices. In a several long-term studies conducted by Aparicio and Costa (2007) they found that the type of cropping system was only secondary to the effect of tillage practice on bulk density.

Overall, the intercropping treatments demonstrated little influence on soil physical and chemical properties with statistical differences seen only in the SOC concentration (g kg<sup>-1</sup>). This was anticipated; however, as equilibrium in soil properties are reportedly reached after a period of five or more years in the rolling pampas (Alvarez et al., 1998; Andriulo et al., 1999). In, one study in the Argentine Pampa there were no detectable differences in SOC after 11 years of

maize/soybean intercrop establishment (Studdert and Echeverria, 2000). However, Malhi et al. (2008) suggested that since soils in the Argentine Pampa are inherently rich in SOC, it might prohibit discernible increases in SOC and N over the short-term (3-5 years). Although this study was in its infancy and thus presents baseline data it is still important to quantify changes in soil quality after intercrop establishment in order to fully understand the complexity the soil system.

Soil bulk density ( $\text{g cm}^{-3}$ ), SOC, soil TN and C/N were significantly different with depth as is reported in most agricultural studies (Chabbi et al., 2009; Hernanz et al., 2009; Halpern et al., 2010). For instance, conventional tillage studies have reported significant differences between the top 20 cm and the lower depths (Wright and Hons, 2004; Hernanz et al., 2009). Tillage promotes SOM loss through the incorporation of crop residues into the soil, which causes increased microbial activity due to soil oxidation and a disruption of the macroaggregates (Halpern et al., 2010). Comparable to other studies, SOC and TN had significantly higher concentrations ( $\text{g kg}^{-1}$ ) in the upper soil layer and decreased with depth (Halvin et al., 1990; Kramer and Gleixner, 2008). In longer-term studies however, some researchers have found that there can be substantial losses of SOC in the surface layers. For instance, in a three year no-till maize cropping system, Verma et al., (2005) found that losses of SOC in the top 15 cm ranged from 80-130  $\text{g C m}^{-2}$ . The higher C/N ratio in upper soil layers is likely a result of a greater accumulation of crop residues due to crop roots however; the decrease in the C/N ratio after 60 cm is likely due to the lack of readily decomposable plant material and thus is dominated by microbially derived products.

In this study, there were no significant differences in SOC or TN between treatments within depths most likely because of the early phase of intercrop establishment. Conversely, other studies have found that crop rotation had significant impacts on the storage of SOC and TN in the upper surface layers (Halvin et al., 1990; West and Post, 2002; Bhattacharyya et al., 2009). Kramer and Gleixner (2008) found that in the 20-40 and 40-60 cm soil depth, SOC was higher in a maize cultivated soil than in a rye-cultivated (*Secale cereale* L.) soil. This variation in C with depth reflects differences in plant residue input. For instance, most of the residues in the rye plot are at the surface layer compared to an increase in residues with depth in the maize plot. Kramer and Gleixner (2008) found the greatest abundance of roots within the top 0-20 cm depth in the rye plot compared to roots found as deep at the 40-60 cm depth in the maize plot. This is important since rhizodeposition accounts for the release of up to 20% of photosynthetically fixed C by roots, additionally, the size of the roots reflect the rate of root exudates which then accounts for an increased C content in the soil profile.

Recent studies show that the location of C and N within the soil profile determines its mean residence time (Chabbi et al., 2009) and therefore its availability to soil microbes. In a long-term field study comparing CT to NT in a sorghum (*Sorghum bicolor* L.)/wheat (*Triticum aestivum* L.) /soybean, a wheat/soybean and a soybean sole crop, researchers found that the soybean sole crop generally had the lowest SOC and TN concentrations in the top 5 cm (Wright and Hons, 2004). This was attributed to the soybean contributing the least amount of C-rich residue to the soil than residues from more intensive cropping sequences. However, in a 40 year study, with a rotational cropping system of wheat-grain-sorghum-fallow, researchers found no

measurable differences in soil properties measured after the top 5 cm (McVay et al., 2006). West and Post (2002) compiled results from 67 long-term experiments (5+ yrs) and found that there was no mean increase in SOC after converting from a maize sole crop to a maize-soybean intercropping system. Again, this was attributed to a decrease in residue C input or possible SOC loss as there were correlations found between SOC and soil residue inputs (Rasmussen et al., 1980; Lal and Bruce, 1999). Studies have shown that even in intensively managed agroecosystems, the composition of plant species as well as litter-quality markedly affect SOM turnover. Moreover, the rotational complexity of crops does not always result in significant increases in SOC (West and Post, 2002).

In the top 0-10 cm depth, SOC concentrations ( $\text{g kg}^{-1}$ ) varied only narrowly between the maize sole crop, the 1:2 and 2:3 intercropping system and there were no significant differences in soil TN ( $\text{g kg}^{-1}$ ) between treatments at any depth. Comparably, Vachon and Oelbermann (2010) found that N input from crop residues were evenly distributed between soybean and maize sole crops and 1:2 and 2:3 maize-soybean intercrops. They attributed this to the incorporation of leguminous soybeans in the intercrops as well as the application of inorganic fertilizer to the maize plants in the intercrops, thus leading to the input of crop residues with higher N concentrations (Vachon and Oelbermann, 2010). In Costa Rica, Chang and Shibles (1985) also observed no significant difference in N-input from crop residues involving maize and cowpea (*Virginia unguiclata* L. Walp.) sole crops compared to a maize-cowpea intercrop. The type of crop residue plays an important role in C sequestration and soil aggregation (Balabane and

Plante, 2004) due to the C/N ratio and thus the complexity of the residue (Wright and Hons, 2004).

In this study, the soil C/N ratio was not significantly different between treatments; but was slightly higher in the surface layer of the intercrop treatments and, there was a slight increase in the 10-20 cm in the sole crops treatments. In a study by Oelbermann and Echarte (2010), conducted on the same field site at INTA, Balcarce in 2007-2008, the sole crops had a slightly higher C/N ratio than compared to the intercrops.

### **3.3.2 Soil Infiltration Rate**

Water infiltration is an indicator of the soil's ability to allow water movement into the soil. A soil with increased infiltration results in storage of water making it available for root uptake, plant growth and a habitat for soil organisms. Intercrops have the ability to improve soil moisture immediately below the soil surface as the soil under a sole crop is generally more exposed to evaporative loss than an intercrop (Olasantan, 1988; Fukai and Trenbath, 1993). They also have the ability to increase SOM, which controls infiltration through the development of stable soil aggregates. Moreover, the root network underneath an intercropping system will be much more extensive than under a sole crop system thus leading to increased infiltration. In this study, both the 1:2 intercrop and the 2:3 intercrop had higher infiltration rates than the sole crops. Similarly, in an alley-cropping study with *Gliricidia* (*Gliricidia sepium* L.) and Pigeon Pea (*Cajanus cajan* L.) intercropped with maize, pumpkin (*Cucurbita Maxima* L.) and okra (*Abelmoschus esculentus* L.) in Sir Lanka, lower bulk density, higher porosity and greater water

infiltration rates were found when compared to the sole *Gliricidia* and sole Pigeon Pea systems (Mapa and Gunasena, 1995). A hillside agricultural study conducted by the International Development Research Centre (IDRC) in northern Honduras also found that second season maize cropped with velvet bean (*Mucuna pruriens* L.) had increased crop productivity, increased infiltration and water-holding capacity. Moreover, the increase in organic matter resulted in a greater resistance to drought in both crops (Buckles et al., 1998). In Uganda, a one-season fallow of an indigenous legume (*Crotalaria ochroleuca*) intercropped with maize resulted in increased water infiltration, increased yield and decreased bulk density (Fischler et al., 1999).

### **3.4 Conclusion**

The objective of this study was to quantify differences in soil chemical and physical characteristics in sole vs. intercropping systems and to determine temporal changes in soil chemical and physical characteristics over two growing seasons. Although, there has yet to be a significant difference in these characteristics between the sole crops and intercrops over the last two growing seasons; equilibrium in soil properties may be reached after a period of five or more years in the rolling Pampas. Discernible trends between 2007-2008 and 2008-2010 have shown that there are continuing changes in the soil properties as the soil begins to reach equilibrium after intercrop establishment. Therefore, it is expected that in the next ten years quantifiable differences in soil chemical properties may be observed.

Continuous monitoring of soil chemical and physical characteristics in sustainable complex agroecosystems continues to be central in aiding researchers and scientists in protocol and policy development. Moreover, an improved understanding of these soil characteristics is fundamental to creating GHG management plans that enable farmers to sequester C and have more sustainable farming practices. Thus, even though this study is in its early stages, it is still important to quantify changes in soil quality after intercrop establishment in order to appreciate the complexity of the soil system. Annual monitoring of this maize-legume experiment is therefore recommended to examine changes in soil physical and chemical characteristics as the agroecosystem begins to stabilize.

## Chapter 4

### Soil Carbon Dioxide and Nitrous Oxide Production Rates during the Growing Season in a Complex Agroecosystem in the Argentine Pampa

#### 4.0 Introduction

Although in some countries the rate of agricultural expansion has slowed in most recent years, crop yields have increased dramatically (Naylor, 1996). This achievement is based largely on intensified management practices, genetic modification of crops to high-yielding, disease resistant varieties; fertilizers, pesticides and mechanization (Matson et al., 1997). These intensive agricultural practices have reduced levels of soil organic carbon (SOC) and have contributed to the production rates of greenhouse gases (GHGs). The Intergovernmental Panel on Climate Change (IPCC) has concluded with 95% certainty that the main drivers of climate change have been anthropogenic increases in GHG production rates (IPCC, 2007c). For instance, agriculture contributes 10-12% of the total estimated GHG production rates annually (5.1 to 6.1 Gt carbon dioxide (CO<sub>2</sub>) equivalents/yr) (Niggli et al., 2009), and as the demand for increased yields and thus increased fertilizer-use continues, nitrous oxide (N<sub>2</sub>O) production rates are expected to increase by 35-60% by the year 2030 (Smith et al., 2007).

Several crop-simulation models and future climate scenarios have predicted that grain yield could be reduced by 30% by 2080 under warmer scenarios in central and south America (IPCC, 2007b). According to the IPCC, this could place an additional 5, 26 and 85 million people at risk of hunger globally by 2020, 2050 and 2080 respectively. Using a Global Climate

Model (GCM), Magrin and Travasso (2002) predicted that a 3°C increase in temperature in the Argentine Pampa would decrease wheat yields by 4%, maize yields by 9% but could increase soybean yields by 29%. However, a 5.6 °C increase as used by the United Kingdom Meteorological Office (UKMO) could result in a decrease of 16%, 17% and increase of 14% for wheat, maize and soybean respectively (Magrin and Travasso, 2002).

With a prediction of decreased yields, in the near future crop producers will continue to intensify cropping systems to maintain crop yields thus increasing the amount of GHGs emitted. However, several intensive agricultural land management practices have been proposed for mitigating GHG production rates (Olesen and Bindi, 2002; Smith et al., 2007). These practices involve improving crop, soil and fertilizer management practices such as conservation tillage including no-till farming, agroforestry, intercropping, integrated crop and livestock farming (Niggli et al., 2009) and implementation of sustainable agricultural productions systems (Studdert and Echeverria, 2000; Verchot et al., 2008). According to Wang et al. (2010), the most simplistic way of sequestering C in agricultural soils is to produce and retain a sizeable quantity of crop biomass or organic C in the soil. Biomass accumulation can be enhanced by growing cover crops between main crop growing seasons, reducing fallow period, implementing crop rotations and intercropping systems (Wang et al., 2010). For example intercropping, where more than one crop is grown on the same land unit at the same time may be a more sustainable agricultural production system compared to conventional or sole crop systems. Maize-legume intercrops for instance provide reciprocal benefits and optimal crop and soil productivity, they

allow crop residue biomass to be returned to the soil and thus aid in sequestering C. (Wang et al., 2010).

The Argentine Pampa is a global producer of maize (*Zea mays* L.) and soybean [*Glycine max* (L.) Merr.], as such, there has been severe soil degradation and loss of SOM. However, landowners are adopting maize-legume intercrops to try to minimize these effects; intercropping with legumes can reduce losses of C and N from cultivated land because of an overall increase in system complexity (Prasad and Brook, 2005). The integration of litter from legume crop residues, which have low C/N ratios, have been shown to increase C and N retention in temperate agroecosystems (Drinkwater et al., 1998) thus reducing CO<sub>2</sub> and N<sub>2</sub>O production rates. The addition of leguminous plants with maize regulates the internal N cycle via dinitrogen-fixation (N<sub>2</sub>-fixation) (Schipanski et al., 2010) associated with its symbiosis with the bacteria *Rhizobium*, and decreases the need for fertilizer application; further mitigating impacts (Inal et al., 2007).

Although agricultural soils have a substantial impact on the global GHG inventory, there are still large uncertainties associated with estimates of regional and global GHG fluxes; this is largely due to insufficient temporal and spatial measurements (Lal, 2004b). What's more, as annual GHG fluxes can have a large variation between sites (Stehfest and Bouwman, 2006) and on the same site in different years (Dobbie et al., 1999), extrapolation of findings from one site or one period to another may cause serious under or over-estimation of production rates. Most studies of soil GHG production rates are from agricultural systems with various tillage or fertilizer applications and there are no known studies on the effect of maize-legume intercrops of

GHG production rates, except those in agroforestry (Verchot et al., 2008). Moreover, there is little understanding of the underlying processes involved in the sequestration and stabilization of SOC in intercropping systems (Oelbermann et al., 2004). Therefore, a field experiment was carried out to quantify GHG production rates from maize-soybean intercrops compared to maize and soybean sole crops. Such information is crucial as it aids in the understanding of how agroecosystem management practices can affect C sequestration and the production of GHGs (Rochette et al., 1999) . Moreover, this study will contribute to already existing global GHG databases (Smith et al., 2007; Verchot et al., 2008) and will help to improve our understanding of optimal agroecosystem design for the long-term sequestration of C and resulting reduction in soil GHG production rates.

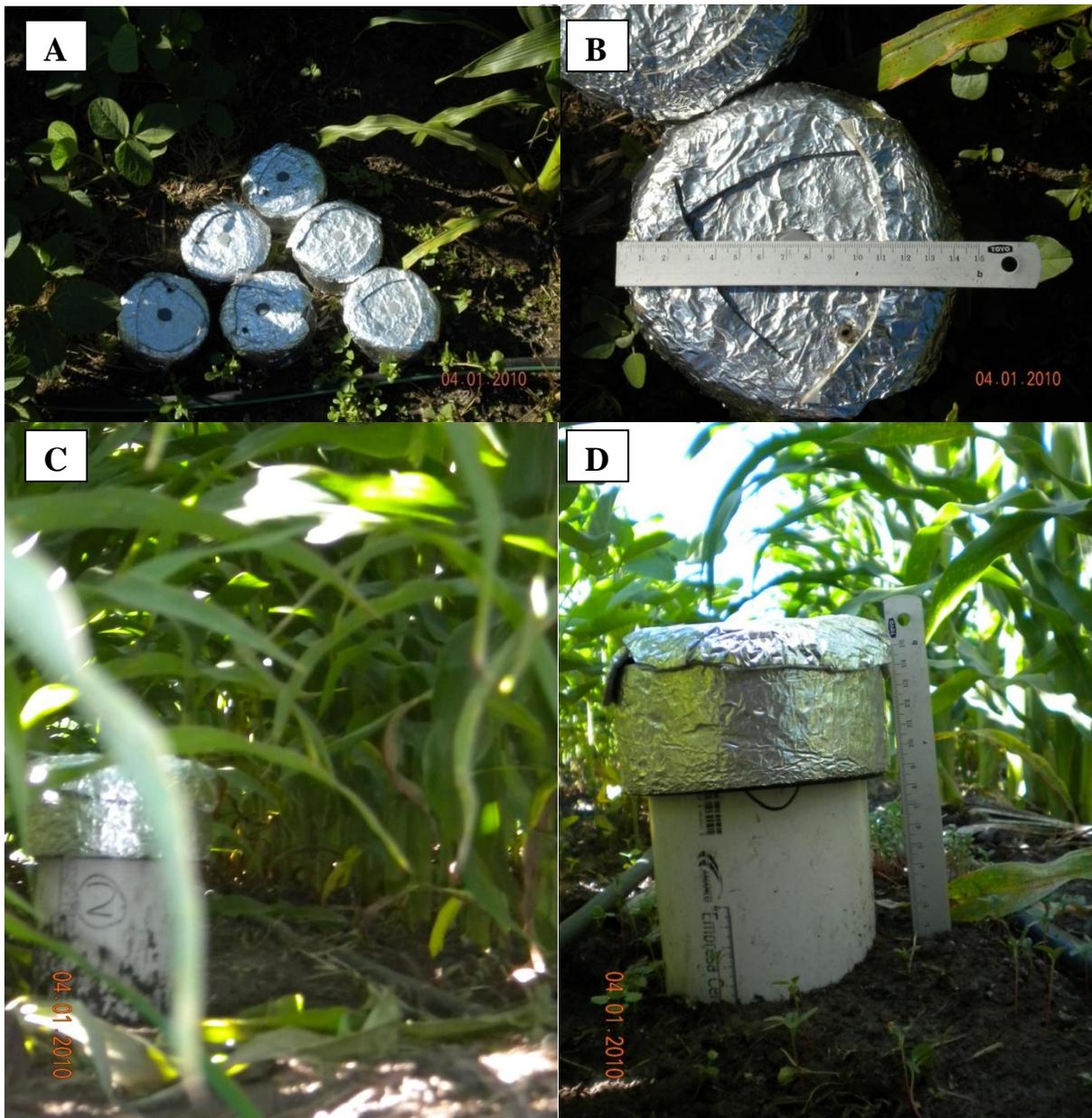
The objectives of this study were therefore:

- 1) To quantify soil GHG production rates from maize-soybean intercrops compared to maize and soybean sole crops over two field seasons.
- 2) To estimate annual changes in GHG production rates rate from maize-soybean intercrops compared to maize and soybean sole crops between Y1 (2008-2009) and Y2 (2009-2010).
- 3) To examine the correlation between soil GHG production rates from a maize-soybean intercropping systems at various ratios and a soybean sole crop system and a maize sole crop system and soil temperature and moisture over two growing seasons.

## **4.1 Materials and Methods**

### **4.1.1 Greenhouse Gas Chamber Design**

Greenhouse gas sampling chambers were constructed from PVC piping and based on a design by the Trace Gas Protocol Development Committee-United States Department of Agriculture (Parkin et al., 2004). Chamber bases were constructed using PVC irrigation pipe (25 cm height and 15 cm inner diameter) and inserted into the soil to a depth of 10 cm (Figure 4.1). Chamber collars were constructed using PVC caps (15 cm diameter) and were insulated [6 mm polyolefin foam (Borealis, Port Murray, USA)] to seal against the top edge of the chamber base. The collars were also fitted with a septa (2 cm diameter) used as a sampling port for extraction of the gases and covered with reflective insulation. The chambers were vented using a 10 cm length of Bev-a-line IV tubing [inside diameter 6 mm (Fisher Scientific, Mississauga, Canada)]. The chambers were left in the ground from December until February and were left open to the atmosphere between sample periods. Both the soybean and maize plants grew past the chamber edges and roots inevitably extended underneath the chambers but any seedlings that grew inside the chamber between sample periods were carefully removed the day before sampling took place. According to Parkin et al. (2003), it is advisable to measure bare soil fluxes using shorter chambers because they have a higher sensitivity.



**Figure 4.1 - Greenhouse gas (GHG) sampling chambers constructed from PVC piping based on a design by Parkin et al. (2004) ; A) Insulated caps used for chamber measurement; B) Cap (7.5 cm radius) with sampling port; C) Chamber and cap under measurement in the maize sole crop; D) Chamber and cap under measurement in the 2:3 intercrop.**

### 4.1.2 Field Sampling

Two sample chambers per replicate plot ( $n=6$ ) were placed in the soil between crop rows at a distance of 2 m from the plot edge to minimize border effects. In the intercrop treatments, chambers were always placed between maize and soybean rows. GHGs were measured over two, 3 month periods (December-February), bi-weekly from 2008-2010, always between 9-am and noon to minimize partiality associated with diurnal variations (Parkin et al., 2004). The collars were placed on top of the chambers and remained in place for 30 mins; during this time the GHG's were measured at  $t=0$ ,  $t=15$  and  $t=30$  mins using 10 mL air-tight syringes (Luer-Lok Tip, BD, Franklin Lakes, NJ, USA). Gases were then transferred into 3 mL evacuated vials (Labco Ltd., High Wycombe, UK). According to Parkin (2003) and Venterea (2009) decreasing the chamber measurement time tends to reduce non-linearity. Note that the syringe was not pumped in the headspace before sampling as it may affect pressure perturbations and/or excess dilution of the headspace gas by entry of outside air through the vent tube (Parkin et al., 2004).

At the same time as sampling GHG production rates, soil moisture content (%) and soil temperature ( $^{\circ}\text{C}$ ) to a 10 cm depth were recorded using a WET-2 sensor (Delta-T Devices, Cambridge, UK). Measurements were sampled at the same time and in the same location as GHGs. Ambient temperature and precipitation data (30 year mean) was obtained from a weather station, operated by the University of Mar del Plata, located adjacent to the study site.

### 4.1.3 Quantification of Greenhouse Gas Production rates

Concentrations of CO<sub>2</sub> and N<sub>2</sub>O were determined by gas chromatography (GC) (Agilent 6890N, California, USA). Samples from individual chambers were analyzed in sequence; t=0, t=15 and t=30 mins to account from problems with GC drift. A gas standard consisting of 99.9 ppm CO<sub>2</sub> and 10.0 ppm N<sub>2</sub>O was injected at a 10 sample interval. While the GC detected CO<sub>2</sub> and N<sub>2</sub>O production rates, CH<sub>4</sub> production rates were not detected in the samples. Production rates of CO<sub>2</sub> and N<sub>2</sub>O for each treatment were calculated using the mean values of the six chambers (*n*=6). They were quantified according to the following equations (Hutchinson and Mosier, 1981):

$$[(C_1 - C_0) / (C_2 - C_1)] \quad (4.1)$$

where C<sub>0</sub>, C<sub>1</sub> and C<sub>2</sub> are the chamber headspace gas concentrations (ppmv) at t=0, t=15 and t=30, respectively. If the Hutchinson and Mosier equation was  $\geq 1$ , linear regression was used, however if the equation  $\leq 1$  than the second equation proposed by Hutchinson and Mosier (1981) was used:

$$f_0 = V(C_1 - C_0)^2 / [A * t_1 * (2 * C_1 - C_2 - C_0)] * \ln [(C_1 - C_0) / (C_2 - C_1)] \quad (4.2)$$

where  $f_0$  is the quantity gas produced ( $\mu\text{l m}^{-2} \text{min}^{-1}$ ); V is the chamber headspace volume (l); A is the soil surface area ( $\text{m}^2$ ) and  $t_1$  is the time interval between gas sampling points (minutes). The  $r^2$  value from the linear regression analysis was substituted for  $f_0$  in the Ideal Gas Law equation:

$$PV=nRT \quad (4.3)$$

where P is pressure (atmosphere); n is the number of moles of gas (mol); R is the gas law constant ( $1 \text{ atm mol}^{-1} \text{ K}^{-1}$ ) and T is temperature ( $^{\circ}\text{K}$ ). Temperature and precipitation data was obtained from the INTA meteorological station, located adjacent to the study site. Resulting values ( $\mu\text{mol m}^{-2} \text{ min}^{-1}$ ) of  $\text{CO}_2$  and  $\text{N}_2\text{O}$  were converted to  $\mu\text{g m}^{-2} \text{ h}^{-1}$  by multiplying by the molecular weight of the analyte. If the value was  $\leq 1$ , then the measurements could be accredited to a build-up of the analyte concentration in the headspace and thus Equation 5.2 proposed by Hutchinson and Mosier (1981) was utilized.

#### **4.1.4 Statistical Analysis**

All data was examined for homogeneity of variance and normality and were found to have normal distributions. Differences between treatments for soil characteristics and GHG production rates were analyzed using the univariate general linear model (ANOVA) in SPSS (SPSS Science Inc., v. 15.0, 2006). Significant differences were further analyzed using Tukey's multiple comparison test (Steel et al., 1997). Since the distribution was normal, a Pearson product moment correlation (r) was used to determine the relationship between treatments and soil  $\text{CO}_2$  and  $\text{N}_2\text{O}$  production rates, soil temperature ( $^{\circ}\text{C}$ ) and soil moisture (% vol). A paired t-test was used to compare differences in GHG production rates between sampling years (Y1 and Y2) within treatments. For each treatment, linear regression analysis was used to determine the relationship between  $\text{CO}_2$  or  $\text{N}_2\text{O}$  production rates and the product of soil temperature and volumetric soil moisture (0-10 cm). For all statistical analyses, the threshold probability level was  $p < 0.05$ .

## 4.2 Results

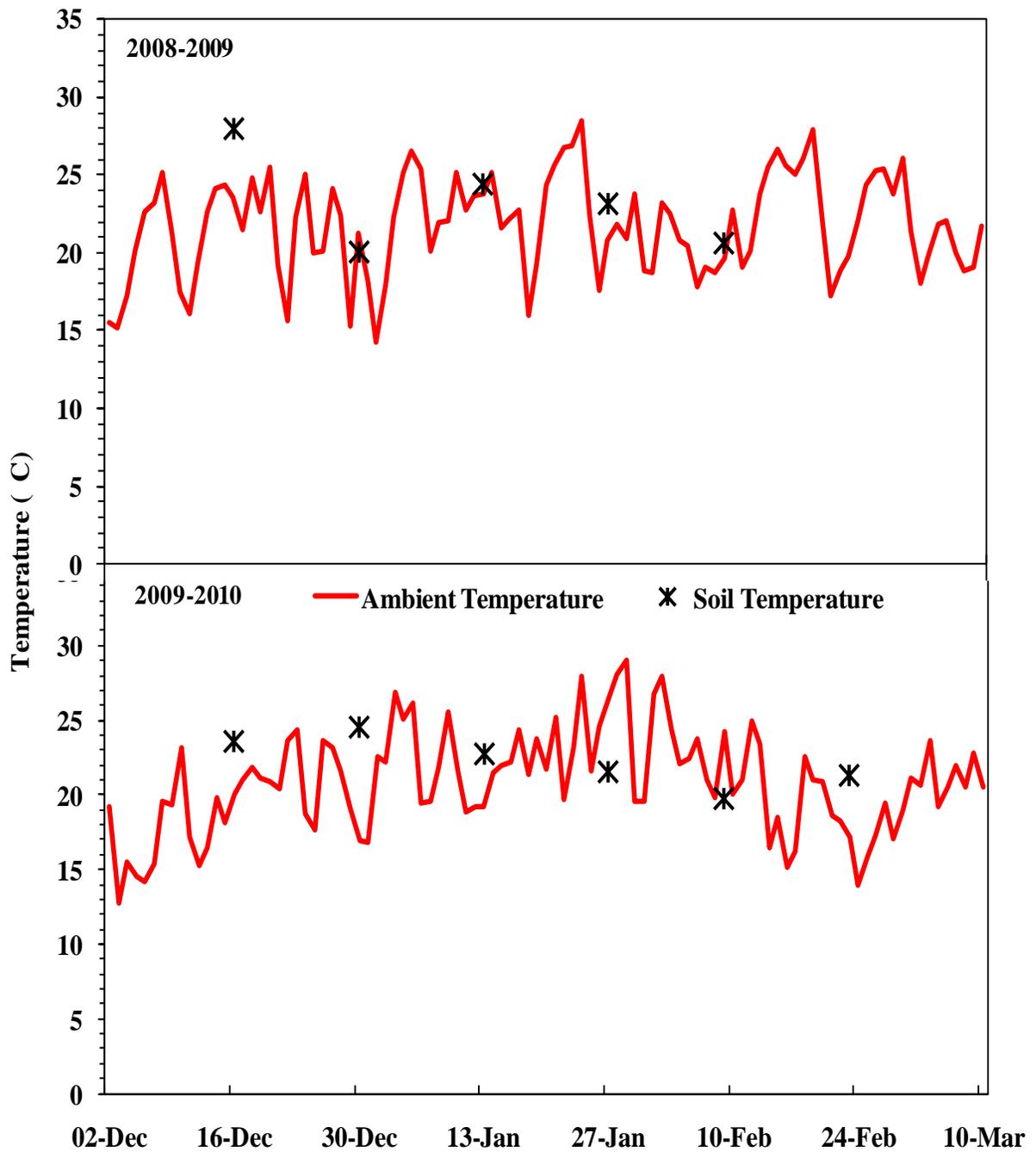
Soil temperature and moisture varied within and between each field season. Both soil moisture and temperature followed a similar pattern to that of the ambient temperature and precipitation. Soil temperature was highest during the latter part of December in both years, and was 1°C higher in Y1 than Y2 (Figure 4.2). Soil moisture was highest in December of Y1 and in February of Y2 (Figure 4.3).

Significant differences in production rates rates of CO<sub>2</sub> and N<sub>2</sub>O ( $\mu\text{g analyte m}^{-2} \text{h}^{-1}$ ) were not found between Y1 and Y2 for all treatments (Figure 4.4 & 4.5), but CO<sub>2</sub> differed significantly between treatments in Y2 ( $p < 0.05$ ). When comparing differences in CO<sub>2</sub> production rates rates between treatments, lower production rates were observed in the intercrop treatments during both years. For example, mean CO<sub>2</sub> production rates rates ranged from 323 to 379  $\mu\text{g C m}^{-2} \text{h}^{-1}$  in the soybean and maize sole crops respectively. Whereas the intercrops ranged from 268 in Y1 to 299  $\mu\text{g C m}^{-2} \text{h}^{-1}$  in Y2 in the 1:2 intercrop and in the 2:3 intercrop ranged from 280 in Y1 to 300  $\mu\text{g C m}^{-2} \text{h}^{-1}$  in Y2. In Y1, all treatments saw a spike in CO<sub>2</sub> production rates rates during the last sample day (February 10, 2008) with 458  $\mu\text{g C m}^{-2} \text{h}^{-1}$  (maize sole crop), 425  $\mu\text{g C m}^{-2} \text{h}^{-1}$  (soybean sole crop), 327  $\mu\text{g C m}^{-2} \text{h}^{-1}$  (1:2 intercrop) and 429  $\mu\text{g C m}^{-2} \text{h}^{-1}$  (2:3 intercrop). Conversely, in Y2 the maize sole crop and soybean sole crop peaked in mid-January (430 and 357  $\mu\text{g C m}^{-2} \text{h}^{-1}$  respectively). The intercrops however had the lowest CO<sub>2</sub> production rates rates in December of both years [233 (Y1) and 256  $\mu\text{g C m}^{-2} \text{h}^{-1}$  (Y2)] and [221 (Y1) and 239  $\mu\text{g C m}^{-2} \text{h}^{-1}$  (Y2)] for the 1:2 and 2:3 intercrops respectively.

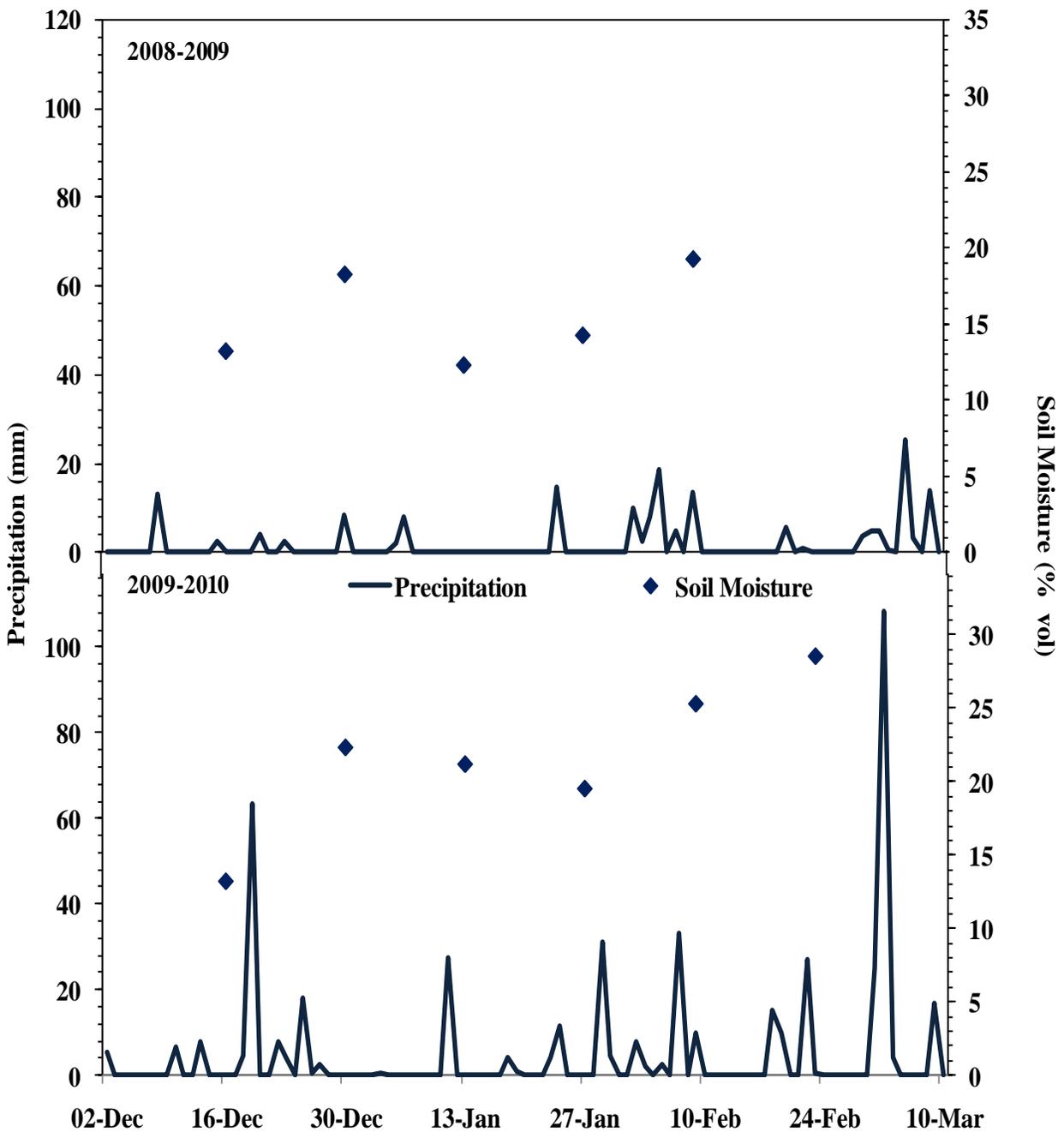
In Y1, 51% of the variation in CO<sub>2</sub> production rates in the soybean sole crop was explained by soil moisture and temperature, alternatively they explained only 20% in the maize sole crop and 1:2 intercrop, and 29% in the 2:3 intercrop. In Y2, soil moisture and temperature explained 10% (soybean), 2% (maize), 6% (1:2 intercrop) and 32% (2:3 intercrop) of the variation in CO<sub>2</sub> production rates (Table 4.1). There was a significant positive relationship between soil CO<sub>2</sub> production rates and soil moisture in the 1:2 intercrop and maize sole crop in Y1, whereas a significant correlation between CO<sub>2</sub> production rates and soil temperature occurred in the soybean sole crop and 2:3 intercrop in Y1 (Table 4.1). In Y2, only the 2:3 intercrop showed a significant correlation between CO<sub>2</sub> production rates and soil moisture.

When comparing differences in N<sub>2</sub>O production rates between treatments (mean of both field seasons) lower production rates were observed in the intercrop systems. For example, N<sub>2</sub>O production rates ranged from 13.5 to 14.0 µg N m<sup>-2</sup> h<sup>-1</sup> in the soybean and maize sole crops respectively, whereas in the intercrops they ranged from 11.5 (1:2 intercrop) to 12.0 µg N m<sup>-2</sup> h<sup>-1</sup> (2:3 intercrop). Similar to the CO<sub>2</sub> production rates, the N<sub>2</sub>O production rates varied over the growing season and different treatments peaked at various times. In Y1 the maize sole crop and soybean sole crop peaked in mid-February (16.94 and 15.57 µg N m<sup>-2</sup> h<sup>-1</sup> respectively) and in Y2 they peaked mid- January (16.09 and 14.77 µg N m<sup>-2</sup> h<sup>-1</sup> respectively). In contrast, the lowest N<sub>2</sub>O production rates occurred in mid-December and late January of both years and was in the 2:3 intercrop [10 µg N m<sup>-2</sup> h<sup>-1</sup> (Y1)] and was the lowest in the 1:2 intercrop (9 µg N m<sup>-2</sup> h<sup>-1</sup>) in Y2.

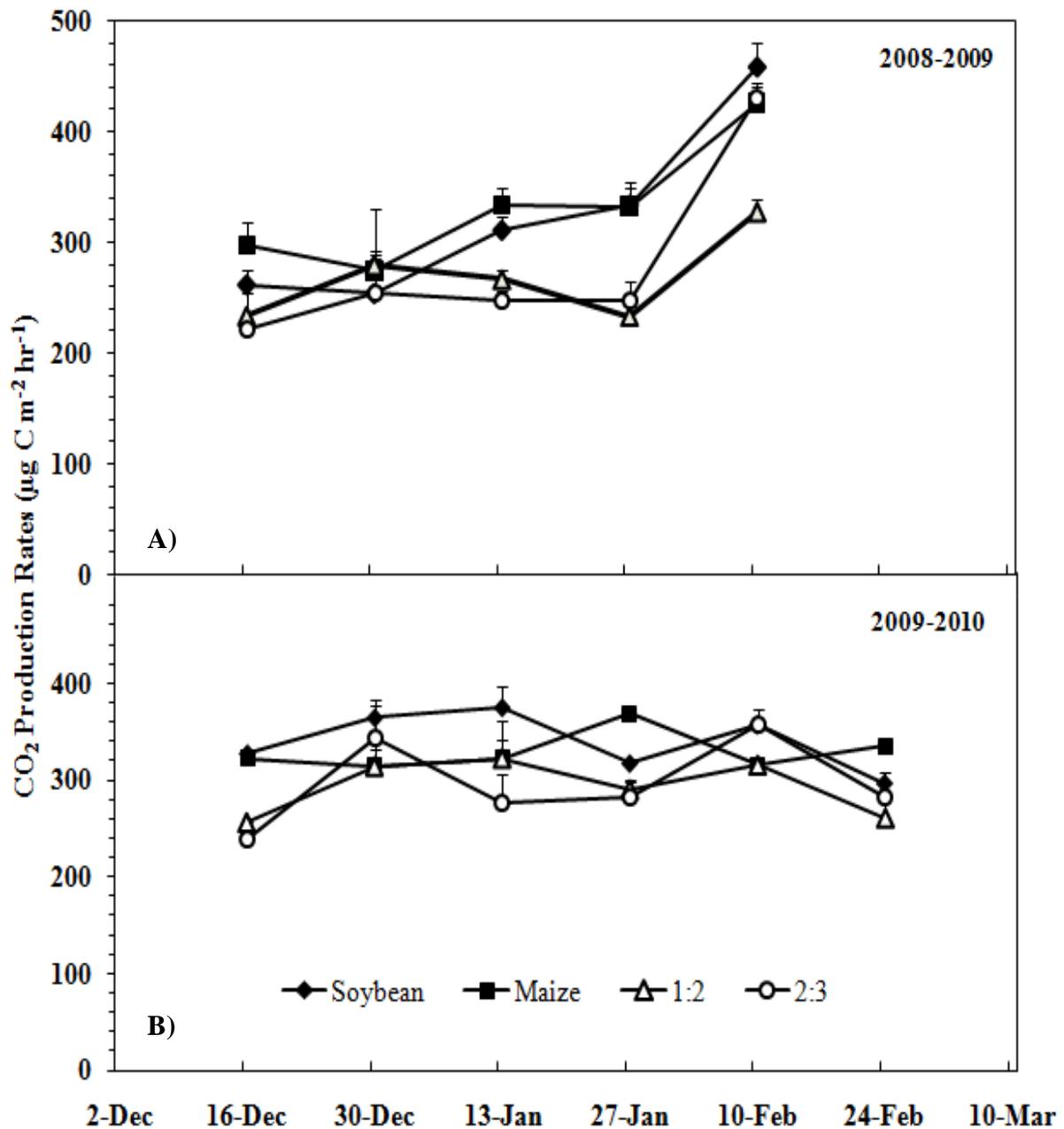
The observed variation in soil N<sub>2</sub>O production rates significantly correlated to changes in soil moisture (% vol) and to a lesser extent soil temperature (°C). For example, there was a significant positive correlation between N<sub>2</sub>O production rates and soil temperature in the soybean sole crop in Y1, but a significant negative correlation for the maize sole crop and 1:2 intercrop in Y2 (Table 4.1). In Y1, 17% of the variation in N<sub>2</sub>O production rates in the soybean sole crop and the 2:3 intercrop was explained by soil moisture and temperature. Whereas 46% and 28% of the variation in N<sub>2</sub>O production rates in the maize sole crop and 1:2 intercrop was explained by soil moisture and temperature. In Y2, however 40% of the variation in N<sub>2</sub>O production rates in the soybean sole crop, 60% in the maize sole crop and 1:2 intercrop, and 34% in the 2:3 intercrop was explained by soil moisture and temperature.



**Figure 4.2 - Mean air and soil (0-10 cm) temperature (°C) of soybean and maize sole crop and intercrops (n=6) over two field seasons A) Y1: December 2008 – February 2009’ B) Y2: December 2009 – February 2010 in the Argentine Pampa.**



**Figure 4.3 - Precipitation (mm) and soil moisture (0-10 cm) (% vol) of soybean and maize sole crop and intercrops (n=6) over two field seasons A) Y1: December 2008 – February 2009; B) Y2: December 2009 – February 2010 in the Argentine Pampa.**



**Figure 4.4 - Soil CO<sub>2</sub> production rates (µg C m<sup>-2</sup> h<sup>-1</sup>) from soybean and maize sole crop and intercrops (n=6) over two field seasons A) Y1: December 2008 – February 2009; B) Y2: December 2009 – February 2010 in the Argentine Pampa. Vertical bars represent standard errors.**

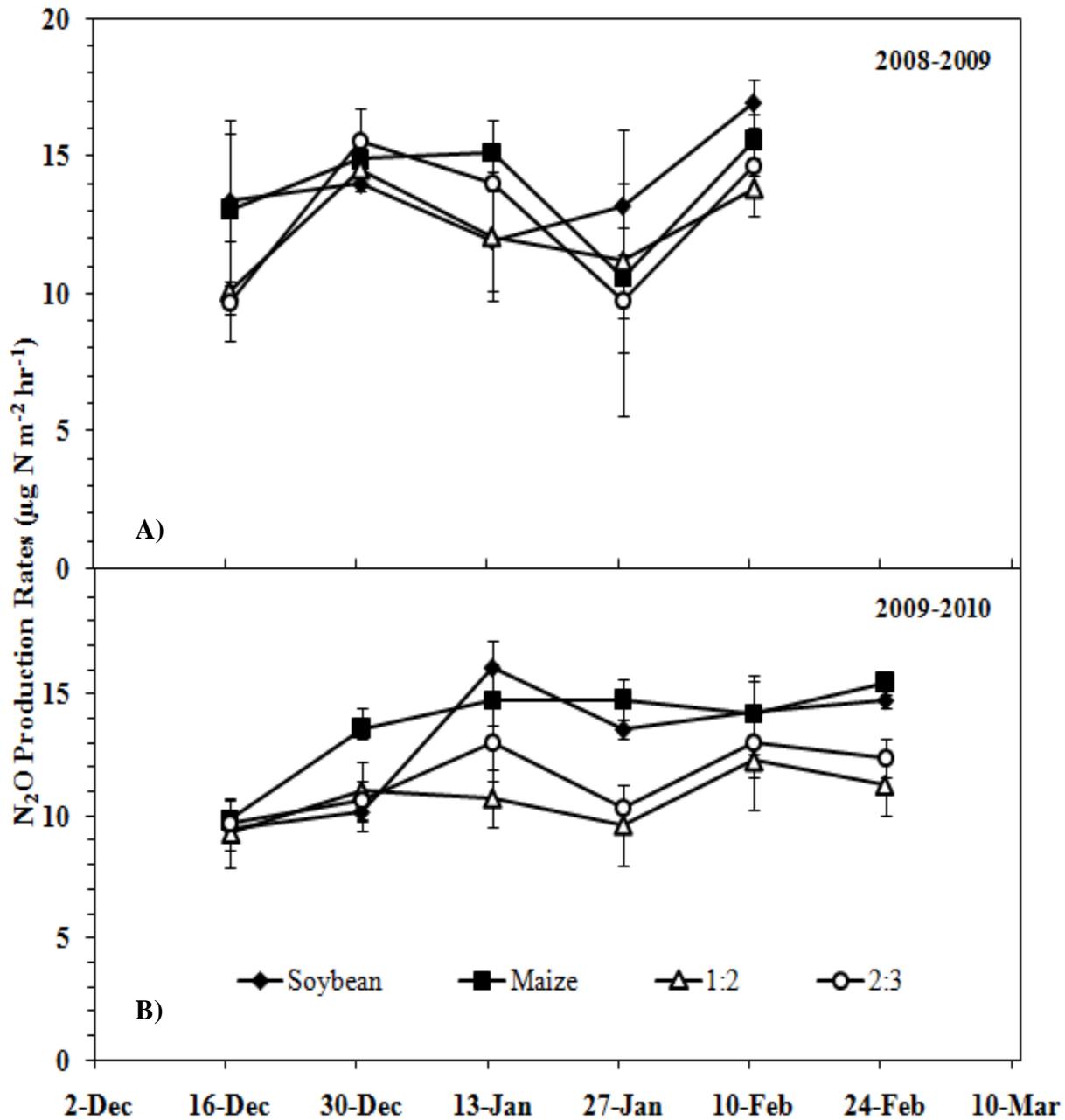


Figure 4.5 - N<sub>2</sub>O production rates (µg N m<sup>-2</sup> h<sup>-1</sup>) from soybean and maize sole crop and intercrops (n=6) over two field seasons A) Y1: December 2008 – February 2009; B) Y2: December 2009 – February 2010 in the Argentine Pampa. Vertical bars represent standard errors.

**Table 4.1 - Pearson product moment correlations (r) between treatment soil CO<sub>2</sub> or N<sub>2</sub>O production rates and soil temperature (°C), or between soil CO<sub>2</sub> or N<sub>2</sub>O production rates and soil moisture (% vol.) for each treatment in Y1 and Y2, in the Argentine Pampa.**

	Soil Temperature vs. CO <sub>2</sub>	Soil Moisture vs. CO <sub>2</sub>	Soil Temperature vs. N <sub>2</sub> O	Soil Moisture vs. N <sub>2</sub> O
<b>Y1 December 2008 – February 2009</b>				
<b>Soybean Sole Crop</b>	-.001**	.901	.026*	-.141
<b>Maize Sole Crop</b>	.114	-.023*	.064	.013*
<b>1:2 Intercrop</b>	-.621	.039*	-.005**	-.008**
<b>2:3 Intercrop</b>	-.020*	.966	-.195	.021*
<b>Y2 December 2009 – February 2010</b>				
<b>Soybean Sole Crop</b>	-.323	.147	-.131	.001**
<b>Maize Sole Crop</b>	.433	-.575	-.008**	.001**
<b>1:2 Intercrop</b>	.647	.208	-.095	.001**
<b>2:3 Intercrop</b>	.725	.004**	-.140	.001**

\* Correlation is significant at p=0.05.

\*\* Correlation is significant at p=0.01.

Note: Data had a normal distribution and variance.

### 4.3 Discussion

During this study both the dry (Y1) and the wet (Y2) growing season showed variable measurements in ambient air temperature and precipitation. Soil moisture was lower in Y1 due to a lower amount of precipitation, which was 171 mm below the 30 year mean from December to February. Comparatively, during Y2, total precipitation was 312 mm (Y1 = 199 mm); 20 mm above the 30 year mean. The variation between Y1 and Y2 can be attributed to natural seasonal variations in local climate patterns that expectedly influenced diurnal CO<sub>2</sub> and N<sub>2</sub>O production rates (Smith et al., 2007). Although, temperature and moisture are two of the most influential environmental factors affecting the rate of nutrient cycling and the production of GHGs (Kirschbaum, 1995; Smith, 1997), there were only a few direct correlations between these variables and CO<sub>2</sub> and N<sub>2</sub>O production rates. This was not expected as precipitation typically affects the production rates of CO<sub>2</sub> and N<sub>2</sub>O from agricultural soils in two main ways: soil water content affects soil aeration, which in turn affects microbial processes of production and consumption of trace gases (Davidson et al., 2004). Decreased precipitation can also amend root turnover, litterfall, decomposition and mineralization, which in turn will influence the variability of C and N substrates from trace gas production (Davidson et al., 2004). However, researchers have found that in areas where water is non-limiting such as in the temperate region, decomposition, mineralization and CO<sub>2</sub> production rates are highly correlated to temperature (Kirschbaum, 1995; Olaniran, 1988). In Y1 however, production rates were highly correlated to precipitation probably because water was limiting.

Soil CO<sub>2</sub> and N<sub>2</sub>O production rates for all treatments were similar to the values reported from other temperate agroecosystems (Ellert and Janzen, 2008; Omonode et al., 2007; Rastogi et al., 2002). However, the 1:2 and 2:3 intercrops had lower CO<sub>2</sub> and N<sub>2</sub>O production rates than those reported by Sey and Whalen (2008) from maize and soybean sole crop systems at McGill University in Quebec, Canada. In this study, soil CO<sub>2</sub> fluxes were highest in the maize sole crop. Comparatively, in a long-term study evaluating the relationship between CO<sub>2</sub> production rates and residue input, the maize sole crops had the greatest C input from crop residues in comparison to maize-soybean rotations, yet the CO<sub>2</sub> production rates from the rotation were much lower. In a similar study, conducted on the same field site at Instituto Nacional de Tecnología Agropecuaria (INTA) in 2007-2008; Oelbermann and Echarte (2010), found that C input from crop residues was greatest in the maize sole crop (904 g C m<sup>-2</sup>); followed by the intercrops [768 g C m<sup>-2</sup> (2:3 intercrop) and 552 g C m<sup>-2</sup> (1:2 intercrop)] and lowest in the soybean sole crop (502 g C m<sup>-2</sup>). Thus the results from this study confirm that the crop with the greatest litterfall and residue addition (maize sole crop), had the greatest CO<sub>2</sub> production rates (West and Post, 2002).

However, it has also been suggested that a decrease in residue input and therefore C may result in a lower rate of C sequestration rate (Lal and Bruce, 1999; Rasmussen et al., 1980; West and Post, 2002). In a 2 year study on maize-soybean rotations in Indiana, Omonode et al. (2007) reported that mean CO<sub>2</sub> production rates from maize sole crops were 16% higher than maize in rotation with soybean. Jacinthe et al. (2002) reported that seasonal CO<sub>2</sub> fluxes are affected by the amount, type and timing of crop residue incorporation. In northern France for instance, Oorts et al. (2007) determined that crop residues contributed significantly to CO<sub>2</sub> production rates in a

no-till maize-wheat rotation. This was attributed to differences in the C/N ratio between the crop residues in the no-till treatment, which had a lower C/N ratio compared to the conventional till treatment, which had a higher C/N and thus contributed significantly to production rates.

Soil CO<sub>2</sub> fluxes differed significantly between the sole crops and intercrops in both years of study. In Y1, CO<sub>2</sub> increased throughout the growing season however in Y2, CO<sub>2</sub> fluxes stayed relatively steady. Moreover, throughout the growing season, treatments showed peak CO<sub>2</sub> production rates rates which corresponded to the active growth (linear phase of biomass accumulation) of the crops (Fearnside, 2000). Adviento-Borbe et al. (2007) found CO<sub>2</sub> production rates were low immediately after planting, then increased with progressing growth of maize, and reached a maximum of 500  $\mu\text{g C m}^{-2} \text{h}^{-1}$  at the peak of the growing season. This was similar to results observed in this study, where soil CO<sub>2</sub> fluxes reached a maximum of 458  $\mu\text{g C m}^{-2} \text{h}^{-1}$  during mid-growing season, and was comparable to results reported by Verma et al. (2005) and Ellert and Janzen (2008).

Furthermore, variability in production rates rates within and between growing seasons were likely due to soil temperature and moisture which led to increased heterotrophic activity (Sehy et al., 2003). Although CO<sub>2</sub> production rates were not significantly correlated to temperature in any of the treatments this is could be due to limitation by water and C availability in the soil. Conversely, other researchers have found that CO<sub>2</sub> production rates reflect seasonal temperature, moisture and root respiration during the active growth phase (Ellert and Janzen, 2008; Omonode et al., 2007; Oorts et al., 2007). Additionally, quantity and quality of crop residue inputs, tillage, and differences in weather patterns may also control CO<sub>2</sub> production rates

rates (Franzluebbers and Arshad, 1996). For example, Franzluebbers (2005) found that soil CO<sub>2</sub> production was greatest during peak crop biomass productivity due to root respiration, and at harvest due to a greater input of crop residues. In the maize sole crop, root respiration contributed to CO<sub>2</sub> production during the growing season, and residue decomposition contributed to increases in production rates following harvest (Franzluebbers and Arshad, 1996).

The greatest N<sub>2</sub>O production rates occurred in the maize sole crop, Verchot et al. (2008) has suggested that biological N<sub>2</sub>-fixation may likely not have contributed substantially to N<sub>2</sub>O production. Biological N<sub>2</sub>-fixation reduces the production of N<sub>2</sub>O compared to the release of N from root exudates and the mineralization of precedent crop residues. Gregorich et al. (2005) and Verchot et al. (2008) found similar results in a legume-based agroforestry system over a 3 year study in eastern Canada. On the contrary, previous studies have shown that a legume, either as a sole crop, an intercrop, or in agroforestry systems may stimulate higher levels of N<sub>2</sub>O production via denitrification, nitrification, or biological N<sub>2</sub>-fixation (Sey et al., 2008).

In general, the majority of N<sub>2</sub>O production in temperate grassland ecosystems occurs within an optimal range of soil temperature, with increased soil moisture, easily metabolized C, and fertilizer application (Müller and Sherlock, 2004). Temperature has been established as an important factor for microbial activities including nitrification and denitrification (Dalal et al., 2003). Rainfall events can also stimulate brief peaks in N<sub>2</sub>O production rates; showing that soil moisture is an important factor, but cannot be used as the sole predictor in production rates (Müller and Sherlock, 2004). Several studies have found one of the key factors influencing N<sub>2</sub>O production rates is soil moisture which is dependent on precipitation, evapotranspiration and soil

properties (Drury et al., 2005). In Y1 soil moisture and temperature accounted for  $\approx 32\%$  of the variability in  $\text{N}_2\text{O}$  fluxes and 49% in Y2. The variation in significance between soil moisture and GHG fluxes during both years of study is most likely because of the timing of rainfall events in relation to the time of the day that the GHG measurements were taken. For example, an unusually large flux of  $\text{CO}_2$  was observed on February 9<sup>th</sup>, 2009 when there was a moderate rain event in the early hours before sampling.

In this study, the sole crops generally had higher soil  $\text{N}_2\text{O}$  production rates when compared to the intercrops, although there was not a significant difference in production rates between the maize sole crop and soybean sole crop or between the 1:2 intercrop and the 2:3 intercrop. Some researchers have attributed differences in production rates of  $\text{N}_2\text{O}$  to seasonal effects rather than crop type (Müller and Sherlock, 2004; Verchot et al., 2008). Variable  $\text{N}_2\text{O}$  production rates over the growing season and between years may be influenced by the availability of labile C sources and the rate of N fertilizer application (Müller and Sherlock, 2004). Mosier et al. (2006) and Sehy et al. (2003) observed low rates of production rates ( $< 50 \mu\text{g m}^{-2} \text{h}^{-1}$ ) throughout the growing season in a maize-winter wheat (*Triticum aestivum* L.) rotation in southern Germany. Soil moisture, temperature and fertilization were described as the main factors affecting seasonal  $\text{N}_2\text{O}$  production rates. Similarly, Drury et al. (2007) observed greater  $\text{N}_2\text{O}$  production rates in a maize sole crop compared to a winter wheat or soybean sole crop; they attributed this to increased N fertilizer application in the maize sole crop.

Ammonia ( $\text{NH}_3$ ) and nitrate ( $\text{NO}_3^-$ ) may act as a substrate for soil microbes and consequently encourage nitrification and denitrification (Hutchinson and Mosier, 1981). Frequently in field (Sehy et al., 2003) and laboratory (Jarecki et al., 2008) studies, denitrification rates and/or  $\text{N}_2\text{O}$  fluxes increased following fertilizer application. In November of this study, N fertilizer was hand applied to the maize in both the sole crop and intercrops, however, no spike in  $\text{N}_2\text{O}$  production rates was observed compared to that reported by Khalil et al. (2002). However, it has been reported that high  $\text{N}_2\text{O}$  production rates are heavily controlled by high soil water content (Olasantan, 1988; Reay et al., 2009; Smith et al., 2003), which was also supported by the significant results of the Pearson product moment correlation in this study.

Soil  $\text{N}_2\text{O}$  production rates are dependent on environment management interactions, that influence the balance and rates of microbial nitrification and denitrification processes and the transport of  $\text{N}_2\text{O}$  (Smith et al., 2003). Many of the key drivers of soil  $\text{N}_2\text{O}$  fluxes; soil temperature, moisture, pH, osmotic stress, and C and N availability (Adviento-Borbe et al., 2007) may be controlled by management practices (Venterea et al., 2009) such as site-specific N fertilizer application (Sehy et al., 2003). Site-specific application of N fertilizers, where only maize and maize in the intercrops received N fertilizer, may be a better management option to mitigate  $\text{N}_2\text{O}$  production rates.

#### **4.4 Conclusion**

The objective of this study was to quantify GHG production rates from maize-soybean intercrops compared to maize and soybean sole crops over two field seasons and to evaluate annual changes in GHG production rates rate from maize-soybean intercrops compared to maize and soybean sole crops. Results for this study indicated that intercropping maize with soybean, with respect to GHG production rates, is more advantageous than sole cropping. The temporal fluctuations in GHG production rates during both field seasons were most likely governed by a combination of soil temperature, soil moisture and crop residue decomposition. Crop residue decomposition is especially important in agroecosystems as it is often the only C input to the soil.

With increasing interest in promoting SOC storage and its stabilization efficiency in soil, it is vital that further field and lab studies examine the effect of complex crop residue addition on GHG production rates. Annual quantification of GHG production rates from this field site and the establishment of further intercropping trials with various maize-soybean ratios will aid in investigating the potential of these agroecosystems to stabilize soil C and help mitigate GHG production rates while at the same time maintaining agricultural productivity. Given that there is a limited amount of research on the environmental advantages of complex agroecosystems in the temperate region, continued long-term research will aid researchers and certified crop advisors in advising farmers and policy-makers on the benefits of maize-soybean intercrop agroecosystems in sequestering C.

## Chapter 5

### Soil Greenhouse Gas Production rates from Soil Amended with Soybean and Maize Crop Residues: An Incubation Study

#### 5.0 Introduction

World soils, if sustainably managed, can mitigate anthropogenic climate change by sequestering carbon dioxide (CO<sub>2</sub>) and converting it into humus through the process of carbon (C) stabilization (Lal, 2007). Numerous studies have been published examining the mitigation potential of sustainable agricultural practices in sequestering C. Some of these practices include; conservation tillage (Lal, 1997), crop rotation and intercropping (Ahmed and Rao, 1982; Prasad and Brook, 2005), use of cover crops (Buckles et al., 1998), reduced fertilizer use (Studdert and Echeverria, 2000; Aita and Giacomini, 2007) which leads to decreased nitrous oxide (N<sub>2</sub>O) production rates, crop residue management (Adiku et al., 2008), reforestation and agroforestry (Jose, 2009).

Although agriculture has the potential to mitigate GHG production rates, GHGs are remain to be highly complex and are affected by the amount, type and timing of crop residue incorporation (Jacinth et al. 2002). Crop residues influence C and nitrogen (N) levels in the soil, and store nutrients that are progressively released when mineralized by soil microorganisms (Guo et al., 2008). They are also important as available energy for soil biota (Buyanovsky and Wagner, 1986). The process of crop residue decomposition is governed first by the C/N ratios of the plant residue but as the residue becomes more decomposed, the process is controlled by the lignin contents or lignin/N ratios (Wright and Hons, 2004). Likewise, each portion of the litter is

composed of different types of compounds varying in their complexity and degree of decomposability that requires different enzymes for their degradation (Ball and McCarthy, 1989; Uphoff, 2006).

Intercropping however, provides a more diverse plant community compared to sole cropping; growing multiple crops on the same land at the same time provides a highly complex soil environment. Diversity in crop residues from intercrops influences soil quality, nutrient cycling, and microbial processes and provides complex decomposition patterns due to the interaction of residue types (McKenney et al., 1993). For example, most intercrops have a combination of both high and low quality residues, which govern faunal activity and nutrient translocation (Chapman et al., 1988; Vachon and Oelbermann 2010). A high N content, low C/N and lignin/N ratio characterize leguminous C<sub>3</sub> crops such as soybean while C<sub>4</sub> crops such as maize have a much lower N content, high C/N and lignin/N ratio. The composition of the soybean allows decomposition to occur much faster than the maize residue (Uphoff, 2006). Hobbie (1992) and Blanco-Canqui and Lal (2004) have found that intercropping can be advantageous to the long-term decomposition and cycling of nutrients as well as the storage and cycling of C and N in the soil. For instance in North Central Missouri, Mungai and Motaville (2006) found that a soybean-maize intercrop significantly increased the retention of C and N in the soil. While, Drinkwater et al. (1998) suggested that the use of low C/N residues to maintain soil fertility combined with increased temporal diversity could restore the biological linkage between C and N cycling in legume-based agroecosystems and could enhance the global C and N balances.

Presently, there is very little research on legume based intercropping and its effects on the underlying processes involved in SOC sequestration and stabilization. Moreover, there is a lack of information on how C<sub>3</sub> and C<sub>4</sub> residue decomposition effects on GHG production. The rate of GHG production from crop residue decomposition is an important step in understanding the fluxes of GHGs from agriculture. Decomposition rates of residues have been determined in the laboratory (Liang et al., 1999; Oelbermann and Schiff, 2010; Pendall and King, 2007) and field (Rochette et al., 1999) where researchers measured CO<sub>2</sub>-C originating from decomposing plant residues by isotopic techniques. However, no known research has been conducted in a laboratory on CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> production rates from maize and soybean decomposition under controlled temperature and moisture.

Therefore, the objectives of this study were too:

- 1) Quantify CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) in a short-term laboratory study where soils were incubated with C<sub>3</sub> crop residues (soybean) or C<sub>4</sub> crop residues (maize).
- 2) To analyze the temporal phases in CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> production rates over 92 days of incubation.

## **5.1 Materials and Methods**

### **5.1.1 Soil Sampling**

The study design was a randomized complete block design (RCBD) with four treatments and three replications per treatment. The treatments were a maize sole crop, soybean sole crop, 1:2 intercrop and 2:3 intercrop. Soil sampling from each treatment took place in February 2010 and represented the third year of intercrop implementation. Six random samples per replicate were extracted from the 0-20cm layer, weighed and then composited from each block using the cone and quarter technique (Schumacher et al., 1990). All samples were then air-dried and passed through a 2 mm sieve to remove coarse mineral fraction and large plant residue. Soils were then packaged and transported back to the Soil Ecosystem Dynamics Laboratory at the University of Waterloo, Ontario, Canada.

### **5.1.2 Incubation Design**

Mason jars (1000 mL) were used to incubate soil and were equipped with a lid, which contained a septum (2 cm diameter) that was used to extract the gas sample from the headspace. The incubation experiment lasted 92 days and included three sets of experiments, the first with no crop residue addition, the second with the addition of C<sub>3</sub> residues and the last with the addition of C<sub>4</sub> residue. In each experiment, 40-g of air-dried, 2 mm sieved soil from each treatment (soybean sole crop, maize sole crop, 1:2 intercrop, 2:3 intercrop) was placed in the Mason jars (Table 5.1). For example, the C<sub>3</sub> residue experiment consisted of nine jars; three with soybean soil (40-g of soil/jar), three with 1:2 intercropped soil and three with 2:3 intercropped soil. Moreover, 0.92-g of ground soybean residue was added to each jar. The C<sub>4</sub> residue

experiment also had nine jars; three with maize soil (40-g of soil/jar), three with 1:2 intercropped soil and three with 2:3 intercropped soil all with 0.92-g of ground maize residue added. An additional set of jars twelve jars from each treatment was used as a reference with no residue addition (four jars with 40-g of soybean sole crop soil; four with maize sole crop soil, four with 1:2 intercropped soil and four with 2:3 intercropped soil).

Prior to incubation, soil in all jars was adjusted to 60% of field capacity (7.05 mL/40-g of soil) using deionized (DI) water, which is considered optimum for microbial activity (Linn and Doran, 1984). The wet soil was then left for two hours to ensure proper and even distribution of water before crop residues were added. Moisture content was kept constant ( $\pm 0.50$  mL) during the 92 day incubation by weighing the jars the day prior to sampling and adding the differences in moisture. All jars were covered on all sides to try to prevent exposure to light and were incubated at a temperature of 21 °C ( $\pm 0.5$ ).

**Table 5.1- Incubation Design for two experiments using C<sub>3</sub> residue (soybean) and C<sub>4</sub> residue (maize) addition to soil as well as a no residue addition used as a control. Soil is from four different field treatments, each treatment was replicated three times (n=3).**

	Type of crop residue added	Amount of crop residue added (grams)	Type of cropped soil added	Amount of soil added (grams)
<b>Experiment #1 - No residue Addition</b>				
<b>Soybean Sole Crop</b>		None	Soybean	40
<b>Maize Sole Crop</b>		None	Maize	40
<b>1:2 Intercrop</b>		None	1:2 Intercrop	40
<b>2:3 Intercrop</b>		None	2:3 Intercrop	40
<b>Experiment #2 - C<sub>3</sub> Residue Addition</b>				
<b>Soybean Sole Crop</b>	C <sub>3</sub> - Soybean	0.92	Soybean	40
<b>1:2 Intercrop</b>	C <sub>3</sub> - Soybean	0.92	1:2 Intercrop	40
<b>2:3 Intercrop</b>	C <sub>3</sub> - Soybean	0.92	2:3 Intercrop	40
<b>Experiment #3 - C<sub>4</sub> Residue Addition</b>				
<b>Maize Sole Crop</b>	C <sub>4</sub> - Maize	0.92	Maize	40
<b>1:2 Intercrop</b>	C <sub>4</sub> - Maize	0.92	1:2 Intercrop	40
<b>2:3 Intercrop</b>	C <sub>4</sub> - Maize	0.92	2:3 Intercrop	40

### 5.1.3 Quantification of Greenhouse Gas Fluxes

CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> were extracted from the headspace of the Mason jar every week and were immediately analyzed in the gas chromatograph (GC) (Aglient 6890N, California, USA). After extraction the Mason jars were opened for a period of 30 minutes after which they were sealed until the next measurement day. A gas standard consisting of 99.9 ppm CO<sub>2</sub>, 5.0 ppm CH<sub>4</sub>, and 10.0 ppm N<sub>2</sub>O was injected at a 10-sample interval.

Daily GHG production rates were determined using the following equation adapted from Hogg et al. (1992):

$$R = (VD / t) [(C_S - C_A) / M] \quad (5.1)$$

where  $R$  ( $\mu\text{g}/\text{g}^{-1}/\text{d}^{-1}$ ) is the quantity of the analyte of interest (CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub>) ( $\mu\text{g}$ ) evolved per gram of dry soil per day;  $V$  is the volume of effective headspace (L);  $D$  is the density of the analyte (CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub>) adjusted for temperature at 21°C, pressure and humidity ( $\text{g L}^{-1}$ );  $C_S$  is the concentration of CO<sub>2</sub>, N<sub>2</sub>O or CH<sub>4</sub> evolved from the soil ( $\mu\text{L}^{-1}$ );  $C_A$  is the concentration of CO<sub>2</sub>, N<sub>2</sub>O or CH<sub>4</sub> in the blank jars ( $\mu\text{L}^{-1}$ );  $M$  is the dry mass of the soil sample (g) and  $t$  is the sampling time interval in days.

#### 5.1.4 Statistical Analysis

All data was examined for homogeneity of variance using the Levene Test and goodness of fit using the Kolmogorov-Smirnov (K-S) Test but CO<sub>2</sub> and N<sub>2</sub>O production rates were found to have unequal variances (Zar, 2007). Therefore, a nonparametric analysis was run using SPSS (SPSS Science Inc., v. 15.0, 2006) to determine significant differences between treatments. The Kruskal-Wallis Analysis of Variance and a Wilcoxon mann-Whitney Test (also referred to as the Mann-Whitney-U) were implemented to determine differences within experiments (blank, C<sub>3</sub> and C<sub>4</sub>) between treatments. To compare between residue addition and no residue addition, within treatments (sole crops and intercrops) a Wilcoxon Signed Ranks Test was run in SPSS. Since GHG production rates data for each experiment was found to an unequal distribution, a Spearman Rank-Order Correlation was used to determine the relationship between GHG production rates in each experiment.

For CH<sub>4</sub> production rates a one-way analysis of variance (ANOVA) was run in SPSS and was used to compare between treatments for jars that received either C<sub>3</sub> or C<sub>4</sub> added residue. Any significant differences were further analyzed using Tukey's least significant difference multiple comparison test (Steel et al., 1997; Zar, 2007). Data comparing between residue addition and no residue addition within treatments were tested for differences using a paired t-test (Zar, 2007). For all statistical analyses, the threshold probability level for determining significant differences was  $P < 0.05$ .

## 5.2 Results

In all experiments, soil CO<sub>2</sub> and N<sub>2</sub>O production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) showed a significantly positive correlation to each other but showed a negative correlation to CH<sub>4</sub> production rates (Table 5.2). GHG production rates varied between treatments and between residue addition for both CO<sub>2</sub> and N<sub>2</sub>O. For example, mean CO<sub>2</sub> production rates ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ) fluctuated between a minimum of  $120.99 \pm 6.60$  (Soybean sole crop – no residue addition) to a maximum of  $217.15 \pm 11.92$  (Maize Sole Crop – C<sub>4</sub> residue addition) (Table 5.2). CO<sub>2</sub> production rates were strongly influenced by the type of residue added. Soils incubated without any crop residue (Experiment 1) addition had the lowest CO<sub>2</sub> production rates ( $11.1 \pm 1.8$  to  $192.0 \pm 20.4$ ), followed by C<sub>3</sub> residue addition (experiment 2) ( $11.5 \pm 0.6$  to  $272.3 \pm 0.7$ ) and lastly the C<sub>4</sub> residue addition (experiment 3) ( $12.1 \pm 0.5$  to  $305.1 \pm 10.6$ ). CO<sub>2</sub> production rates were significantly different within treatments between experiments (i.e. soybean without residue addition compared to soybean with C<sub>3</sub> residue addition), but were not significantly different within treatments between experiment 2 and 3 (with crop residue addition) (Table 5.3).

Expectedly, mean CH<sub>4</sub> production rates ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ) varied only narrowly between treatments and experiments ( $-0.04 \pm 0.01$  to  $0.04 \pm 0.01$ ). No significant differences ( $P > 0.05$ ) were observed in CH<sub>4</sub> production rates between experiments or treatments. There was a negative correlation between CH<sub>4</sub> and N<sub>2</sub>O and CH<sub>4</sub> and CO<sub>2</sub> ( $P < 0.001$ ), however there was a positive correlation between CO<sub>2</sub> and N<sub>2</sub>O. Mean N<sub>2</sub>O production rates ( $\mu\text{g N g}^{-1} \text{d}^{-1}$ ) varied between a minimum of  $2.5 \pm 0.2$  (maize sole crop – no residue addition) to a max of  $5.7 \pm 0.3$  (soybean sole crop – C<sub>3</sub> residue addition). Moreover, N<sub>2</sub>O was strongly influenced by residue addition;

production rates were the lowest in experiment 1 ( $0.1 \pm 0.1$  to  $4.7 \pm 0.8$ );  $0.2 \pm 0.1$  to  $7.8 \pm 0.4$  in experiment 3 and highest in experiment 2 ( $0.2 \pm 0.1$  to  $8.2 \pm 0.2$ ). Significant differences were found between all treatments within experiment 1, except for the soybean sole crop and maize sole crop, which were not significantly different. In experiment 2, the 1:2 and 2:3 intercrop were significantly different from the soybean sole crop while in experiment 3 ( $C_4$  residue addition) the maize sole crop and 2:3 intercrop were significantly different from each other but were not different from the 1:2 intercrop. Moreover, when comparing differences between experiments, within treatments, all treatments in experiment 1 were significantly different between the experiments with residue addition (experiment 2 and 3).

In experiment 1, there was no distinct temporal change in  $CO_2$  production rates ( $\mu g C g^{-1} d^{-1}$ ). All treatments steadily increased until day 28 (Figure 5.1), although the sole crops had a greater increase in production rates compared to the intercrops (sole crops  $5.6 \pm 1.3$ ; intercrops  $3.3 \pm 0.1$ ). After day 28, treatments generally reached a steady-production rates for the remainder of the incubation period (mean for all treatments from day 28-91:  $-0.6 \pm 1.0$ ). Experiment 2 had two main temporal phases (Figure 5.1); phase 1: all treatments had a steady  $CO_2$  production rates rate increase (Soybean  $6.36 \pm 1.03$ ; 1:2 intercrop  $5.6 \pm 0.1$ ; 2:3 intercrop  $5.6 \pm 0.3$ ). After day 42 all treatments steadily declined at rates of  $4.0 \pm 1.1$  (soybean),  $2.3 \pm 0.6$  (1:2 intercrop),  $2.5 \pm 1.6$  (2:3 intercrop) (Phase 2). Conversely, experiment 3 had distinct temporal phases (Figure 5.1). There was a gradual increase in all treatments (Mean rate of change:  $8.0 \pm 0.9$ ) until day 21 (Phase 1). After which the maize continued to increase at a much faster rate ( $3.1 \pm 1.7$ ) compared to the 1:2 and 2:3 intercrop which increased at much slower rates (1:2 intercrop:  $1.0 \pm$

1.2; 2:3 intercrop:  $1.7 \pm 0.1$ ) (Phase 2). The third phase is split with the intercrops steadily decreasing from day 63 to day 92 (1:2 intercrop  $3.4 \pm 2.0$  and the 2:3 intercrop  $3.7 \pm 1.2$ ). However the maize sole crop reached almost steady state between day 63 and 70 ( $-0.2 \pm 0.6$ ) and then steadily decreased until the end of the incubation ( $-5.8 \pm 1.4$ ).

$\text{CH}_4$  (Figure 5.2) and  $\text{N}_2\text{O}$  (Figure 5.3) production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) showed much more variability than  $\text{CO}_2$  with few visible temporal phases throughout the incubation. Methane production rates ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ) had only one visible trend; all treatments in all experiments had a positive flux between 0.01 and 0.04 during day 1 of sampling and decreased by day 7 fluctuated at or near 0 ( $-0.035$  to  $0.016 \pm 0.001$ ). All experiments demonstrated an increase in  $\text{N}_2\text{O}$  production rates ( $\mu\text{g N g}^{-1} \text{d}^{-1}$ ) during the first 14 days of incubation; experiment 1 had a mean increase of  $0.2 \pm 0.1$  experiment 2,  $0.4 \pm 0.1$  and experiment 3,  $0.3 \pm 0.4$ . After the first 2 weeks of incubation, mean rates of increase fluctuated between 1.5 and 4.0 (Experiment 1) while experiment 2 was very variable fluctuating between 3.6 and 8.2 and experiment 3 had extreme weekly fluxes in all treatments.

**Table 5.2 – Spearman Rank-order Correlation between soil CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> production rates (µg analyte g<sup>-1</sup> d<sup>-1</sup>) rates for each incubation experiment. One using C<sub>3</sub> residue (soybean) (Experiment 2) and the other using C<sub>4</sub> residue (maize) addition (Experiment 3), as well as a control with no residue addition used (Experiment 1) (n=6).**

	<b>CO<sub>2</sub> Production rates</b>	<b>N<sub>2</sub>O Production rates</b>	<b>CH<sub>4</sub> Production rates</b>
<b>Experiment #1 - No residue Addition</b>			
<b>CO<sub>2</sub> Production rates</b>	1.000		
<b>N<sub>2</sub>O Production rates</b>	0.228**	1.000	
<b>CH<sub>4</sub> Production rates</b>	-.560**	-.332	1.000
<b>Experiment #2 - C<sub>3</sub> Residue Addition</b>			
<b>CO<sub>2</sub> Production rates</b>	1.000		
<b>N<sub>2</sub>O Production rates</b>	.445**	1.000	
<b>CH<sub>4</sub> Production rates</b>	-.741**	-.658**	1.000
<b>Experiment #3 - C<sub>4</sub> Residue Addition</b>			
<b>CO<sub>2</sub> Production rates</b>	1.000		
<b>N<sub>2</sub>O Production rates</b>	.412**	1.000	
<b>CH<sub>4</sub> Production rates</b>	-.661**	-.388	1.000

\*\* Correlation is significant at the 0.01 level (2-tailed)

**Table 5.3 – Mean soil CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> production rates (µg analyte g<sup>-1</sup> d<sup>-1</sup>) from a 92 day incubation using two experiments. One using C<sub>3</sub> residue (soybean) (Experiment 2) and the other using C<sub>4</sub> residue (maize) addition (Experiment 3), as well as a control with no residue addition used (Experiment 1). Standard error is in parenthesis (n=42).**

Treatment	CO <sub>2</sub> (µg C g <sup>-1</sup> d <sup>-1</sup> )	N <sub>2</sub> O (µg N g <sup>-1</sup> d <sup>-1</sup> )	CH <sub>4</sub> (µg C g <sup>-1</sup> d <sup>-1</sup> )
<b>Experiment #1: Blank – No residue Addition</b>			
<b>Soybean Sole Crop</b>	120.99 (6.60) <sup>A, a</sup>	2.48 (0.15) <sup>A, a</sup>	-0.007 (0.003) <sup>A, a</sup>
<b>Maize Sole Crop</b>	142.65 (6.29) <sup>B, a</sup>	2.45 (0.16) <sup>A, a</sup>	0.004 (0.003) <sup>A, a</sup>
<b>1:2 Intercrop</b>	121.19 (5.27) <sup>A, a</sup>	2.94 (0.15) <sup>B, a</sup>	0.002 (0.003) <sup>A, a</sup>
<b>2:3 Intercrop</b>	108.44 (9.84) <sup>A, a</sup>	2.98 (0.34) <sup>C, a</sup>	0.000 (0.003) <sup>A, a</sup>
<b>Experiment #2 - C<sub>3</sub> Residue Addition</b>			
<b>Soybean Sole Crop</b>	159.31 (10.10) <sup>AB, b</sup>	5.69 (0.29) <sup>A, b</sup>	-0.007 (0.003) <sup>A, a</sup>
<b>1:2 Intercrop</b>	150.84 (9.10) <sup>A, b</sup>	4.74 (0.23) <sup>B, b</sup>	-0.008 (0.004) <sup>A, a</sup>
<b>2:3 Intercrop</b>	166.92 (12.07) <sup>B, b</sup>	4.68 (0.25) <sup>B, b</sup>	-0.004 (0.004) <sup>A, a</sup>
<b>Experiment #3 - C<sub>4</sub> Residue Addition</b>			
<b>Maize Sole Crop</b>	217.15 (11.92) <sup>A, b</sup>	4.30 (0.27) <sup>AB, b</sup>	-0.002 (0.003) <sup>A, a</sup>
<b>1:2 Intercrop</b>	167.35 (9.38) <sup>B, b</sup>	4.76 (0.29) <sup>B, c</sup>	-0.005 (0.003) <sup>A, a</sup>
<b>2:3 Intercrop</b>	178.42 (9.45) <sup>C, b</sup>	5.31 (0.32) <sup>CB, b</sup>	-0.002 (0.003) <sup>A, a</sup>

Means followed by the different uppercase letter are significantly different (0.05) between treatments within experiments. Means followed by the different lowercase letter are significantly different (0.05) between experiments within treatments.

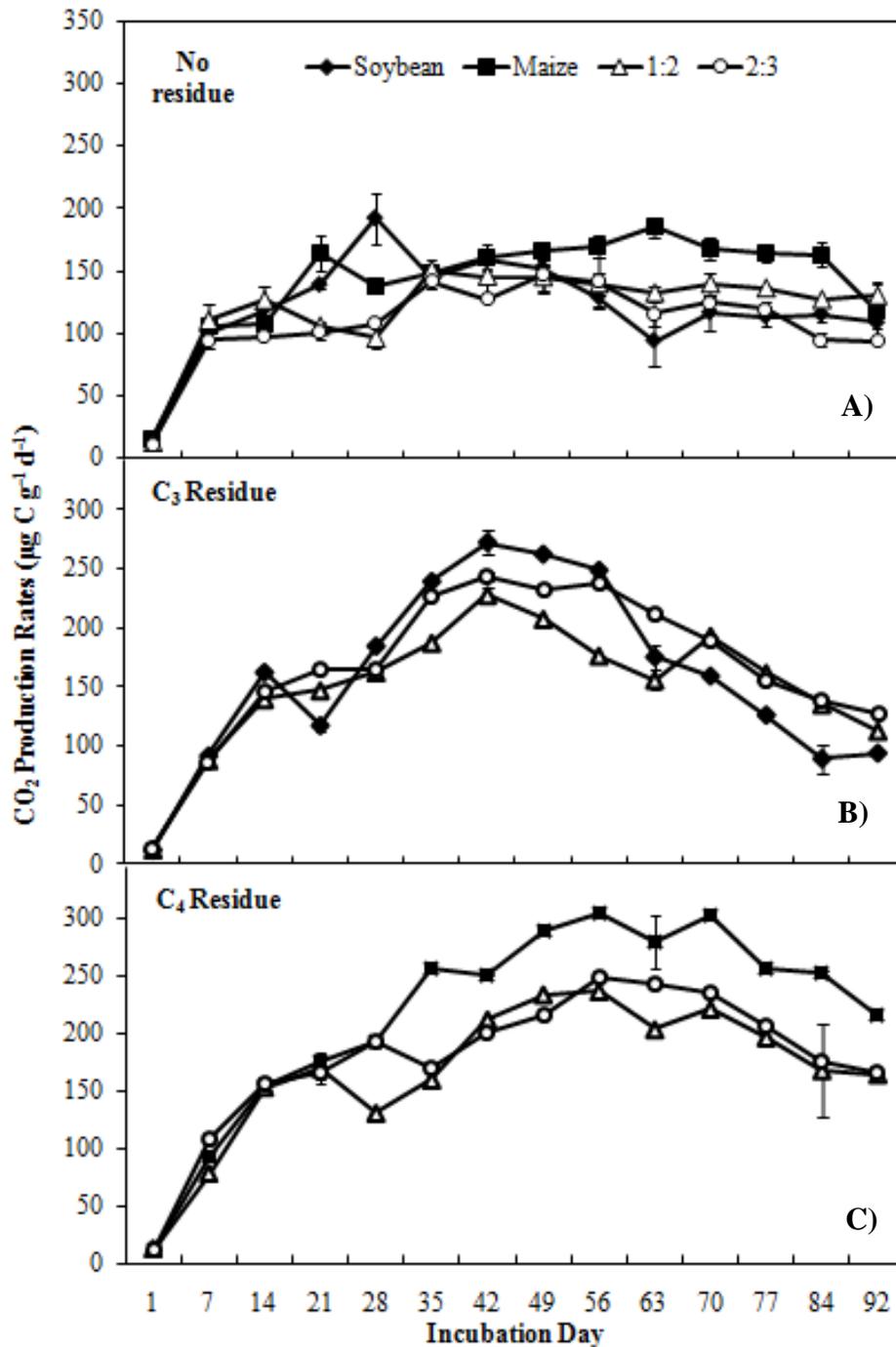


Figure 5.1- CO<sub>2</sub> Production rates (µg C g<sup>-1</sup> d<sup>-1</sup>) from soils taken from four different treatments (Soybean sole crop, Maize sole crop, 1:2 and 2:3 intercrops) in the Argentine Pampa. A) No residue added to the soil B) C<sub>3</sub> (Soybean Residue Addition) C) C<sub>4</sub> (Maize Residue Addition). Vertical Bars represent standard errors.

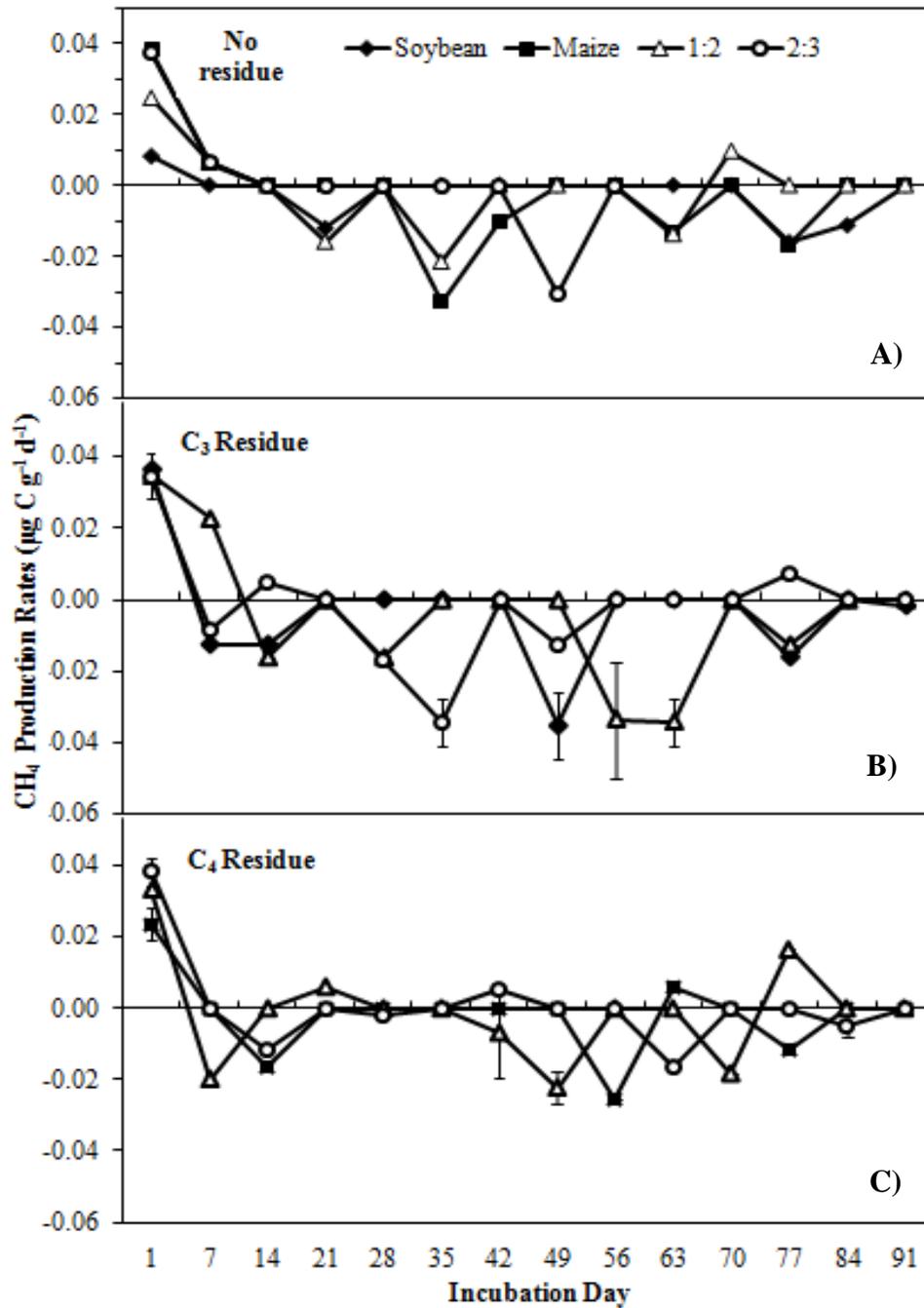


Figure 5.2 - CH<sub>4</sub> Production rates (µg C g<sup>-1</sup> d<sup>-1</sup>) from soils taken from four different treatments (Soybean sole crop, Maize sole crop, 1:2 and 2:3 intercroops) in the Argentine Pampa. A) No residue added to the soil B) C<sub>3</sub> (Soybean Residue Addition) C) C<sub>4</sub> (Maize Residue Addition). Vertical Bars represent standard errors.

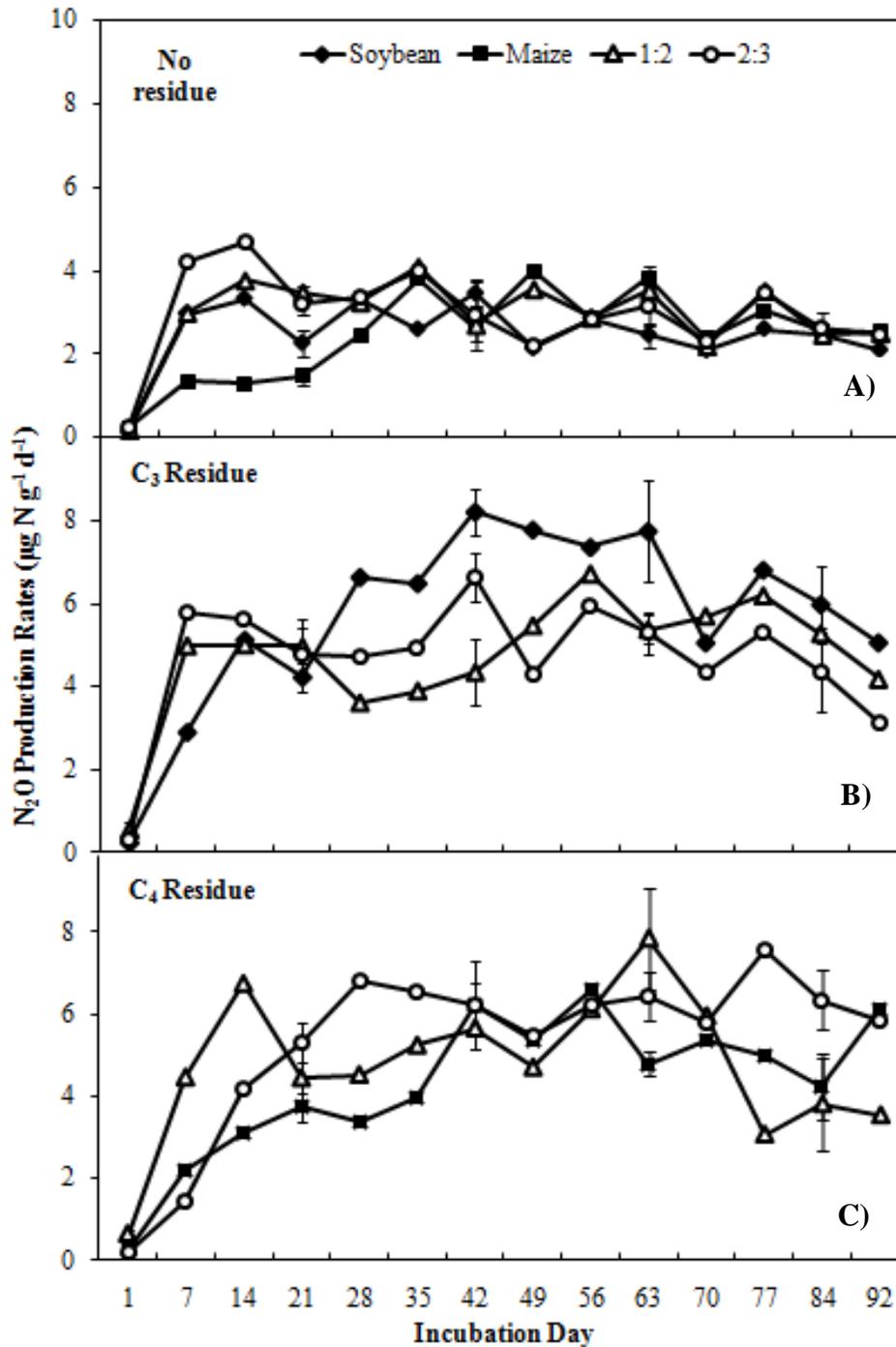


Figure 5.3 - N<sub>2</sub>O Production rates (µg N g<sup>-1</sup> d<sup>-1</sup>) from soils taken from four different treatments (Soybean sole crop, Maize sole crop, 1:2 and 2:3 intercrops) in the Argentine Pampa. A) No residue added to the soil B) C<sub>3</sub> (Soybean Residue Addition) C) C<sub>4</sub> (Maize Residue Addition). Vertical bars represent standard errors.

### 5.3 Discussion

CO<sub>2</sub>, N<sub>2</sub>O, and CH<sub>4</sub> production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) were quantified in a short-term incubation study where soils were amended with C<sub>3</sub> (soybean) or C<sub>4</sub> crop residues (maize). Although, CH<sub>4</sub> varied only narrowly between treatments and experiments ( $-0.035 \pm 0.009$  to  $0.039 \pm 0.004$  ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ); CO<sub>2</sub> and N<sub>2</sub>O production rates had visibly higher production rates between the different types of crop residue additions. The cumulative amount of CO<sub>2</sub> and N<sub>2</sub>O ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) evolved from the incubated soil was highest in both experiments with residue addition but was lowest in experiment 1 when no crop residue was added, this suggests that production rates were strongly influenced by type of crop residue added. For example, the addition of soybean residue likely influenced C and N mineralization through the parallel assimilation of C and N by heterotrophic soil organisms (Vachon and Oelbermann, 2010). The conversion of a sole crop to a maize-soybean intercrop may result in a greater contribution to the long-term availability of N for crop utilization and the stabilization of SOM and sequestration of C (Vachon and Oelbermann, 2010).

According to Rastogi et al. (2002), CO<sub>2</sub> production rates are approximately two to three times greater in soil amended with crops residues than from bare soil. Similarly, the type of crop residue present in the soil and the aeration status of the soil influences both nitric oxide (NO) and N<sub>2</sub>O production rates (Drury et al., 1991; McKenney et al., 1993; McKenney and Drury, 1997). In a 2 year field study in the United Kingdom, Baggs et al. (2003) established that greater N<sub>2</sub>O production rates were measured in the presence of wheat (*Triticum aestivum*) residues than that from the bare soil controls. Generally, residue decomposition reduces SOC because of increased

contact between the residue and soil microbes thereby accelerating the decomposition and loss of soil C (Adiku et al., 2008). Evidence from medium to long-term (14 year) studies, suggest that the quality of soil C is derived from the quality of the added residue and this affects the SOC mineralization rates (Huggins et al., 2007).

In this study, there was a rapid increase in CO<sub>2</sub> and N<sub>2</sub>O production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) during the first two weeks of sampling. Adiku et al. (2008) used mineralization rates as a measure of C dynamics in Ghana and found that treatments involving maize-legume rotations had significantly higher mineralization rates than those treatments that derived their biomass inputs solely from cereal and grass sources. This was attributed to differences in soil quality, which affected mineralization rates, even in such a short-term study (120 days). Soil CO<sub>2</sub> and N<sub>2</sub>O production rates have also been shown to increase after application of inorganic fertilizer or incorporation of crop residues; this initial increase in CO<sub>2</sub> is generally thought to be due to the rapid degradation of readily decomposable, water-soluble, low molecular weight components of applied crop residue (Robertson and Paul, 2000). For example, Berg et al. (1987) attributed the rapid release of C from clover (*Trifolium pretense L.*) roots to almost complete decomposition of all polysaccharides. Some researchers however, argue that CO<sub>2</sub> evolution rates increase during the first week of incubation due to microbial stimulation caused by disturbance (Kirschbaum, 1995). Franzluebbers et al. (1994) found that the rate of C loss increased in a loamy sand soil from East Texas, USA, during the first seven days of incubating with cowpea residue (*Vigna unguiculata L.*) Walp) addition. This however, was directly correlated to the size of the active microbial biomass pool during the first seven days after cowpea incorporation. During the

remainder of their incubation period, the active microbial biomass was much smaller which coincided to the reduced rate of cowpea decomposition (Franzluebbers et al., 1994). According to Oelbermann et al. (2008), the highest peak in CO<sub>2</sub> production rates and rate of decomposition can be attributed to a peak in microbial activity before the labile C is depleted, eventually leading to a decreased CO<sub>2</sub> flux and stabilization as only recalcitrant materials are available for decomposition.

As discussed previously, decomposition of crop residues plays a key role in the C and N cycle in agroecosystems. Decomposition is firstly governed by C/N ratios but as residues become more decomposed, the process is controlled by the lignin contents or lignin/N ratios (Wright and Hons, 2004). Likewise, each portion of the litter is composed of different types of compounds varying in their complexity and degree of decomposability that requires different enzymes for their degradation (Ball and McCarthy, 1989; Uphoff, 2006). During incubation, CO<sub>2</sub> production rates ( $\mu\text{g C g}^{-1} \text{ d}^{-1}$ ) from C<sub>3</sub> residue decomposition sharply increased until day 42 after which they steadily declined. Conversely, CO<sub>2</sub> production rates from the C<sub>4</sub> residue had a more gradual increase overtime and showed very little decline by the end of the incubation. This is attributed to the low C/N ratio (31) in the soybean residue (Appendix B), which results in rapid decomposition and thus increases CO<sub>2</sub> and N<sub>2</sub>O production rates more rapidly than following the incorporation of maize residues, which have a much higher C/N of 66 (Appendix B) (Cattanio et al., 2008). The early stage of soybean residue decomposition has a rapid loss of N from the residue, which is associated with the release of soluble and readily decomposable N containing substances (Howarth, 2007).

During this incubation, CH<sub>4</sub> production rates ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ) were highest during the first sampling day; this may be attributed to the addition of crop residues that are easily biodegradable, which enhances mineralization thereby increasing O<sub>2</sub> consumption by heterotrophs (Wuebbles and Hayhoe, 2002). Moreover, the initial addition of water at the beginning of the incubation could have contributed to anaerobic conditions, which may cause increased methanotroph activity and thus CH<sub>4</sub> production (Wuebbles and Hayhoe, 2002). Methane production rates ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ) did not differ significantly between treatments or experiments and were barely detectable after 92 days of incubation. Similarly, Ellert and Janzen (2008) found that CH<sub>4</sub> uptake was scarcely detectable in a maize sole crop system with soybean residue addition. In a comparable experiment, Mosier et al. (2006) reported that CH<sub>4</sub> production rates ranged between -2 and + 2 ( $\mu\text{g C m}^{-2}\text{h}^{-1}$ ). While in an agroforestry field study in Brazil, Verchot et al. (2008) found that maize intercropped with leguminous tree species resulted in a net sink of CH<sub>4</sub>.

N<sub>2</sub>O production rates ( $\mu\text{g N g}^{-1} \text{d}^{-1}$ ) were visibly higher when C<sub>3</sub> residues were added compared to the C<sub>4</sub> residue addition. This is in strong agreement with previously reported results (Kaiser et al., 1998; Baggs et al., 2000) and was attributed to rapid release of N from N-rich legumes resulting in available N for nitrification or denitrification. Similarly, McKenney et al. (1993) found that N<sub>2</sub>O production was greater in soils amended with a legume residue than with corn residue. This is because the low C/N ratio of a leguminous residues results in greater NO and N<sub>2</sub>O production rates than residues with higher C/N ratios such as maize (McKenney et al., 1993). For example, Baggs et al. (2003) found that N<sub>2</sub>O production rates were higher in

unfertilized conventional and no-till treatments following incorporation of bean residues than following the incorporation of rye residues (*Secale cereale* L.), which have a high C/N ratio. Leguminous plants release higher levels of N<sub>2</sub>O production in the soil directly, by rhizobia denitrification or indirectly by increasing inputs of N to the soil and thus increasing the substrate stock for nitrification and denitrification (O'Hara and Daniel, 1985).

In this study, CO<sub>2</sub> and N<sub>2</sub>O production rates ( $\mu\text{g analyte g}^{-1} \text{d}^{-1}$ ) showed a significantly positive correlation to each other in all experiments (Table 5.2). Correspondingly, numerous studies have found a significantly positive relationship between CO<sub>2</sub> and N<sub>2</sub>O production rates (Rice et al., 1988; Baggs et al., 2003; Sehy et al., 2003; Liu et al., 2007; Sey et al., 2008). This indicates that the CO<sub>2</sub> and N<sub>2</sub>O production rates peaked simultaneously with maximum soil microbial activity (Rice et al., 1988). Conversely, CH<sub>4</sub> production rates ( $\mu\text{g C g}^{-1} \text{d}^{-1}$ ), were negatively correlated to both CO<sub>2</sub> and N<sub>2</sub>O production rates (Table 5.2). Similarly, several studies found that agricultural systems with improved CH<sub>4</sub> uptake negatively affected N<sub>2</sub>O production rates (Koga et al., 2004 and Mullet and Sherlock, 2004). Johnson et al., (2007) found that the factors that regulate a soil's CH<sub>4</sub> capacity include moisture and N levels. However, in terms of CO<sub>2</sub> and CH<sub>4</sub>, this study contradicts what one would expect because CO<sub>2</sub> and CH<sub>4</sub> have similar flux-controlling processes such as soil moisture, amount of leaf litter, and temperature (Dong et al., 1998). For instance, Dong et al. (1998) found a positive correlation between CH<sub>4</sub> uptake and CO<sub>2</sub> production in a deciduous forest soil in Southern Germany. Similarly, significant positive relationships between inorganic N stocks, N availability or rates of nitrification and CH<sub>4</sub>

flux rates have been reported in agriculture (Hütsch, 1996), fertilized forests (Castro et al., 1994; Steudler et al., 1996) and fertilized grasslands (Mosier et al., 1997).

## **5.4 Conclusion**

The objective of this study was to quantify GHG production rates from maize-soybean intercrops compared to maize and soybean sole crops. There is a beneficial effect when a leguminous plant is intercropped with a non-leguminous plant such as a cereal. Results from this study provide evidence of this beneficial effect, as intercropping maize with soybean proved to be more advantageous than sole cropping in regards to GHGs. The temporal fluctuations in GHG production rates during this incubation were most likely governed by crop residue decomposition, as soil moisture and temperature were controlled during the length of the incubation. Crop residue decomposition is especially important in agroecosystems as they are often the only C input to the soil. As such, it is essential to understand the dynamics of their decomposition in order to determine the effects of soil and crop management on soil C sequestration and GHG production rates.

With increasing interest in promoting SOC storage and its stabilization efficiency in soil, it is vital that further field and lab studies examine the effect of complex crop residue addition on GHG production rates. Although this study adds to the current literature on the influence of residue quality on GHG production rates in agroecosystems, it is imperative that further detailed studies are developed to evaluate C and N transformations in soil from intercropping systems.

This will broaden the knowledge base and understanding of GHG dynamics in different land-use systems and the most effective intercrop design. Moreover, in order to further investigate the processes of decomposition and C stabilization in maize-soybean intercrops it is recommended that stable isotope techniques be employed. Recently, the analysis of isotopic composition of SOM stocks has resulted in further insight into turnover rates and microbial processing. Recent advances in the use of  $\delta^{13}\text{C}$  values of respired  $\text{CO}_2$  to identify sources of stocks for the  $\text{CO}_2$  efflux from the soil surface have been made. With the use of  $\delta^{13}\text{C}$ , the flow of C between  $\text{C}_3$  and  $\text{C}_4$  residues could be identified.

## Chapter 6 Conclusion and Recommendations

### 6.0 Summary and Conclusions

Trends in greenhouse gas production rates (GHG) and soil quality in the agricultural sector depend mainly on the level and rate of socio-economic development, population growth, adequate technologies and agricultural policy. However, mitigation potentials in the agricultural sector are indeterminate, making a consensus difficult to achieve and hindering policy making (IPCC, 2007c). The long-term outlook for GHG mitigation in agriculture suggests that there is a significant potential, but many uncertainties will determine the level of implementation. Thus, further research is needed to determine the long-term effects of sustainable agricultural management practices on GHGs. Moreover, with appropriate policies, education and incentives it may be possible for agriculture to make a significant contribution to climate mitigation.

The purpose of this research was to determine the effects of maize-legume intercrops compared with maize and soybean sole crops on GHG production rates and soil physical properties over two field seasons, and to quantify GHG production rates from soils amended with maize and soybean crop residues. Although results from the field study showed that soil organic carbon (SOC) concentrations were significantly greater in the maize sole crop and intercrops than the soybean sole crops this was expected as the maize would contribute a greater input of litterfall and biomass than the soybean. Soil CO<sub>2</sub> production rates were significantly greater in the maize sole crop but did not differ significantly for N<sub>2</sub>O production rates. However,

over the two field seasons both trace gases showed a general trend of greater production rates in the maize sole crop followed by the soybean sole crop and were lowest in the intercrops.

Linear regression between soil GHG production rates and soil temperature or volumetric soil moisture accounted for up to 51% of the variability in soil CO<sub>2</sub> production rates and 60% of the soil N<sub>2</sub>O production rates. Moreover, the variation between field seasons can be attributed to natural seasonal variations in local climate patterns that expectedly influenced diurnal CO<sub>2</sub> and N<sub>2</sub>O production rates (Smith et al., 2007). Temperature and moisture are two of the most influential environmental factors affecting the rate of nutrient cycling and the production of GHGs (Kirschbaum, 1995; Smith, 1997).

In the laboratory study, soil GHG production rates varied between treatments and between residue addition for both CO<sub>2</sub> and N<sub>2</sub>O but varied only narrowly between treatments and experiments for CH<sub>4</sub>. However, this was expected as it reflects the contribution of soybean and maize derived C. Drury et al. (1991) found that C substrate supply is one of the key factors controlling denitrification and CO<sub>2</sub> production rates in incubation studies involving soils with a range of soil physical and chemical characteristics. According to Rastogi et al. (2002), CO<sub>2</sub> production rates are approximately two to three times greater in soil amended with crops residues than from bare soil.

## 6.1 Recommendations for Future Research

This study provided further insight into the effect of agroecosystem management practices on GHG production rates and soil physical and chemical characteristics. However, due to time and resources there are limitations to these experiments that need to be addressed in the future. Specifically, GHG production rates from the field need to be sampled weekly if not twice weekly and sampling needs to take place over a longer time period, ideally the entire growing season. With a larger sampling scheme, a better portrait of GHG production rates can be seen in these complex agroecosystems as opposed to a snap shot of only part of the growing season. Moreover, if GHG production rates are taken over a longer time-period the global warming potential (GWP) can be calculated in order to better cross-examine this agroecosystem with others.

Secondly, the analysis of isotopic composition of SOC in the incubation experiment could have provided further insight into turnover rates and microbial processing. Recent advances in the use of  $\delta^{13}\text{C}$  values of respired carbon dioxide ( $\text{CO}_2$ ) have been used to identify sources of  $\text{CO}_2$  efflux from the soil surface; moreover with the use of  $\delta^{13}\text{C}$  the flow of C between  $\text{C}_3$  and  $\text{C}_4$  residues could be identified. In broader terms, the effect of increased atmospheric GHG production rates from anthropogenic climate change needs to be studied, as it will likely affect the growth, timing and productivity of agricultural crops. Different combinations of crop species and densities also need to be included in order to improve our understanding of optimal agroecosystem design. Although all studies will have limitations, there

is a need for an improved understanding of sustainable agricultural practices and their impacts on anthropogenic climate change.

GHG production rates from agriculture will always have uncertainties and it is difficult to assess the effectiveness of GHG mitigation measures under the changing climate conditions (Smith et al., 2007). However, reduced GHG production rates from sustainable agricultural practices look promising and as a society, we need to take ownership of the impacts we are having on the environment and begin to share our innovative technologies for efficient use of land resources, as this will be the first step in mitigating GHG production rates from agriculture. However, our greatest challenge will be to remove the social, economic and political barriers in order to ensure that GHG mitigation options will be implemented in the future.

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**Appendix A**  
**Agronomic information of experimental treatments in a RCBD in the Argentine Pampas. Balcarce, Argentina.**

<b>Growing Season</b>		<b>2008/2009</b>	<b>2009/2010</b>	
<b>Crop Variety</b>	Maize Sole Crop	DK 682 (Long-cycle)		
	Maize Intercrop	DKC51-45AR2 (Short-cycle)		
	Soybean	NIDERA 4613		
<b>Weed Control</b>		Glyphosate		
<b>Row Spacing</b>		52 cm		
<b>Plant Spacing</b>	Maize Sole Crop	24 cm		
	Soybean Sole Crop	6.5 cm		
	Maize Intercrop	14.5 cm		
	Soybean Intercrop	4 cm		
<b>Number of Rows</b>	Sole Crops	14		
	Intercrops	17		
<b>Sowing</b>	Maize Sole Crop & Intercrop	Oct 22/08	Oct 21/09	
	Soybean Sole Crop & Intercrop	Dec 3/08	Nov 18/09	
<b>Inoculation of soybean</b>		Bradyrhizobium Japonicum		
<b>Fertilizer</b>	Maize Sole Crop & Intercrop	150 k ha <sup>-1</sup> of urea		
<b>Harvest</b>	Maize Sole Crop & Intercrop	Mar 30/09	Mar 26/10	
	Soybean Sole Crop & Intercrop	May 4/09	Apr 28/10	
<b>Plant Densities</b> (plants/m <sup>-2</sup> )	Maize Sole Crop	8 plants m <sup>-2</sup>		
	Maize 1:2 Intercrop	4.4 plants m <sup>-2</sup>		
	Maize 2:3 Intercrop	5.3 plants m <sup>-2</sup>		
	Soybean Sole Crop & Intercrop	30 plants m <sup>-2</sup>		
<b>Grain Yield</b> (g/m <sup>-2</sup> )	Maize Sole Crop	1120	1332	
	Soybean Sole Crop	234	214	
	1:2 Intercrop	Maize	784	764
		Soybean	45	82
	2:3 Intercrop	Maize	797	793
		Soybean	45	53
<b>Final Biomass</b> (g/m <sup>-2</sup> )	Maize Sole Crop	2187	2593	
	Soybean Sole Crop	681	569	
	1:2 Intercrop	Maize	1306	1363
		Soybean	170	355
	2:3 Intercrop	Maize	1496	1597
		Soybean	177	358

**Appendix B**  
**Aboveground (shoots and leaves) crop residue biomass carbon and nitrogen input ( $\text{g m}^{-2} \text{yr}^{-1}$ ) from maize and soybean in a maize and soybean sole crop in two differently configured intercropping systems in the Argentine Pampa.**

	<b>Maize Sole Crop</b>	<b>Soybean Sole Crop</b>	<b>1:2 Intercrop</b>	<b>2:3 Intercrop</b>
<b>Carbon</b>	795 (25) <sup>a</sup>	407 (32) <sup>b</sup>	451 (34) <sup>b</sup>	667 (63) <sup>a</sup>
<b>Nitrogen</b>	12 (2) <sup>a</sup>	13 (3) <sup>a</sup>	8 (1) <sup>a</sup>	11 (2) <sup>a</sup>
<b>C/N Ratio</b>	66 <sup>a</sup>	31 <sup>b</sup>	56 <sup>a</sup>	61 <sup>a</sup>

Values followed by the different lower case letter are significantly different (0.05) between treatments. Standard errors are in parenthesis.

Mean values of C and N concentrations of crop residue biomass in all treatments for maize was 42.2% carbon and 0.66% nitrogen and for soybean was 44.8% carbon and 1.4% nitrogen.

Table adapted from Vachon and Oelbermann (2010)