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EFFECT OF METHOD OF TILLAGE ON LOSS OF CARBON FROM SOILS

A thesis presented in partial fulfilment of the

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Abstract

Soils represent the largest terrestrial carbon (C) pool. Different tillage practices have been shown to result in variable losses of soil C. Among these, No-tillage is regarded as an effective management practice for conserving SOC and reducing soil CO_2 emissions. Overseas research shows that No-tillage practice could reduce CO_2 emissions by approximately 3 Mg CO_2 ha⁻¹yr⁻¹.

Quantitative information comparing soil CO_2 emissions with No-tillage and conventional tillage is limited in New Zealand. Furthermore, little quantitative information is available on the effect of soil and climatic conditions in modifying these emissions. This Ph.D. study evaluated the potential for Cross Slot[®] No-tillage cultivation to reduce CO_2 emissions from cropped soils in New Zealand conditions.

A series of preliminary experiments were conducted to establish a suitable chamber method to collect and measure CO_2 emissions from soil. The alkali trap method was selected for use in traditionally cultivated agricultural soils. Another experiment was conducted to test whether pressure fluctuations caused by wind velocity differentially influence soil CO_2 emissions from conventionally and Cross Slot[®] No-tillage cultivated soils. Carbon dioxide emissions from conventionally cultivated soils rapidly equilibrated to the onset of lower (negative) pressure, whereas CO_2 emissions from No-tillage soils took longer to equilibrate.

Experiments on the potential savings of soil C with Cross Slot[®] No-tillage cultivation (NT) compared to simulated tillage, measured in the laboratory showed reduced (between 113 and 393 kg CO₂-C ha⁻¹) CO₂ losses in three out of four soils. This reduction in CO₂ losses was further verified with measurements made for one of the soils at a field site during autumn and summer seasons. Overall the results of field studies suggest that Cross Slot[®] No-tillage cultivation reduced ~3.0 Mg CO₂ ha⁻¹ compared with rotary tillage for combined autumn and summer sowings i.e. two cultivations.

A subsequent laboratory incubation study assessed CO_2 loss with different levels of residue addition to the four soils used in the previous laboratory and field experiments. A number of labile C fractions extracted from these soils were measured in an attempt to predict CO_2 losses. These did not show any relationship with the CO_2 respired during the incubation period. It was, therefore, not possible to develop a soil test to predict CO_2 losses using these extractions.

Modelling laboratory CO_2 respiration data for predicting the CO_2 losses from conventional and No-tillage soils was explored using relationships between short-term CO_2

respired and total CO_2 loss. The model developed from laboratory incubations was further improved with parameterising the soil temperature and moisture effects. The temperature and moisture sensitive model was used to predict the CO_2 emissions measured during the summer season. The model precisely predicted the amount of C lost from No-tillage soils but the amount predicted for rotary tilled soils was 30 per cent less than the amount of C that was lost in the field. Moreover, the model predicted C loss was higher for the No-tillage soils than the rotary tilled soils which was contradictory to the findings from the field study. Therefore, further work is required as the data obtained during this Ph.D. study was insufficient to provide, or develop a model that could be used to predict CO_2 loss from conventional and No-tillage cultivation in New Zealand soils.

DEDICATION

This work is dedicated to my wife Rupinder Kaur who gave me, more than strength, a reason to go on.

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Chapter-1

Introduction

1.1 Issue

Soil organic carbon (SOC) has long been studied due to its beneficial effect on physical, chemical and biological properties of soils, and is frequently reported as an indicator of soil quality and environmental sustainability (Ngwira *et al.* 2012). Rising environmental concerns about global warming, mainly due to increased CO₂ emissions, has increased awareness about the potential for SOC storage and greenhouse gas mitigation in agricultural soils (Ussiri and Lal 2009). Soil organic C is the largest active terrestrial C pool (>1550 Pg to 1m depth), with more than twice the C present in the atmospheric pool (Batjes 1996; Lal 2004), and is composed of both living and decomposed forms of plant tissues and soil microbes (Christopher *et al.* 2009).

Soil is a dynamic component of the global C cycle and may behave as a source or sink of CO_2 depending upon the management practices (Ogle *et al.* 2005; Franzluebbers 2005). Currently, the concentration of CO_2 in the atmosphere is 390.5 ppm and is increasing at the rate of 2 ppm per year (<u>http://cdiac.ornl.gov/pns/current_ghg. html</u>). Although fossil fuel consumption is the major cause of the increase in CO_2 concentration, agricultural activities have also been a significant contributor (Lal 2001; 2002).

Carbon dioxide is regarded as one of the most dynamic greenhouse gases related to agriculture (Sauerbeck 2001). Agricultural activities like deforestation, the conversion of natural to agricultural ecosystems, biomass burning, and excessive tillage (Lal 2004, 2007) lead to emissions of greenhouse gases, in particular CO₂, from soils. Historically, soils have lost about 40 to 90 Pg C (estimated value) to the atmosphere (Lal 2001; Smith 2008). Full cultivation has been regarded as a major cause of this C loss in cropping soils (Reicosky 2003), and tillage induced SOC loss in general, has been well documented (Huggins *et al.* 2007; Hermle *et al.* 2008). The depleted SOC in agricultural cropping soils can be restored by eliminating tillage, decreasing fallow periods and incorporating cover crops in the crop rotation cycle (Paustian *et al.* 2000; Jarecki and Lal 2003).

No-tillage is a conservation management practice in which there is least soil disturbance (nearly 90 per cent of the soil surface remaining untouched), and which conserves both SOC and soil nutrient stocks in comparison to conventional tillage. This practice has been adopted worldwide to combat soil erosion, as it leads to the development of a protective layer of crop residues at the soil surface (Puget and Lal 2005; Sa *et al.* 2001; Mazzoncini *et al.* 2011). The

adoption of No-tillage has increased over the past few decades, as it allows farmers to capture efficiencies in crop production - saving time, money and energy. Specialised openers are used for No-tillage as soil disturbance is limited to a spot where seed would be placed. Generally, No-tillage openers create "V", "U" and inverted "T" shaped slots (Baker and Saxton 2007). New Zealand scientists developed a unique inverted-T-shaped opener for No-tillage. The opener was first commercially available as the "Baker Boot" which underwent further development to become the current Cross Slot[®] opener. Several field and laboratory trials have shown that Cross Slot[®] openers cause less soil disturbance and better seedling emergence in comparison to other No-tillage openers (Baker and Saxton 2007). V-shaped slots created by double and triple discs are the most commonly used No-tillage openers worldwide, which makes No-tillage with inverted "T" openers in New Zealand different from other countries.

Overseas research, mostly conducted in North America, has compared the CO₂ emissions from cropping soils under No-tillage and conventional tillage systems, and shown that conversion to No-tillage seeding could reduce CO₂ emissions, currently estimated at 20.1 to 24.2 Mg CO₂ ha⁻¹ yr⁻¹, by up to 3 Mg CO₂ ha⁻¹ yr⁻¹ (Omonode *et al.* 2007; Ussiri and Lal 2009). Currently, about 6 per cent (1556 million hectares) of cultivated land in the world is under No-tillage, mostly in North and South America (Christopher *et al.* 2009; Mishra *et al.* 2010). Therefore there is the potential that further conversion to No-tillage systems and the adoption of appropriate management practices could enhance the role of soil as a major sink for C, and offset CO₂ emissions from fossil fuel combustion.

In New Zealand, approximately 1 million hectares are sown in agricultural crops annually. Quantitative information on potential changes in the soil CO_2 emissions with Notillage compared to conventional tillage in New Zealand systems is currently limited. Undoubtedly, soil and climatic conditions and the amount of plant residue at cultivation are the key factors that further modify soil emissions. However, the effect of these factors on CO_2 fluxes from conventionally and No-tilled cropping/pasture systems under a temperate environment, such as New Zealand, are not well documented, nor are they well quantified.

The Kyoto Protocol aims to reduce greenhouse gases by setting legally binding emission targets for member countries. The provision for assigning carbon credits for carbon conserved in forestry (article 3.3) and agricultural soils (article 3.4) under the Kyoto Protocol has made carbon a marketable commodity (Lal 2003). Carbon conserved in trees and soils can be traded as any farm produce, leading to an additional income for the farmer which can be used in soil restoration (Lal 2008).

Being a signatory to the Kyoto Protocol New Zealand has committed itself to reduce its greenhouse gas emissions to 1990 levels by 2012. To manage its Kyoto commitments, New Zealand introduced a financial market-based approach known as the emissions trading scheme (ETS). The scheme introduces a price on greenhouse gas emissions to provide an incentive to people to reduce greenhouse gas (GHG) emissions. The unit of trade is the New Zealand Unit (NZU) which is often referred to as carbon credits; and it is equivalent to one tonne of CO2-eq emissions. The New Zealand ETS covers six Kyoto Protocol managed greenhouse gases: carbon dioxide (CO_2), methane (CH_4), nitrous oxide (N_2O), hydro fluorocarbons (HFCs), per fluorocarbons (PFCs), and sulphur hexafluoride (SF6). So far New Zealand is the only country that has signalled its intent to include agricultural greenhouse gas emissions in its ETS but this has been delayed until 2015. However, mandatory monitoring and reporting for agricultural methane and nitrous oxide emissions has already started as of 1 January 2012. No commitment has been made for agricultural related carbon dioxide emissions to be included in carbon trading due to a lack of techniques to monitor the emissions of carbon dioxide related to agricultural activities, in particular tillage. No-tillage is seen as a promising strategy to conserve soil carbon leading to lower CO_2 emissions. There is also the possibility of inclusion of soil carbon losses or gains in the future revisions of the agricultural ETS scheme. Such inclusion would necessitate the development of a credible technique/tool for the farming community and policy makers in order to accurately determine and verify the reduction in soil C loss with No-tillage.

This project aims to quantify short term C loss from soils due to the decomposition of crop and/ or pasture residues after tillage, and attempts to provide a simple tool of either a soil test or decomposition simulation model to predict this C loss.

1.2 Research Objectives

- To determine CO₂ emissions with Cross Slot[®] No-tillage cultivation compared with full cultivation.
- To determine the soil textural influences on decomposition of crop residues to establish a relationship between soil texture, residue input and CO₂ emissions.
- ➤ To identify the labile C pools contributing to these losses and establish a relationship between the oxidisable C pool and CO₂ emissions.
- To develop a simple tool of either a soil test or decomposition simulation model that can quantify emissions reductions with Cross Slot[®] No-tillage to give the end-user the tools

necessary to implement advice on the use of Cross Slot[®] No-tillage to reduce the GHG footprint of arable farming in New Zealand.

1.3 Thesis structure

Significant research effort has been undertaken globally to understand the reductions in CO_2 emissions and increased soil C storage from No-tillage practice. However in New Zealand, research on the reduction of CO_2 emissions from agricultural soils using No-tillage practice, in comparison to conventional tillage, is limited. The single study conducted by Aslam *et al.* (2000) is not sufficient to devise strategies to conserve soil C and its loss as CO_2 .

This thesis (Figure 1.1) begins with an introductory Chapter-1 which covers the research background, conceptual framework, objectives and significance of this study in relation to New Zealand conditions. Chapter-2 presents an analysis of the available literature on various tillage practices and their role in conservation of soil C, factors affecting C loss and different methods for measuring soil CO₂ emissions. Various physical, chemical and biological techniques to quantify the labile soil C fractions and the effect of wind (Venturi effect) on soil CO₂ fluxes are reviewed. The review concludes with an overview of Notillage practices and highlights the need for a soil test to predict conservation of CO₂ with No-tillage. Chapter-3 provides a comparison between three chamber methods (static chamber alkali trap method, static chamber flux gradient method and dynamic chamber method i.e. EGM-1) to measure CO₂ emissions, which was used as the basis for choosing an appropriate method for the current study; this chapter also examines the venturi effect on CO₂ emissions from tilled and No-tilled soils under laboratory conditions. Chapter-4 quantitatively determines the loss of CO₂ from tilled soils under laboratory and field conditions in comparison to No-tilled soils. Chapter-5 quantifies the effect of soil texture and residue rate on decomposition of residues to CO₂ and attempts to provide a simple test for end users to use to quantify the reductions in emissions using No-tillage. Chapter-6 develops a model to predict CO₂ emissions in field conditions using laboratory measurements; and Chapter-7 provides a summary of results from the previous chapters along with the main conclusions derived from the research undertaken during this Ph.D., and the recommended direction for future research.



Figure.1.1: Thesis structure.

1.4 Significance of this study

This research will test the hypothesis that No-tillage cultivation is a management strategy for increasing soil C in cropping and pasture soils, and attempts to provide a mitigation tool to farmers and policy makers. This tool will allow quantitative determination of the influence of No-tillage practices in reducing CO_2 losses and maintaining soil C, in comparison to current conventional cultivation techniques practised in New Zealand. This study will provide a process-based understanding of the soil and climatic factors controlling CO_2 losses.

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Chapter-2

Review of literature

2.1 Soil carbon contribution to global warming

The atmospheric concentration of the three major terrestrial greenhouse gases, carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O) have increased markedly from 280 to 390.5 ppm (CO₂), 700 to 1750 ppb (CH₄) and 270 to 322 ppb (N₂O) (http://cdiac.ornl.gov/pns/current ghg.html), since the industrial revolution as a result of human activities. The majority of climate change scientists around the world agree that increasing concentrations of greenhouse gases in the earth's atmosphere are contributing to global warming and climate change (IPCC 2007). Concerns about global warming have increased interest in soil organic carbon (SOC) storage to counterbalance the rising atmospheric CO₂ levels (Sanaullah et al. 2011; Ussiri and Lal 2009). Soil organic C plays a pivotal role in soil functional processes, through its influence on most biological, physical and chemical processes in the soil. The decrease in SOC due to changes in land use and land management practices can critically affect the sustainability of our productive soil resource (Lal 2002). Historically, ploughing of natural soils resulted in the considerable loss of 55 Pg (billion tons of C) from the global SOC pool thereby converting a large fraction of SOC to CO₂ (Pacala and Socolow 2004). A loss of 55 Pg of SOC represents approximately 8.0% of the SOC (724 Pg) contained in the world's soils up to 0-30 cm depth (Batjes 1996); most of the SOC is lost from surface soils as ploughing does not go beyond 30 cm of soil depth (Lal 1991). Under some circumstances adoption of conservation tillage, growing cover crops, and using organic manures can restore SOC thus making soil a net sink.

No-tillage is regarded as an effective management practice in conserving SOC and reducing soil CO_2 emissions, and has so far received the most attention in North and South America. In New Zealand, about 20% of annual crop and pasture establishment is undertaken using No-tillage seeding (Personal Communication C J Baker 2009).

Given the importance of SOC to crop production, the impact of tillage on SOC storage has been well documented (Lal and Kimble 1997; West and Post 2002; Huggins *et al.* 2007; Ussiri and Lal 2009; Dalal *et al.* 2011). However, in New Zealand previous research on soil tillage measured the changes in soil physical properties as affected by No-tillage (Francis *et al.* 1987; Horne *et al.* 1992; Francis and Knight 1993; Hermawan and Cameron 1993; Ross *et al.* 2002), but little information is available on comparative effects of

No-tillage and conventional cultivation systems on CO_2 emissions in New Zealand cropping systems. Based on the North America's estimates of CO_2 savings and estimated areas of No-tillage in New Zealand, No-tillage has a potential of conserving approximately 2.4-3.0 million tonnes of CO_2 per year in New Zealand (see appendix 1). This amount of conserved CO_2 could reduce the contribution of agriculture to total greenhouse gas emissions by 7.0% per year in New Zealand (detailed explanation in Chapter-7). The possibility of inclusion of soil C losses or gains in any future revisions of the agricultural emissions trading scheme (ETS) has increased interest in the use of No-tillage practice.

The aim of this chapter is to review the role of traditional tillage practices in loss of soil C, various soil & climatic factors affecting soil C loss and labile C fractions contributing to this loss, and concludes by presenting the main research needs. Therefore this chapter first describes the range of current conventional tillage practices commonly used by farmers, and how these practices influence soil C dynamics. It then outlines various climatic & soil factors affecting soil C turnover and various methods of measuring soil CO_2 fluxes.

2.2 Common tillage practices

A tillage practice describes the nature and sequence of tillage operations used in preparing a seed bed for planting (Acquaah 2005). Tillage practices vary depending on soil characteristics, the crop to be grown, agro-ecological environment, and the socio-economic status of the farming community (Lal 1991; Koller 2003; Plaster 2009). Three basic goals of tillage are: 1) weed control 2) alteration of physical soil conditions, and 3) incorporation of the crop residues into the soil (Leij *et al.* 2002; Conant *et al.* 2007). Conventional tillage practices consist of primary cultivations to break the soil mass into a loose system of clods of mixed sizes followed by secondary cultivation for further pulverisation, repacking and smoothing of the soil surface, and is still used as the preferred tillage option (Koller 2003). Tillage is specifically designed to optimize edaphological conditions (soil water and temperature regimes, soil aeration, seed-soil contact, nutrient availability, porosity and pore size distribution and minimal incidence of pests) for seed germination, seedling establishment and crop growth (Lal 2004). In this practice natural soil structure is destroyed and a lot of energy is wasted (Blanco and Lal 2008).

2.2.1 Conventional tillage

Conventional tillage (CT), also called "intensive tillage", comprises a series of tillage operations to prepare a seed bed that leaves less than 15% of crop residues on the soil surface after planting the next crop (Blanco and Lal 2008). Conventional mouldboard ploughing followed by secondary tillage operations is the most common and preferred tillage practice in the world (Lal 2004; Acquaah 2005). During primary tillage operations, topsoil is ploughed to a depth of 15 to 36 cm and inverted, burying the vegetation and debris on the soil surface. Depth and time of primary tillage depends upon soil type, soil moisture, nature and amount of crop residues on soil surface, climatic conditions and crop season. Commonly used primary tillage implements are the mouldboard plough, disc plough, chisel plough, and powered rotary tiller. Secondary tillage operations are performed at shallow depths from 5 to 15 cm with an aim to improve seedbed level, moisture conservation, and increased soil pulverisation. Ploughing (primary tillage), disking and harrowing (secondary tillage) are carried out in sequence in order to produce a fine seedbed for seeding (McKyes 1985; Acquaah 2005; Plaster 2009).

2.2.2 Conservation tillage

Conservation tillage and conservation agriculture are collective umbrella terms given to No-tillage, minimum tillage and/or any other non-inversion soil tillage practice that has a conservation goal of some nature (Baker and Saxton 2007). Conservation tillage practices may retain at least 30% or more of the soil surface with crop residue after planting, improve soil structure, conserve soil moisture, reduce soil erosion, increase soil organic matter and improve environmental quality and agricultural sustainability (Lal 1991; Choudhary and Baker 1994; Acquaah 2005; Govaerts *et al.* 2009).

Different conservation tillage systems as described by Acquaah (2005); Baker and Saxton (2007); Blanco and Lal (2008) and Plaster (2009) are as follows:

1) Mulch tillage *(mulch-till)*: The soil is disturbed by tillage prior to seeding. A chisel plough, which loosens the soil but does not invert, is used as the primary tillage followed by light disking and seeding, leaving about 30-50 per cent of the crop residues. Weeds are controlled by a combination of cultivation and herbicides.

2) Strip tillage (*strip-till or zone-till*): The soil is not disturbed prior to seeding. About onefourth of the soil is tilled at the seeding time in a narrow strip ahead of the drill openers; so that seed is sown in the strip of tilled soil but the soil between the sown rows remain undisturbed, leaving behind approximately 50 per cent of crop residues. This tillage system is suitable for poorly drained soils. Weeds are controlled by a combination of cultivation and herbicides.

3) Ridge tillage (*ridge-till*): The soil is left undisturbed from harvest to planting except for fertilizer application. In ridge tillage system, 15 cm high ridges are formed by tillage during the second cultivation or after harvest in preparation for following year's crop. Seed is planted on 15 cm ridge tops with crop residues swept into shallow furrows. About two-thirds of crop residues remain after planting. Ridges may remain in place for several seasons or they might be reformed annually. Weeds are controlled by a combination of cultivation and herbicides.

4) Minimum tillage (*reduced tillage*): Any conservation tillage practice that minimizes the total number of primary and secondary tillage operations for seeding and leaves at least 30% residue cover after planting to control soil erosion and sustain crop production.

2.2.3 No-tillage

No-tillage (NT) (no-till, direct drilling, direct seeding, zero tillage, slot-till and slot planting) is a practice in which soil disturbance is limited only to a spot where seed would be placed. The soil is left undisturbed from harvest to planting. About 90 per cent of the soil surface is untouched i.e. minimal soil disturbance, which maximizes the benefits of conservation tillage. This practice commonly retains 70-95% of surface residues intact. Weeds are controlled primarily by herbicides.

No-tillage preserves soil organic matter (SOM) near the soil surface, reduces greenhouse gas production and stores the most soil C in comparison to other tillage systems (Lal 2004). However, No-tillage has some disadvantages as it results in a build-up of grass weeds, increases pest and disease problems, decreases crop yields due to poor germination and uneven crop growth, and requires specialised machinery which requires up-skilling. In addition, No-tillage can influence nitrous oxide (N₂O) emissions from soils (Ball *et al.* 2008). There are contradictory results regarding the impact of No-tillage on N₂O emissions. Higher (Ball *et al.* 1999; Beheydt *et al.* 2008) and lower (Chatskikh and Olesen 2007; Gregorich *et al.* 2008) N₂O emissions were observed from No-tillage soils than for tilled soils. Reviews conducted by Rochette (2008) and Powlson *et al.* (2012) suggest that soil aeration, which in turn is a combination of soil type and rainfall, and timing of nitrogen fertilizer application, are the key factors determining the N₂O emissions from soils.

Conventional mouldboard plough followed by secondary cultivation has been the traditional method for New Zealand farmers to establish crops and pasture (Hamilton-Manns

et al. 2002). Repeated cultivation leads to degradation of soil structure and loss of soil C as CO_2 due to decomposition of plant residues, exposing previously protected SOC in aggregates to micro-organisms (Jimenez and Lal 2006). Soil C has become important in recent times due to greenhouse gas emissions and climate change.

This review suggests that the No-tillage is receiving a lot of global attention as an alternative tillage practice to conserve SOC (Puget and Lal 2005). Large numbers of farmers are turning to No-tillage farming to stop the loss of valuable topsoil by erosion; to curb the run-off of sediment, fertilizer, and pesticides into rivers and lakes; to save time, money, and energy; and to conserve SOC (Lal et al. 2007). However, successful implementation and adoption of No-tillage strongly depends upon the farmers' knowledge of the technology, soil and climatic conditions of a given area.

2.3 Effect of tillage practices on soil carbon dynamics

2.3.1 Carbon storage

Tillage is an integral part of traditional agriculture and has strong impact on soil organic carbon (SOC) storage (Bayer *et al.* 2000; Reicosky 2003). Gregorich *et al.* (1998) found that most soils lost about 20-30% of SOC following 20 years of cultivation, and that maximum loss occurs within the first five years. Lal (2002) estimated that many soils in the Midwestern United States have lost 30-50% of SOC in about 50 years, which they contained prior to cultivation and which has been attributed to conventional cultivation practices. Recent studies recommended conversion from conventional to No-tillage as an efficient strategy to offset the stimulating effect of C emissions on global warming (Puget and Lal 2005; Luo *et al.* 2010).

West and Post (2002) used a global database of 67 long-term agricultural experiments, and analysed data of 93 paired conventional and No-tillage plots. They estimated that, on average, changing from conventional mouldboard plough tillage to No-tillage can restore SOC at the mean rate of 0.57 ± 0.14 Mg C ha⁻¹yr⁻¹ leading to new equilibrium SOC pool in 40-60 years. Similarly, with 96 comparisons of contrasting tillage systems in the south-eastern United States, Franzluebbers (2005) estimated that moving from conventional to No-tillage can restore SOC at the mean rate of 0.42 ± 0.46 Mg C ha⁻¹yr⁻¹. Both of these studies focussed on C change in surface soils (< 30cm) so this may simply be the artefact of sampling methodology (Blanco-Canqui and Lal 2008; Lal 2009). Baker *et al.* (2007) have shown examples where No-tillage had little effect on C storage particularly when deeper (>30cm) soil depths were considered i.e. when the whole soil profile (0-60/100

cm) is considered No-tillage plots contained similar or lower amounts of C in comparison to conventional mouldboard plough plots. Angers *et al.* (1997a) observed significantly higher SOC in conventionally tilled plots in comparison to No-tillage plots near the bottom of plough layer in several sites in eastern Canada.

The data from 24 studies are presented in Table 2.1, and shows the change in SOC at different soil depths due to No-tillage; the majority of these studies were conducted in North America. The duration of the experiments varied from 2 to 44 years and soil texture varied from loamy sand to clay. Studies included in this table differed in the tillage methods used such as chisel plough, disc plough, and mouldboard plough, and in crop, fertilizer and residue management practices.

The statistical significance of the results by depth (as reported in the original studies) are stated in Table 2.1 (last two columns). These results suggest that SOC under No-tillage was significantly greater in surface soil layers (0-5; 0-7.5; 0-10 cm) and significantly lower in deeper soil layers (20-30; 20-40; 30-50 cm) in comparison to conventional tillage. The 24 studies reviewed are presented as a scatter plot (Figure 2.1). Each data point represents the relative soil C change. The relative soil C change at each depth was calculated as:

(NT-CT)/CT..... (Eq. 2.1)

where, NT= SOC at the end of the experiment for NT soil (Mg C ha^{-1})

and CT= SOC at the end of the experiment for CT soil (Mg C ha^{-1})

Due to differences in the depth of soil sampling between studies (0-5; 0-10; 0-15; 0-20; 0-30; 0-40; 0-45; 0-60; 10-15; 10-20; 10-30; 15-30; 20-30; 30-40; 5-10; 5-15; 5-20 cm), each data point in the graph (Figure 2.2) represents the relative soil C change at the mean depth of the soil layer from which soil was collected (e.g. a soil sample collected from a 20-30 cm core has a mean depth of 25 cm). At each depth, the student's t value was used to test the hypothesis that the relative soil C change was zero (using SAS version 9.1). In general, relative soil C change data point values were >0 for 0-15 cm suggesting lower soil C stock under CT than NT at that depth (Figure 2.1). The relative soil C change was significantly greater than zero at 0-5, 0-10 and 0-15 cm depth intervals (p<0.05) i.e. the cultivation zone. The surface accumulation of SOC with NT practice irrespective of texture and duration of experiment has been well documented (West and Post 2002). With increase in depth intervals many data points' values were lower than 0, suggesting that average soil C stock could be greater under CT than NT at lower depths. However, at lower depth intervals (10-30, 15-30, 20-30, 30-40 cm) the relative soil C change values were not significantly different

from zero (p> 0.05). This suggested that the tillage effect on SOC is stratified with soil depth.

The presence of carbon in the form of crop residue on the soil surface is beneficial because surface residue accumulation has shown to reduce wind and water erosion, moderate soil temperature effects, conserve soil moisture, and provide an energy source for soil microorganisms (Lal and Kimble 1997; Govaerts *et al.* 2009). Franzluebbers (2002 a; b) suggested that the degree of change in SOC with depth can be used as an indicator of 'soil quality', because surface accumulation of organic C is essential to control erosion, water infiltration, and the conservation of nutrients. Furthermore it provides energy, substrates, and the biological diversity necessary to sustain numerous soil functions.

From the available data set, 13 comparison studies which measured SOC to at least 30 cm are presented in a scatter plot (Figure 2.2). Soil organic C stocks in 0-10, 10-20, 20-30 cm depths were summed and then relative soil C changes (Eq. 2.1) were calculated. In general, when the soil depth (0-30cm) was considered the soil C change with No-tillage showed a weak but significant correlation with duration of the experiment (Figure 2.2). Only a small proportion of variation (R^2 =0.32) was explained by this factor; however the relationship seems to be biased as the majority of studies have a short duration. Moreover, the important factor which affects the relative impacts of tillage practices on soil C change is crop residue inputs. No meta-analysis could be conducted on this factor due to insufficient data.

Results of NT and CT comparisons can be variable; Christopher *et al.* (2009) observed a change from -20.3 to 22.8 Mg ha⁻¹ in the carbon stock in 0-60cm depth from 12 paired NT and CT experiments varying in duration from 5-23 years in USA. Similarly, Angers and Eriksen-Hamel (2008) analysed data of 237 paired SOC depth measurements and suggested that No-tillage significantly increased the SOC in surface soil layers whereas conventional tillage has higher SOC near the bottom of the plough layer (23 cm). However higher SOC stocks at bottom of plough layer were not able to offset the gain under No-tillage in surface layers, resulting in higher SOC stocks under No-tillage.

In conclusion literature suggests significantly higher SOC stocks at the soil surface with No-tillage cultivation. Greater SOC content at deeper soil layers with conventional tillage practices did not completely nullify the gain at the soil surface with No-tillage; however, the view that No-tillage cultivation increases SOC over conventional tillage practices is questionable when sampling depth is considered. **Table 2.1:** Differences in soil organic carbon (SOC) between No-tillage (NT) and conventional plough tillage (CT) across a range of soils in different countries, most of the studies reported in this table used mouldboard plough except few and are arranged by publication year.

th significant (0.05) in SOC ound	NT <ct< th=""><th></th><th>20-30</th><th></th><th></th><th></th><th></th><th></th><th></th><th></th><th></th></ct<>		20-30								
Depth at whic differences (p< were f	NT>CT	0-10	0-5;5-10	0-10	0-10	0-10	0-10	0-10	0-15		
Dofound	vererence	Dalal <i>et al.</i> (2011)+¥	Lopez-Fando and Pardo (2011) ¥	Mazzoncini <i>et al.</i> (2011)¥	Sombrero and de Benito (2010)¶ ¥	Mishra <i>et al.</i> (2010) ¥	Mishra <i>et al.</i> (2010) ¥	Mishra et al.(2010) ¥	Ussiri and Lal (2009)¥	Hermle <i>et al.</i> (2008) ¥	Abid and Lal (2008)+¥
∆ SOC	(Mg C ha ⁻¹ yr ⁻¹)	-0.01	0.36	0.45	1.32	0.57	0.57	0.17	0.84	0.00	1.37
Layers	sampled	3	4	2	3	4	4	4	2	4	3
Depth of	cm) (cm)	0-30	0-30	0-30	0-30	0-40	0-40	0-40	0-30	0-40	0-30
Plough	depth (cm)	10	25	30	25	25	25	25	25	25	20
Duration	(years)	40	17	15	10	42	44	43	42	19	13
	Locauon	Queensland, Australia	Santa Olalla, Toledo, Spain	Pisa, Tuscany, Itlay	Castile-Leon, Spain	Ohio, USA	Ohio, USA	Ohio, USA	Ohio, USA	Tanikon, Switzerland	Ohio, USA
Coll toother	amitan	Clay	Loamy Sand	Loam	Clay loam	Silt loam	Silt loam	Silty clay loam	Silt loam	Sandy loam	Silt loam

	:	Duration	Plough	Depth of	Lavers	∆ SOC	ç	Depth at whic differences (p< were f	h significant 0.05) in SOC ound
texture	госаноп	(years)	depth (cm)	samping (cm)	sampled	(Mg C ha ⁻¹ yr ⁻¹)	Kelerence	NT>CT	NT <ct< td=""></ct<>
oam	Minnesota, USA	14	25	0-45	4	1.90	Huggins <i>et al.</i> (2007) ¥	0-7.5	
clay loam	Indiana, USA	28	25	0-100	9	0.36	Gal <i>et al.</i> (2007) ¥	0-5;5-15	30-50
am	Minnesota, USA	23	NA	0-45	7	0.00	Dolan <i>et al.</i> (2006)Ŭ		
am	Minnesota, USA	23	NA	0-45	7	-0.48	Dolan <i>et al.</i> (2006)Ř		
am	Illinois, USA	12	17	0-75	7	0.78	Olson <i>et al.</i> (2005)	0-15	
_	Iowa, USA	3	25	0-15	3	4.23	Al-Kaisi and Yin (2005)	0-5	
clay loam	Ohio, USA	8	ΝA	0-30	1	0.75	Puget and Lal (2005) ¥		
	Passo Fundo, Brazil	13	20	0-30	1	0.33	Sisti <i>et al.</i> (2004)¶ * ¥	0-30	
	Ponta Grossa, Brazil	22	20	0-40	5	0.86	Sa et al. (2001)	0-5	20-40
y clay	Rio Grande do Sul, Brazil	6	20	0-30	6	0.61	Bayer <i>et al.</i> (2000)¶ * ¥	0-30	

		Duration	Plough	Depth of	Layers	∆ SO C	Ę	Depth at whic differences (p< were f	h significant 0.05) in SOC ound
Soll texture	Госанон	(years)	depth (cm)	samping (cm)	sampled	(Mg C ha ⁻¹ yr ⁻¹)	Kelerence	NT>CT	NT <ct< th=""></ct<>
Silt loam	Palmerston North, New Zealand	2	20	0-10	1	3.20	Aslam <i>et al.</i> (2000)	0-10	
Loam	Nebraska, USA	26	NA	0-20	7	0.20	Six <i>et al.</i> (2000a)	0-5	
Silt loam	Ohio, USA	33	NA	0-20	2	0.13	Six et al. (2000a)	0-5	
Sandy loam	Michigan, USA	6	NA	0-20	2	0.24	Six et al. (2000a)		
Silty clay loam	Kentucky, USA	24	NA	0-20	7	0.26	Six et al. (2000a)	0-5	
Fine Sandy Ioam	Quebec, Canada	8	25	09-0	4	-1.00	Angers <i>et al.</i> (1997a)	0-10	20-40
Clay	Quebec, Canada	9	25	09-0	4	-3.38	Angers <i>et al.</i> (1997a)	0-10	20-40
Silty clay	Quebec, Canada	3	25	09-0	4	06.0	Angers <i>et al.</i> (1997a)	0-10	20-40
Sandy loam	Ontario, Canada	5	25	09-0	4	1.20	Angers <i>et al.</i> (1997a)	0-10	20-40
Sandy loam	Ontario, Canada	S	25	09-0	4	2.97	Angers <i>et al.</i> (1997a)	0-10	20-40

	0-5	Horne <i>et al.</i> (1992)	0.60	2	0-10	20	10	Palmerston North, New Zealand	Silt loam
	0-7.5	Francis and Knight (1993)	0.71	2	0-15	20	6	Lismore, Canterbury, New Zealand	Silt loam
	0-5	Costantini <i>et al.</i> (1996)	0.48	1	0-5	15	6	Cordoba, Argentina	Silty clay loam
		Franzluebbers and Arshad (1996)+	0.45	3	0-20	15	6	Alberta, Canada	Clay
20-40	0-10	Angers <i>et al.</i> (1997a)	80.0-	4	09-0	25	11	Ontario, Canada	Clay loam
 20-40	0-10	Angers <i>et al.</i> (1997a)	-0.77	4	09-0	25	4	Ontario, Canada	Sandy loam
 NT <ct< td=""><td>NT>CT</td><td></td><td>(Mg C ha⁻¹yr⁻¹)</td><td>sampled</td><td>sampung (cm)</td><td>depth (cm)</td><td>(years)</td><td>госацон</td><td>2011 LEXILLE</td></ct<>	NT>CT		(Mg C ha ⁻¹ yr ⁻¹)	sampled	sampung (cm)	depth (cm)	(years)	госацон	2011 LEXILLE
 ch significant <0.05) in SOC ound	Depth at whic differences (p< were f	Doformero	∆ SOC	Layers	Depth of	Plough	Duration		Coll touting

¶=mean of crop rotations +=used chisel plough *=used disc plough Ř= Crop residues retained Ř=Crop residues harvested NA=Information not available ¥=Studies used in figure 2.2 ASOC=SOC in No-tillage minus SOC in Conventional tillage divided by the duration of experiment


Figure.2.1: Relative fractional change in soil C (calculated as (NT-CT)/CT) under No-tillage (NT) compared to conventional tillage (CT) as function of soil depth in a range of studies conducted in Canada, USA, Australia, Europe and New Zealand (Data source given in Table 2.1).



Figure.2.2: Relative fractional change in soil C (calculated as (NT-CT)/CT) under No-tillage (NT) compared to conventional tillage (CT) in 0-30 cm soil depth from a range of studies conducted in Canada, USA, Australia, Europe and New Zealand (Data source given in Table 2.1).

The relative soil C change at each depth was calculated as (NT-CT)/CT where, NT= SOC at the end of the experiment for NT soil (Mg C ha⁻¹) and CT= SOC at the end of the experiment for CT soil (Mg C ha⁻¹)

2.3.2 Carbon dioxide losses

Soil CO₂ efflux plays a major role in the terrestrial C cycle (Lund *et al.* 1999). The annual flux of CO₂ from soils to the atmosphere at the global scale is estimated to an average 68 Pg C yr⁻¹ (Raich and Schlesinger 1992). At the field scale, soil CO₂ produced by microbial decomposition is stored in soil pores and emitted to the atmosphere either by diffusion due to soil-atmosphere concentration gradients or by diffusion plus mass flow (Alvaro-Fuentes *et al.* 2007; Reicosky *et al.* 2008). Both these processes interact and occur simultaneously in the field. The rate of loss of CO₂ from soil can be increased during tillage (Reicosky and Lindstrom 1993; Prior *et al.* 1997; Reicosky *et al.* 1997; Ellert and Janzen 1999; Alvarez *et al.* 2001).

Tillage effects on soil CO₂ fluxes are complex and highly variable. Short term CO₂ loss from soil due to tillage varied from 2.50 kg C ha⁻¹ d⁻¹ to 285.05 kg C ha⁻¹ d⁻¹ (Table 2.2). The average difference for short term studies (1-97 days) was 46.4 kg C ha⁻¹ d⁻¹. Increases in soil CO₂ flux have been observed hours or days after tillage operations (Rochette and Angers 1999). Reicosky and Lindstrom (1993) measured soil CO₂ fluxes under contrasting tillage systems (Mould board Plough and No-tillage), using a canopy chamber attached to an infrared gas analyser, and found high CO₂ fluxes from tilled soils immediately after tillage; even 19 days after the tillage event CO₂ flux rates were substantially higher for tilled than No-tilled soils. This increase was related to increased soil roughness and tillage intensity. In another study, where Reicosky *et al.* (1997) measured CO_2 fluxes from soils under different cropping systems with different soil inorganic N levels, there was no clear relationship between high CO₂ flux after tillage and N levels. They suggested that the increase in CO₂ flux was attributed to the release of CO₂ entrapped in soil pores rather than changes in microbial activity following tillage. Tillage accelerates SOM decomposition by the process of mixing of fresh residues with soil, modifying soil profile characteristics (e.g. aeration, moisture and temperature) and promoting soil microbial activity leading to high soil CO₂ emissions (Dao 1998; Rochette and Angers 1999; Ellert and Janzen 1999; Alvarez et al. 2001; Sanchez et al. 2002; Al-Kaisi and Yin 2005; Bauer et al. 2006; Omonode et al. 2007; Chatskikh and Olesen 2007; Alvaro-Fuentes et al. 2007; Akbolat et al. 2009; Ussiri and Lal 2009; Morell et al. 2010).

Long-term studies have shown conflicting results on the effects of No-tillage on soil CO_2 emissions. In Georgia, Hendrix *et al.* (1988) using a static alkali trap reported higher CO_2 fluxes from 5-6 year old No-till soils than from conventional tilled soil under sorghum

(Sorghum bicolor L.) and Soybean (Glycine Max L.) during a 17 month monitoring period. Although the cumulative difference was not clearly explained, there was a strong relationship between CO_2 fluxes and soil temperature for both tillage systems; however, no relationship could be found with soil moisture content. Similarly, Franzluebbers *et al.* (1995) and Aslam *et al.* (2000) observed that CO_2 fluxes in Texas and Palmerston North were similar or greater in No-tillage compared to conventional tillage systems during 1 and 2 year monitoring periods.

From the review it is concluded that CO_2 fluxes for a period after sowing are generally lower from No-tillage than conventional cultivation. However, a single study conducted in New Zealand found no significant differences in CO_2 emissions from Notillage relative to conventional tillage. As No-tillage is gaining momentum in New Zealand, monitoring CO_2 emissions from conventionally tilled and No-tilled soils could provide an improved scientific understanding and information to account for savings in CO_2 emissions from No-tillage cultivation for New Zealand cropping systems.

2.3.2.1 Effect of wind speed on CO₂ emissions from soil

Production of CO_2 in soil is regulated by microbial processes and transport takes place both by diffusion and mass flow (Widen and Lindroth 2003), where diffusion is controlled by the CO_2 concentration gradient and mass flow by pressure fluctuations at the soil surface (Xu *et al.* 2006). Wind has two effects. The first is turbulence at the soil surface which prevents a stagnant CO_2 rich layer forming. Lower CO_2 concentrations above the soil surface increase the concentration gradient. Wind due to its turbulent nature and interaction with various obstacles in the field like trees & complex terrain contributes significantly to surface pressure fluctuations which in turn affect soil CO_2 efflux (Takle *et al.* 2003; Xu *et al.* 2006). Pressure differences as low as 1 pa have been shown to cause differences in CO_2 flux measurements (Fang and Moncrieff 1996); using open dynamic chambers in field conditions, Kanemasu *et al.* (1974) showed that a static pressure deficit of 1 pa inside the chamber resulted in significant mass flow from soils. When wind blows a low pressure is created above the soil surface, a phenomenon known as the Venturi effect (Conen and Smith 1998), which pulls CO_2 rich air from the soil.

Baldocchi and Meyers (1991) observed an increase in CO_2 flux from the forest floor with increasing air turbulence using micro-metrological approaches. A seven fold increase in CO_2 flux was observed by Hanson *et al.* (1993) from the forest floor using a closed dynamic chamber when wind speed changes from near zero to 0.6 m sec⁻¹ by varying the speed of the chamber mixing fan. Using an empirical equation Longdoz *et al.* (2000) quantified the possible error in mean flux i.e. a depression of 1 pa resulted in overestimation of 50% of the CO₂ efflux using a closed dynamic chamber. Similarly, an over estimation of fluxes of up to 300% under strong wind conditions creating a depression of >2pa inside the chamber was observed by Suleau *et al.* (2009) from a forest floor. Takle *et al.* (2004) observed that pressure changes due to wind speed and direction penetrated in soil to depths of 45 to 60 cm and found that CO₂ fluxes at the surface to be approximately three times what would be expected from calculations based on diffusional flux rates. Reicosky *et al.* (2008) found higher CO₂ concentrations in No-tillage soils in comparison to ploughed soils and suggested that tillage induced changes in the soil bulk density and air porosity enabled wind speed to affect the gas exchange and resulted in a rapid decline in the CO₂ concentration. The review suggests that pressure differences created by wind speed inside the chambers or on the soil surface results in an increase in CO₂ fluxes from soils.

Table 2.2: Summary of the results of studies conducted on tillage induced CO2 fluxes.		
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	Table 2.2: Summary of the results of studies conducted on tillage induced CO ₂ fluxes.	

rbon content C kg ⁻¹ soil)	Location	Duration	CO ₂ -C loss during CT (kg ha ⁻¹ d ⁻¹)	CO ₂ -C loss during NT (kg ha ⁻¹ d ⁻¹)	$\Delta CO_2 - C \ loss \\ (kg \ ha^{-1} \ d^{-1})$	Reference
	USA	19 days	131.05	26.26	104.79	Reicosky and Lindstrom (1993)
	USA	1 day	115.00	18.20	96.80	Reicosky et al.(1997)
	USA	60 days	4.80	2.50	2.30	Dao (1998)
Са	nada	97 days	40.10	27.60	12.50	Rochette and Angers (1999)
Car	ıada	2 days	13.03	10.32	2.71	Ellert and Janzen (1999)
Arge	ntina	40 days	58.25	43.75	14.50	Alvarez et al. (2001)
N	SA	20 days	6.97	4.09	2.88	Al-Kaisi and Yin (2005)
n:	SA	51 days	285.05	41.24	243.81	Bauer <i>et al.</i> (2006)

Soil Texture	Carbon content	Location	Duration	CO ₂ -C loss during CT	CO ₂ -C loss during NT	$\Delta CO_2 - C \log S$	Reference
	(g C Kg SOII)			$(kg ha^{-1} d^{-1})$	$(kg ha^{-I} d^{-I})$	(kg na a)	5
	*U 9C8C						Alvaro-
Silt loam	z m ⁻² (0 20 m)	Spain	16 days	105.30	52.00	53.30	Fuentes
	g III (0-70 CIII)						et al. (2007)
							Chatskikh
Loamy Sand	20.3 (0-20 cm)	Denmark	91days	40.11	30.00	10.11	and Olesen
							(2007)
C:14 10000	10.0	Tudiout	16 1000	11 70	1 06	C0 0	Akbolat
	(0-30 cm)	ı uıkey	40 days	11./0	1.70	7.02	<i>et al.</i> (2009)
,	*5°L			1			Morell <i>et al.</i>
Loam	(0-28 cm)	Spain	8 days	9.50	6.30	3.20	(2010)
	35.3	New	Ţ			(Aslam <i>et al</i> .
Silt loam	(0-10 cm)	Zealand	l year	60.00	02.20	-2.50	(2000)
	9						Franzliiehhers
Silty clay loam	(0-20 cm)	USA	2 years	19.80	20.70	-0.90	et al. (1995)
Sandy clay	32.3*	USA	17 months	27.64	32.13	-4.49	Hendrix
loam	(0-20 cm)						<i>et al.</i> (1988)
$\Delta CO_2 - C \log S = CC$	D_2 -C loss from conv	entional tillage	minus CO ₂ -C los	s from No-tillage			

*indicates average carbon content from two contrasting tillage systems # indicates carbon content from three cropping systems

2.3.3 Carbon fractions

Six *et al.* (1998, 2000b, 2002a) concluded that a reduced rate of macro-aggregate turnover under No-tillage in comparison to conventional tillage led to the formation of stable micro-aggregates that increased C storage under No-tillage in the longer term. Many techniques based on chemical, physical and biological separation have been used to measure the size and turnover of SOM pools (von Lutzow *et al.* 2007). Physical fractionation has been widely used to study the turnover of SOM pools because of its direct relationship with SOM structure and function compared to chemical fractionation which is not useful in following the dynamics of SOM in natural and cultivated systems (Golchin *et al.* 1994a; Duxbury *et al.* 1989; Christensen 2001; Dou and Hons 2006). Physical fractionation of soils on the basis of density and/ or size of particles, allows the separation of uncomplexed/unprotected SOM (Christensen 2001; Six *et al.* 2002a).

Two most commonly isolated forms of physically uncomplexed/unprotected SOM are the light fraction (LF) and particulate organic matter (POM) (Gregorich *et al.* 2006). The LF is isolated by floatation in a high density liquid (density range 1.4-2.2 g cm⁻³) and POM by size (> 53 μ m), or by combination of both size and density fractionation procedures (Christensen 1992; Christensen 2001). Both LF and POM are composed of plant residues, but also contain microbial debris such as fungal hyphae and spores, and are considered to be sensitive indicators of the effects of tillage and cropping practices (Janzen *et al.* 1992; Cambardella and Elliott 1992; Golchin *et al.* 1994b; Boone 1994; Hassink 1995; Magid *et al.* 1997; Dominguez *et al.* 2009). Other measurable C fractions are the heavy fraction (HF) and mineral-associated fraction; these are organomineral complexed SOM, which is more stabilized than POM and LF therefore less sensitive to soil management (Alvaro-Fuentes *et al.* 2008).

Adoption of No-tillage can lead to the accumulation of POM through increases in macro-aggregation (Beare *et al.* 1994a, b; Six *et al.* 1998, 1999, 2000b; Chan 2008). Cambardella and Elliott (1992) measured the percentage of total organic C present as POM-C and found 39, 18, 19 and 25% in sod, bare fallow, stubble mulch and No-tillage after 20 years for a loam soil, respectively. They suggested that No-tillage reduced POM loss caused by tillage and aeration. After 13 years of continuous No-tillage and conventional mouldboard plough with grain sorghum [Sorghum bicolour (L.) Moench] and winter rye (Secale cereal L.), Beare *et al.* (1994b) found that No-tillage had 20% more POM-C than mouldboard plough. Tiessen and Stewart (1983) observed a 43% loss of POM after 4 years of continuous

tillage and concluded that POM is important in macro-aggregate formation, and it is this labile fraction that was affected by tillage.

Similar results have been observed in a number of studies (Bayer *et al.* 2001; Mrabet *et al.* 2001; Bayer *et al.* 2002; Bayer *et al.* 2006; Alvaro-Fuentes *et al.* 2008; Huang *et al.* 2010). Zhang *et al.* (2007) found that cultivation resulted in a decrease in LF-C and HF-C but the decrease in LF-C was considerably greater than the change in HF-C showing the LF-C as a fraction sensitive to management induced changes in organic carbon. Several researchers found similar results while studying the impact of No-tillage and conventional tillage on C dynamics (Yang and Kay 2001; Freixo *et al.* 2002; Roscoe and Buurman 2003; Soon *et al.* 2007; Tan *et al.* 2007; Murage *et al.* 2007; Zotarelli *et al.* 2007).

The review above suggests that physical fractionation is commonly used to study the turnover of the soil organic matter in cultivated and No-tillage soils. Soil C conserved by No-tillage in the light fraction (LF) and particulate organic matter (POM) may result only in temporary C storage because it could easily be lost through microbial decomposition with change in management practice.

2.4 Factors affecting soil carbon loss

Carbon dioxide emissions from soil to the atmosphere are the primary mechanism of C loss from soils and a pathway in the terrestrial C budget (Parkin and Kaspar 2003). The emission of CO_2 from soils is controlled by many factors such as the amount of crop residues added, C: N ratio of the added residues, crop rotations, total C content of the soil and added residues, labile soil-C fractions, tillage history of soil, fertilizer management, manure application, soil temperature, moisture and texture. In the research to follow, on the effect of tillage techniques, many of these factors will remain constant therefore this review considered soil temperature, moisture and texture, to understand their effects on the crop residue decomposition. In addition to these, soil tillage practices affect the production and release of CO_2 by influencing the soil physical conditions (Reicosky *et al.* 1997; Ball *et al.* 1999).

2.4.1 Soil temperature

Temperature is the key factor controlling soil CO_2 fluxes (Maljanen *et al.* 2002; Parkin and Kaspar 2003; Dilustro *et al.* 2005; von Lutzow and Kogel-Knabner 2009). Soil temperature depends upon atmospheric/air temperature and is influenced by presence/ absence of vegetation, water content and depth within soil (Aslam 1998). Soil temperature is the primary rate determinant of microbial processes and a central input to models simulating the effects of mineralisation of soil C pools on global warming (Parton et al. 1987). According to their temperature requirements micro-organisms are divided into three groups cryophiles, mesophiles and thermophiles - with their respective optimum temperatures being <20, 20 to 40, and >40°C (Luo and Zhou 2006). Soil microbial activity is considered to be negligible at temperatures <5°C (Wood 1989). There is an exponential increase in soil respiration with respect to temperature increases observed for biological systems (O'Connell 1990). Seasonal variations in the CO₂ emissions under long term field measurements are attributed to changes in soil temperature both under forest (Longdoz et al. 2000; Liang et al. 2004) and agricultural soils irrespective of the tillage systems (Aslam et al. 2000; Sanchez et al. 2002; Bauer et al. 2006; Omonode et al. 2007; Elder and Lal 2008; Alvaro-Fuentes et al. 2008; Ussiri and Lal 2009; Almagro et al. 2009) i.e. higher CO₂ emissions were measured in the summer season in comparison to the autumn/winter season due to higher temperatures in the summer season than autumn and winter season. Diurnal variations in CO₂ efflux are correlated to soil temperature (Jensen et al. 1996) with maximum CO2 fluxes measured during mid-afternoon and minimum during early morning (Akinremi et al. 1999; Shi et al. 2006).

Laboratory incubations provide the best estimates of temperature dependence of soil organic matter decomposition (Kirschbaum 1995). Field soil organic matter decomposition estimates are biased due to confounding factors of soil moisture and seasonal substrate availability, and the contribution of root respiration, which is not separated from that of soil microorganisms and fauna in the field studies (Reichstein *et al.* 2005). There is a range of problems associated with field measurements, but with laboratory measurements there are no such problems (Kirschbaum 2006). However, there is unease about accepting the results from controlled temperature under laboratory conditions for predicting responses under field conditions. Due to the complexity of the processes involved in soil organic matter decomposition both laboratory and field studies should be done simultaneously to test whether laboratory results can be extrapolated to the field.

The mathematical relationship between soil respiration and temperature is often described as the Q_{10} relationship/ Q_{10} factor or temperature coefficient, where the Q_{10} factor is the factor by which soil respiration increases for 10° C rise in temperature. Under controlled laboratory conditions, Sato and Seto (1999) found that the rate of CO₂ evolution from forest and arable soils increased exponentially with increase in the incubation

temperature from 4 to 40°C. The temperature coefficients (Q_{10}) for forest and arable soils were 2.0 and 1.9. A similar rise in CO₂ production was observed by Kechavarzi *et al.* (2010) with increase in temperature from 22 to 30°C under laboratory conditions for peat soils, the mean Q_{10} value was 3.2. Yuste *et al.* (2007) and Rey *et al.* (2008) in separate incubation studies on forest soils from U.S and Europe observed higher C mineralization rates with increase in temperature from 4-35°C, average Q_{10} values were 2.0 and 2.98 for U.S and European soils.

Kirschbaum (1995) reviewed many laboratory decomposition studies in which temperature varied and soil CO_2 evolution was measured in the absence of plants to assess the effect of temperature on organic C decomposition. He observed increased laboratory decomposition rates with increasing temperatures; Q_{10} values varied from 2 to 8, with lower Q_{10} values at higher temperatures. These laboratory decomposition experiments were used to further develop the temperature-response function to be used in soil organic matter simulation models (Kirschbaum 2000).

2.4.2 Soil moisture

Soil moisture is another important factor influencing soil respiration and hence CO_2 emissions (Rastogi et al. 2002). Water content in soil is essential for growth and activity of microbes, affecting the availability of substrates such as organic C, ammonium and nitrate that are essential for microbial functions (Weitz et al. 2001). Soil water content influences the concentration of O₂ and CO₂ in the soil profile (Aslam 1998) and soil temperature (Davidson et al. 1998; Akinremi et al. 1999). Respiration rates under high soil moisture contents are regulated by O₂ concentration (Linn and Doran 1984; Skopp et al. 1990). Bouma and Bryla (2000) suggested that high soil water content obstructs CO₂ production in soils by limiting both O₂ and CO₂ diffusion. Low soil water content, below a certain limit can hinder the soil microbial activity and root respiration because the aquatic habitat is reduced (Davidson et al. 1998; Xu and Qi 2001; Tang and Baldocchi 2005). Luo and Zhou (2006) suggested that optimum water content is near field capacity, where macro-pores are mostly air-filled, thus facilitating O₂ diffusion and micro-pores are mostly water-filled thus providing a habitat for bacteria and fungi and facilitating the diffusion of soluble substrates. In addition, low soil moisture content strongly limits the response of soil respiration to soil temperature.

At low soil moisture content, high temperature/warmer soil conditions decrease soil CO₂ fluxes (Reichstein *et al.* 2002; Xu *et al.* 2004; Tang and Baldocchi 2005; Yuste *et al.*

2007) due to a decrease in soil metabolic activity. A rapid increase in soil CO₂ fluxes within minutes after precipitation and rewetting of dry soils has been observed in a variety of ecosystems (Sponseller 2007; Jenerette et al. 2008; Almagro et al. 2009; Morell et al. 2010). A flush of microbial growth in the rewetted soil influences the soil CO₂ fluxes during the wetting and drying cycle. The rapid release of CO₂ immediately after precipitation is due to the infiltration of rainfall water into the soil pores, which replaces highly concentrated CO₂ air, resulting in degassing (Alvaro-Fuentes et al. 2008). Degassing is not considered to be soil respiration as it releases the stored CO_2 in soil from past microbial and root respiration. Microbial activity increases from several hours to a few days following precipitation, with an up to 40% increase in microbial cell counts observed within 2 days after precipitation (Saetre and Stark 2005). Adu and Oades (1978) argued that the rapid pulse of CO₂ is derived from microbial oxidation of labile soil organic C substrates derived from the physical disruption of soil aggregates due to the wetting and drying cycle, whereas Fierer and Schimel (2003) suggested that the pulse of CO₂ is generated by the rapid mineralization of highly enriched intracellular compounds as a response of the microbial biomass to the rapid increase in the soil water potentials.

2.4.3 Soil texture

Soil texture or size distribution of primary particles is the most influential quality of soil (Brady and Weil 2007). On the basis of the percentages of sand, silt and clay, 12 soil texture types are characterised globally (Eswaran 2003). Soil texture, particularly soil clay content, influences soil CO_2 emissions by affecting pore-size distribution and pore continuity, which in turn controls soil water availability, gas movement by diffusion and mass flow and microbial activity (Scott *et al.* 1996; McInerney and Bolger 2000; Thomsen *et al.* 2003; Galantini *et al.* 2004). The interaction of clay mineral type and clay content with soil C is very important in the process of formation of stable soil aggregates and soil structure (Six *et al.* 2000c).

Monitoring soil CO_2 fluxes from sandy and clayey forest soils for three years, Dilustro *et al.* (2005) found that soil moisture and respiration were significantly correlated in sandy soils but less so in clayey soils. This was because soil respiration was suppressed in sandy soils during warm and dry periods in comparison to fine textured clayey soils, which have more water holding capacity resulting in slow release of moisture during warm periods. Bouma and Bryla (2000) measured soil CO_2 fluxes from three different soil texture mixtures varying from 1 to 28% clay under laboratory conditions. They found that the rate of CO_2 release in sandy soil after rewetting returned to pre-wetting rates more quickly than in fine textured soils. They suggested that this may be due to lower water holding capacity of sandy soils resulting in the diffusion of CO_2 more freely through air-filled pores.

Soil C storage is positively correlated with the clay content of soil (Burke *et al.* 1989; Bird *et al.* 2003; Galantini *et al.* 2004; Tan *et al.* 2004; Plante *et al.* 2006; Homann *et al.* 2007; Kong *et al.* 2009). Within the same climatic area, with similar inputs, higher microbial biomass and more organic matter are expected in soils with high clay content than with low clay content (Hassink 1994; Muller and Hoper 2004). Because of the large specific surface area and negative charge, clay particles interact with organic matter to form stable clayorganic complexes that make organic matter less susceptible to decomposition (Plante *et al.* 2006).

Numerous studies conducted in the past to determine the influence of soil texture (clay content) on organic residue decomposition (Sorensen 1983; Ladd et al. 1981, 1985, 1992; Sharkov and Bukreeva 2004) have shown that in comparison to sandy soils, fine textured soils (with higher clay content) have lower rates of residue decomposition and retain higher proportions of residues by complexing with decomposition products, thereby reducing the losses of residue C from soil. Saggar et al. (1999) in a laboratory incubation study found that soil clay content and clay surface area played a significant role in controlling the decomposition of added C¹⁴ labelled glucose through stabilisation and protection of microbial biomass. Saggar et al. (1996), using two silt loam soils (clay content 16 and 24%) and two clay soils (clay content 56 and 60%) varying in mineralogy and surface area, found that, during the first 9 weeks of the decomposition in micro-lysimeters under field conditions, a greater portion of ¹⁴C-labelled ryegrass was retained in clay soils than in silt loam soils. After 5 years, the amount of remaining ¹⁴C was better correlated with the soil surface area than clay content alone. However, some other studies reported a weak or nonexistent relationship between soil carbon storage/ decomposition and clay content (Scott et al. 1996; Hassink 1997; Percival et al. 2000; Wang et al. 2010).

The review suggests that the loss of soil carbon as CO_2 is interactively affected by multiple factors and it is often difficult to separate these interactions. Carbon dioxide production in soil is directly controlled by soil temperature and moisture by their influence on microbial activity. The rate of CO_2 production and release depends upon temperature over a wide range of moisture contents but becomes responsive to moisture content as soil dries out below a certain limit. Soil texture particularly clay content influences CO_2 fluxes by modifying pore-size distribution and pore continuity, which in turn affects soil water availability and gas diffusion. Soil C content is positively correlated with clay content, as the clay content increases the capacity of the soil to protect organic matter against decomposition.

2.5 Methods for measuring carbon dioxide fluxes

Soil surface CO_2 flux measurements have been widely used to construct ecosystem C budgets (Norman *et al.* 1992) although accurate measurements of soil CO_2 flux are difficult to obtain (Lund *et al.* 1999). There are several methods available to measure CO_2 flux, including chamber and micrometeorological techniques. These methods vary in their accuracy, spatial variability and applicability. To date, there is no one recognised standard for accurate measurements (Rayment and Jarvis 1997). Therefore, methods are often chosen based on a trade-off between accuracy and feasibility.

2.5.1 Chamber methods

Chamber methods are the most commonly used method to measure CO_2 flux. Chamber methods directly measure CO_2 flux at the soil surface. Chamber size is a critical factor, which must be considered in designing measurement protocols i.e. sampling intervals. In addition either a chamber with adequate geometry or an increased number of chambers may be required to integrate the spatial variability across the soil surface (Rochette and Hutchinson 2005). To cope with the spatial variability, either a history of measurements is required or a reconnaissance study is required to design the chamber size and number.

2.5.1.1 Alkali trap approach

One chamber technique is the alkali trap approach. This is the oldest method of measuring soil CO₂ flux, which was introduced by Lundegardh (1927), and which is often known as static chambers, absorption chambers, or alkali trap chambers. This approach uses a chemical absorbent (alkali trap) to capture CO₂ released from the soil into the sealed chamber headspace. There are three common alkali traps: NaOH, KOH or soda lime (NaOH and Ca(OH)₂) (Buyanovsky *et al.*1986; Jensen *et al.* 1996; Janssens and Ceulemans 1998; Grogan 1998). The total mass of CO₂ trapped at the end of the absorption period is determined by titrating the alkali solution with dilute HCl.

The CO_2 flux (total amount of CO_2 trapped over the absorption period) is then calculated using the following calculation:

$$F = (C_{trap} - C_{blank})/t A.... (Eq. 2.2)$$

Where C_{trap} is the amount of CO₂ trapped; A is the surface area covered by the chamber; t is the absorption period; and the use of a blank (C_{blank}) accounts for any bias due to the contamination of the alkali solution (Kabwe *et al.* 2002).

Minderman and Vulto (1973) tested various alkali concentrations (0.25 - 2.0M) under field conditions and suggested that when sufficient alkali is used (approximately 30 ml) increasing the concentration from 0.25 to 2.0 M did not affect the CO₂ fluxes measured for a 24 hour interval. In order to achieve efficient CO₂ absorption, Kirita (1971) recommended a 13% alkali/chamber area ratio, depending upon chamber design and deployment conditions. Gupta and Singh (1977) suggested that absorption area has no significant effect on flux rates when at least 35% of the alkali remains unused after CO₂ absorption. However, the appropriate concentration of alkali to use will depend upon the expected CO₂ flux. The use of static chambers with alkali traps eliminates any convective or venturi effects that might happen during the field CO₂ flux measurements as molecular diffusion is the only major process of CO₂ exchange in this technique.

2.5.1.2 Flux gradient approach

The closed static chamber, flux gradient approach is a method in which there is no movement or replacement of air in the sealed chamber headspace. This means that the CO₂ gas concentration will increase steadily (Liang *et al.* 2004; Denmead 2008). The rate of increase is monitored by taking gas samples using polypropylene syringes which are fitted with 3-way stopcocks (Hutchinson and Mosier 1981). The samples are taken after the chambers are sealed, at different time intervals with the first sample taken at time zero (t0) (Pumpanen *et al.* 2003, 2004; Liang *et al.* 2004). The gas samples are then transferred to evacuated vials and analyzed using gas chromatography (GC) (Bekku *et al.* 1997; Saggar *et al.* 2004). Carbon dioxide flux (mg m⁻² hr⁻¹) is estimated from the measurements made at different time intervals. The sample of the ambient air taken just after closing the chamber (t0) is used as a reference for calculating CO₂ gas fluxes. During gas sampling by syringe, the chamber must be tightly sealed so that the gas extraction samples are not contaminated with ambient air. Secondly, size of the gas sample withdrawn should be small relative to the chamber headspace to get a reliable flux value with this technique. A large sample size may create negative pressure in the chamber leading to biased flux values.

The advantage of using static chambers is that they are easy to use and inexpensive; however they are highly labour intensive due to the large number of chambers required to account for temporal and spatial variability under field conditions. Long-term placement of the chambers may also lead to changes in the soil microclimate inside the chamber.

2.5.1.3 Closed dynamic chamber approach

In closed dynamic systems, air is circulated between the chamber and infrared gas analyser (IRGA). Once the chamber covers the soil surface, the CO_2 concentration in the chamber rises due to the release of CO_2 beneath the soil surface. The rate of CO_2 release is proportional to the soil CO_2 flux. Soil CO_2 flux is calculated by measuring the difference between CO_2 values at the start and end of the measurement period (Rochette *et al.* 1992, 1997; Rayment 2000). Closed dynamic chambers are advantageous as the CO_2 fluxes can be measured quickly, with minimal effect on enclosed soil surface temperature and moisture content. This system of flux measurements is very labour intensive when used manually, but can be automated and run for long periods of time to collect data, although this makes it costly to use. This method involves the insertion of a soil collar several centimetres into the soil to accommodate the chamber and to prevent CO_2 leakage out of the chamber (Hutchinson and Livingston 2001). A soil collar must be inserted in advance, a day before the CO_2 flux measurement to avoid the effect of the disturbance associated with the insertion of the collar. However, this is a concern for systems with long closure times and/or known pressure artefacts (Heinemeyer *et al.* 2011).

2.5.1.4 Open dynamic chamber approach

In dynamic open chamber systems, fresh air of a known concentration of CO_2 flows into the chamber while an equal volume of air is withdrawn. The air leaving the chamber has a higher concentration of CO_2 relative to the air entering the chamber, due to the CO_2 released from the soil surface. This concentration is measured by infrared gas analyzer (Kanemasu *et al.* 1974) and soil CO_2 flux is calculated using the flow rate and difference in CO_2 concentrations (Fang and Moncrieff 1996, 1998). This approach is also known as the steady state through flow chamber technique. It provides continuous CO_2 measurement with high accuracy due to its steady state. However, pressure differences inside and outside the chamber may lead to underestimation (at over pressurization or higher pressure) or overestimation (at under pressurization or lower pressure) of CO_2 fluxes (Pumpanen *et al.* 2001; Liang *et al.* 2004).

2.5.2 Micrometeorological techniques

Micrometeorological techniques are based on the concept that gas transport from the soil surface is accomplished by eddies that move air parcels from the soil to the measurement height. They also assume that vertical flux, measured at the reference level, is identical to the flux from the soil (Mosier 1990; Janssens *et al.* 2000; Baldocchi 2003).

Micrometeorological measurement methods include eddy covariance, flux gradient and mass balance calculations (Denmead 2008). Micrometeorological techniques have advantages over chamber systems as they achieve spatial and temporal integration of CO_2 flux, and do not modify the microenvironment of the soil surface (Dugas 1993; Janssens *et al.* 2000). They can also measure soil CO_2 flux over long periods and integrate large areas (Baldochhi 1997, 2003). However, successful applications of micrometeorological techniques are dependent on homogeneous upwind fetch and appropriate weather conditions (Baldocchi and Meyers 1991). In addition, the presence of vegetation between the soil and the measurement height may alter the measured fluxes (Norman *et al.* 1997) and a key limitation to its use is the high cost of instrumentation. It is unlikely that micrometeorological technique will replace widespread use of chamber methods as the most common means of measuring soil CO_2 efflux (Norman *et al.* 1997).

From the review it is concluded that different techniques have been used to measure soil CO_2 fluxes, but to date there is no universal consensus on one standard method. In the past, different methods have been compared throughout the world either against known CO_2 fluxes or directly on the soil surface (Rochette et al. 1992, 1997; Nay et al. 1994; Norman et al. 1997; Bekku et al. 1997; Jensen et al. 1996; Le Dantec et al. 1999; Janssens et al. 2000; Yim et al. 2002; Pumpanen et al. 2003, 2004; Liang et al. 2004). Several comparison studies suggest that each technique has its own strengths and weaknesses, depending on its application. Chambers are the most commonly used method of measuring CO_2 fluxes due to their simple operating principle, flexibility, portability and low cost. Literature suggests that we may expect chamber and micrometeorological techniques to be combined in the future, in order to achieve an accurate measurement of CO_2 fluxes with spatial and temporal integration.

2.6 Nature of soil organic carbon and separation techniques

Soil organic matter (SOM) is composed of plant, animal and microbial residues at various stages of decomposition and all intimately associated with soil inorganic components, and is an indicator of soil fertility and productivity. The content of organic

matter in soil changes with any change in land use and soil management practices, particularly labile C fractions. A range of techniques are used to measure and characterize soil organic C in relation to its dynamics under various management practices. Various chemical and physical techniques are discussed in the context of soil organic C turnover rates with change in soil management practices.

In a Ph.D. study at Massey University, Bhupinder-Pal Singh (2000) undertook an intensive review of the chemical and physical characterisation of SOM and discussed acid hydrolysis and oxidative techniques using potassium permanganate for labile-C extractions. He also discussed the density separation of soils. This review adds to the information provided in his thesis. In addition to potassium permanganate, the use of hydrogen peroxide, sodium hypochlorite and disodium peroxodisulphate are discussed in this review for oxidising the labile-C fractions. In addition to density separation, aggregate and particle size separations are discussed. Furthermore this review also provides the biological characterisation of SOM and various biological approaches used to estimate microbial biomass.

2.6.1 Chemical characterisation

Historically, chemical fractionation of SOM is based upon the differences in solubility properties of humic substances in alkaline and acid solutions, which yields three major fractions: humic acids (HA), fulvic acids (FA) and humin (Kononova 1966). Fulvic acids are soluble in both alkali and acid, humic acids are soluble in alkali but precipitated by acid, and humins are insoluble in both acid and alkali (Collins *et al.* 1997). Schulten and Schnitzer (1997) observed that humic and fulvic acids have similar chemical structures, however, in comparison to humic acids, fulvic acids have lower molecular weight and C content but higher oxygen content. Humins are condensed humic acids and consist of polysaccharides, phenolic or methoxy substituted aromatic structures and paraffinic structures (Hatcher *et al.* 1985; Hayes and Clapp 2001). They form strong complexes with clays and hydrous oxides and are not easily separated by alkali and acids (Schulten and Schnitzer 1997). Humic acids and humins have slow turnover rate, and are usually older (1130-1410 years) than the fulvic acids (50-550 years) (Cheng *et al.* 2007); however, Campbell *et al.* (1967) found a portion of humic acid and humin with short turnover rate (25-465 years) indicating incomplete separation with conventional SOM fractionation.

The total SOC can be differentiated into pools or fractions of varying stability (Zimmermann *et al.* 2007). Wander (2004) divided SOC into labile/active, slow/intermediate

and recalcitrant (passive/stable/inert) pools on the basis of biologically, physically and chemically regulated dynamics. However, different scientists have used different terms for these pools (Krull *et al.* 2003). A range of chemical fractionation techniques have been used to differentiate total SOC (Jagadamma *et al.* 2010) by acid hydrolysis (Leavitt *et al.* 1996; Collins *et al.* 2000; Rovira and Vallejo 2002; Cheng *et al.* 2007; Silveira *et al.* 2008) and chemical oxidation (Mikutta *et al.* 2005; Siregar *et al.* 2005; Helfrich *et al.* 2007; Jagadamma and Lal 2010). The aim of both fractionation techniques is to separate labile/active fractions from relatively stable/recalcitrant humic (fulvic acid, humic acid and humin) fractions (von Lutzow *et al.* 2007). However, there is no single appropriate chemical oxidation causes changes in SOM composition which mimics biodegradation i.e. natural microbial oxidation processes (Helfrich *et al.* 2007; Eusterhues *et al.* 2003) and is more efficient in isolating labile/young SOC fractions as discussed in the coming section.

2.6.2 Chemical fractionation techniques

Several oxidants like potassium permanganate (Loginow *et al.* 1987; Blair *et al.* 1995), hydrogen peroxide (H₂O₂) (Theng *et al.* 1999; Plante *et al.* 2004; Favilli *et al.* 2008), disodium peroxodisulphate (Na₂S₂O₈) (Eusterhues *et al.* 2003; Bruun *et al.* 2008) and sodium hypochlorite (NaOCl) (Siregar *et al.* 2005; Kleber *et al.* 2005; Zimmermann *et al.* 2007) are used in oxidising easily and strongly oxidisable C fractions. Potassium permanganate has been used to measure easily oxidisable/labile C fractions (Blair *et al.* 1995; Whitbread *et al.* 1998; Blair and Crocker 2000). Loginow *et al.* (1987) introduced the chemical separation of SOC based on its susceptibility to oxidation with various concentrations of KMnO₄ (33-333mM). The method is based on the concept that the oxidative action of KMnO₄ is similar to enzymatic breakdown of SOM which involves uptake of oxygen and liberation of CO₂ i.e. an oxidative process (Blair *et al.* 1995; Bell *et al.* 1998).

Lefroy *et al.* (1993) used three different concentrations (33, 167 and 333mM) of KMnO₄ to measure soil C fractions to relate which fraction is more sensitive to cropping and observed a decline in the C fractions oxidised by all the three different concentrations (33, 167 and 333mM) of KMnO₄ with long term cultivation, however, the major decline was observed in the fraction oxidised by 33mM KMnO₄.

Blair et al. (1995) standardised and simplified the original KMnO₄ oxidation technique introduced by Loginow et al. (1987) and suggested the use of one concentration of KMnO₄ (333mM) to distinguish labile soil C (oxidised by 333mM KMnO₄) from non-labile soil C (not oxidised by 333mM KMnO₄). Blair et al. (1995) compared cropped and uncropped soils and observed a major decline in the 333mM KMnO₄-oxidisable C fraction. The decline was proportionally greater than the decline in total-C. The KMnO₄ oxidises about 5-35.2% of the total C (Blair et al. 1995; Conteh et al. 1997; Bell et al. 1999; Haynes 2005; Vieira et al. 2007; Chen et al. 2009; Saha et al. 2011). Murage et al. (2000) and Xu et al. (2011) observed a relationship between KMnO₄ oxidisable C with water soluble, particulate and microbial biomass C in Kenyan and Chinese soils. However, Tirol-Padre and Ladha (2004) and Skjemstad et al. (2006) reported only a weak correlation between KMnO₄ oxidisable C with water soluble, particulate and microbial biomass C. They suggested that KMnO₄ rapidly oxidises less readily available organic compounds (like lignin) than water soluble carbohydrates and is not able to distinguish between labile and non-labile C and therefore should be referred to as permanganate-oxidisable-C (POC) when used as a parameter in characterizing soil.

Hydrogen peroxide was proposed for soil texture analysis by Robinson (1922) and is a widely used oxidant to remove soil C to date. Eusterhues *et al.* (2005) used hydrogen peroxide and found that the oxidation resistant C fraction was 500-3900 years older than C in the bulk soil suggesting that hydrogen peroxide preferably oxidises younger C fractions. About 42 to 97% of SOC can be oxidised by hydrogen peroxide (von Lutzow *et al.* 2007).

Sodium hypochlorite introduced by Anderson (1963) was used to remove soil organic matter for mineralogical analysis of clays. Sodium hypochlorite removes about 26-96% of SOC in soils (Kleber *et al.* 2005; Mikutta *et al.* 2005; Siregar *et al.* 2005). An increase in radiocarbon ages from 75-6350 years after treatment with sodium hypochlorite compared to bulk soils was observed by Kleber *et al.* (2005).

Disodium peroxodisulphate was proposed by Meier and Menegatti (1997) to remove SOM and have a higher efficiency than hydrogen peroxide and sodium hypochlorite in oxidising soil C. Between 16-99% of SOC was oxidised by disodium peroxodisulphate (Eusterhues *et al.* 2003). The C fraction resistant to hydrogen peroxide, sodium hypochlorite and disodium peroxodisulphate treatment is aliphatic in nature (Leifeld and Kogel-Knabner 2001; Cuypers *et al.* 2002; Jagadamma *et al.* 2010). A wide range of per cent SOC that can be oxidised by hydrogen peroxide, sodium hypochlorite and disodium peroxodisulphate in soils was due to differences in the soil's clay mineralogy (Kleber *et al.* 2005). From the review it is concluded that chemical fractionation is very useful in understanding structure and composition of SOM. Chemical fractionation techniques help in measuring the relative distribution of labile and stable organic C fractions to monitor the effect of treatments. However, chemical fractionations have several limitations as chemical extracts used may modify SOM structure. Moreover, chemical extracts may isolate that fraction which may be physically protected from microbes and not available for decomposition, which cannot be termed as a labile fraction. Due to the complex nature of SOM, chemical fractionation may have limited success.

2.6.3 Physical characterisation

Commonly chemical extractions are used to differentiate SOM fractions based on their respective solubilities in various extractants (acids and bases) and considers interaction between the SOM and soil mineral particles, chemical bonding within SOM but not the three dimensional arrangement of organo-mineral complexes (Six et al. 2000d; Olk and Gregorich 2006). Therefore, they are not very useful in identifying SOM fractions that reduce with intensive management (Collins et al. 1997) because turnover rates of different SOM fractions are partly controlled by microbial decomposition and are affected by both the chemical and physical nature of SOM (Bhupinderpal-Singh 2000). The physical and/or physicochemical binding between SOM and soil minerals is the mechanism supposed to be responsible for protection/stability and turnover rate of SOM fractions (Six et al. 2002b; Chan 2008). To study the C turnover rates, physical fractionation methods which are less destructive than chemical fractionation are employed (Christensen 1992) and results obtained from physical fractionation techniques are more strongly related to the structure and function of SOM in-situ (Golchin et al. 1994a). Physical fractionation methods are based on the premise that the association of primary soil particles with SOC and their spatial arrangement plays an important role in the dynamics of SOM (Gregorich et al. 2006; von Lutzow et al. 2007). Physical fractionation employs a number of different methods, each designed for a specific purpose, application of which, involves combinations of mechanical, chemical and ultrasonic dispersion for size fractionation (wet and dry sieving, slaking) and/or using heavy liquids (1.4-2.4 g cm⁻³) for density fractionation. Detailed reviews on methodology have been published by Elliott and Cambardella (1991); Christensen (1992) and Collins et al. (1997). The objective of every fractionation technique is to avoid chemical changes in SOM during the fractionation step and to provide a clear differentiation between separated SOM fractions. The recovery of SOM and soil material depends upon the complexity of the physical fractionation. More complex the fractionation procedure, more are the chances of errors and losses of SOM (Abdul-Kader 2006). Different methods of physical fractionation are discussed in detail in the coming section.

2.6.4 Physical fractionation techniques

2.6.4.1 Aggregate fractions

Aggregates are also referred to as secondary organo-mineral complexes and reflect the degree of aggregation of primary particles (Christensen 2001). The formation and dynamics of soil aggregates has been reviewed in detail by Golchin *et al.* (1997); Wander (2004); Six et al. (2004); Yadav and Malanson (2007). The aggregate hierarchy concept introduced by Tisdall and Oades (1982) described how primary mineral particles (<20 µm) are bound together with microbial products, root exudates and polyvalent cations into microaggregates (20-250 µm). These micro-aggregates are bound into macro-aggregates (>250 µm) by transient (microbial and plant derived polysaccharides) and temporary (roots and fungal hyphae) binding agents. The hierarchy concept by Tisdall and Oades (1982) was modified by Oades (1984), who suggested that 'roots and hyphae which hold together the macro-aggregate, forms the nucleus for micro-aggregate formation in the centre of the macro-aggregate'. As roots and hyphae are temporary binding agents they crumble into fragments, get coated with mucilage produced during the decomposition process and became encrusted with clay particles and microbial products resulting in the formation of microaggregates within a macro-aggregate. Further studies by Golchin et al. (1994a); Beare et al. (1994b); Jastrow et al. (1996) supported the concept of the formation of micro-aggregates within macro-aggregates. In a field incubation study, Angers et al. (1997b) traced C and N in macro- and micro-aggregates during decomposition of C¹³ N¹⁵-labelled wheat straw and found reorganization of C¹³ from macro-aggregates to micro-aggregates with time. Carbon distribution in micro-aggregates happens only after distribution at the macro-aggregate level, indicating that micro-aggregate OM is older and in a much more stabilized state than macroaggregate OM.

Aggregate fractionation is based on the separation of free-OM and occluded (intraaggregate) OM that is trapped within secondary organo-mineral complexes (Six *et al.*1998, 1999). Aggregate fractions are isolated by wet or dry sieving and slaking (Elliott and Cambardella 1991). Dry sieving checks the stability of aggregates against mechanical disruption when dry soil is rotary sieved and wet sieving includes the effect of the wetting process also (Christensen 2001). Slaking is the breakup of large aggregates into smaller ones due to build-up of internal pressure during rapid wetting of soil (Six *et al.* 1999). Six *et al.* (2000b) proposed a fractionation technique of wet sieving and slaking to completely break up macro-aggregates while minimizing the breakdown of the released micro-aggregates (53-250 μ m). Six *et al.* (2000a) studied the effect of conventional and No-tillage on aggregation and aggregate associated-C and found that increased C under No-tillage was due to a reduced turnover rate of macro-aggregates in comparison to conventional tillage. Denef *et al.* (2004) suggested that more than 90% of the difference in SOC between conventional and No-tillage was because of a difference in micro-aggregate associated C. Various studies have shown that turnover times were about 100-300 years for OM in micro-aggregates (< 250 μ m) and 15-50 years for OM stored in macro-aggregates (> 250 μ m) (Puget *et al.* 2000; Six *et al.* 2002b; Yamashita *et al.* 2006).

2.6.4.2 Particle size fractions

Particle size fractionation is based on the concept that SOM associated with particles of different size and mineralogical composition, differs in structure and function, and therefore plays different roles in SOM turnover. Particle size fractionation can be applied to whole soil or to heavy fractions following density fractionation (Christensen 1992).

Particle size fractionation divides SOM into size classes by sieving (dry/wet depending upon the purpose of fractionation) and sedimentation following dispersion (Six *et al.* 1998; Wander 2004; Sleutel *et al.* 2006). Size fractionation separates soil into sand, silt and clay sized fractions related to texture of soil. Sand particles show weak bonding to SOM whereas clay particles provide large surface area and numerous reactive sites where SOM can be sorbed by strong ligand exchange and polyvalent cation bridges (Sposito *et al.* 1999). Based on the sorption stabilization mechanism, SOM in the sand fraction is more labile (active pool) than silt and clay fractions (intermediate and passive pool) (Evans *et al.* 2001; Chenu and Plante 2006).

Various degrees of dispersion are used to break down macro- and micro-aggregates to separate uncomplexed OM and different size organomineral complexes (Christensen 2001; Gregorich *et al.* 2006). Sonication and shaking are commonly used dispersion techniques (Elliott and Cambardella 1991). Sonication produces vibration energy to the soil suspension causing cavitation, which disrupts bonding agents. Presently, there are no standard protocols for sonication, and it has potential to redistribute organic matter among size/density fractions (Collins *et al.* 1997). Shaking is a more gentle alternative dispersion

method to sonication (Elliott and Cambardella 1991). However, Christensen (1992) observed that high-speed shakers may cause abrasion of particles and simple shaking in water may not provide complete dispersion even after prolonged treatment periods.

Chemical dispersants and prolonged shaking times can improve soil dispersion but chemically assisted dispersion can introduce unintended changes in SOM structure and should be avoided unless the action of the dispersant is specific and is well documented (Elliott and Cambardella 1991; Christensen 1992). Sieving, sedimentation and centrifugation are collectively used to determine particle size fractions (von Lutzow *et al.* 2007). Generally, about 50-75% of the total SOM is associated with the clay sized fraction (< 2 μ m), about 20-40% with the silt sized fraction (2-20 μ m) and < 10% with the sand sized fraction (> 20 μ m) (Christensen 2001).

2.6.4.3 Density fractions

Density fractionation techniques can separate SOM into two discrete fractions representing different stages of decomposition (Gregorich et al. 2006; Crow et al. 2007). Density fractionation is applied to the whole soil to separate the SOM fraction which is not firmly attached to soil minerals i.e. light fraction (LF) from organo-mineral complexes or the heavy fraction (HF) with floatation in inorganic salt solutions with a specific density varying from 1.4 to 2.2 g cm⁻³ and limited sample dispersion (Strickland and Sollins 1987; Christensen 1992; Crow et al. 2007). The light fraction of SOM has density <1.4 to 2.0 g cm⁻ ³ (lower than that of soil minerals), is mineral free, and composed of partially decomposed plant and animal residues with a wider C: N ratio (Janzen et al. 1992; Boone 1994; Magid and Kjaergaard 2001). The light fraction is regarded as highly labile with a rapid turnover rate (Boone 1994; Gregorich et al. 2009) due to the labile nature of its constituents i.e. easily decomposable carbohydrates (Six et al. 2002b). The heavy fraction of SOM is composed of highly processed decomposed products with a narrow C: N ratio adsorbed on to the surface of silt & clay particles (Six et al. 2002b). The heavy fraction has a slower turnover rate and higher specific density > 2.0 g cm⁻³ due to strong close association with soil minerals (Meijboom et al. 1995; Olk and Gregorich 2006; Crow et al. 2007).

Prior to density fractionation, various degrees of sample dispersion are used to break soil aggregates to separate uncomplexed OM and organo-mineral complexes (Christensen 1992). However, compared to particle size based fractionation, density based methods rely on less vigorous dispersions or on shaking (Wander 2004). Historically, heavy organic liquids have been used for density fractionation i.e. tetrabromoethane $C_2H_2Br_4$ (2.96 g cm⁻³),

bromoform CHBr₃ (2.88 g cm⁻³), tetrachloromethane CCl₄ (1.59 g cm⁻³) but aqueous solutions of inorganic salts i.e. sodium Iodide NaI and sodium polytugstate Na₆ ($H_2W_{12}O_{40}$) have gained popularity as previously organic liquids were halogenated hydrocarbons that were highly toxic to humans (Christensen 1992; Crow et al. 2007). The light fraction has been suggested as the indicator of changes in labile organic C as affected by tillage, cropping practices and addition of crop residues (Janzen et al. 1992; Boone 1994; Magid et al. 1997; Six et al. 2002b; Liang et al. 2003; Soon et al. 2007; Zhang et al. 2007). Golchin et al. (1994a; b) modified the basic two-fraction method and separated LF located between soil aggregates (inter aggregate/ free LF) from protected intra-aggregate (occluded LF) by sonication and floatation and suggested that slow turnover of occluded LF compared to free LF is due to difference in stability. Murage et al. (2007); and Gregorich et al. (2009), while studying the effect of tillage practices on SOC fractions, observed higher turnover of C in free LF than occluded LF. In order to relate SOC fractions to stable soil aggregates to better understand the SOC dynamics, Six et al. (1998; 1999; 2000a) developed a complex technique using size and density fractionation to isolate organic C within soil aggregates having different roles in nutrient cycling and stabilization mechanisms.

The density of plant residues changes during decomposition due to breakdown of vascular structure and loss of entrapped air. Therefore the density separation used to separate LF-C is also used to distinguish the nature and extent of organic matter decomposition (Gregorich *et al.* 2006) as a part of labile organic matter in soil originates from live and dead root & plant residues (Bhupinderpal-Singh *et al.* 2005). However, the ability of density separation techniques to recover decomposing residues is affected by the size of the residues and their interaction with mineral surfaces (Magid *et al.* 1996; Bhupinderpal-Singh *et al.* 2005). In order to overcome this problem, Magid *et al.* (2010) and Bhupinderpal-Singh *et al.* (2009) suggested using a combined size-density procedure to recover plant residues from soil rather than using a density procedure alone.

The review suggests that physical separation techniques provide a differentiation between labile/active and passive soil carbon fractions. Particulate organic matter and/or the light fraction are representative of the labile pool whereas the mineral or heavy fraction is a mixture of intermediate and passive pools. Therefore, using density or combined size-density procedures would be helpful to identify the fate of the light fraction like plant or root residues during their decomposition.

2.6.5 Biological characterisation

Soil microbial biomass is the living or/ active component of soil organic matter and is defined as the mass of soil micro-organisms $< 5000 \text{ } \mu\text{m}^3$ in volume, which excludes soil animals and roots (Brookes et al. 2008; de Araujo 2010). Microbial biomass plays a crucial role in C, N, P and S transformations; acts as both a source and sink for nutrients; and contributes to soil structure (Lagomarsino *et al.* 2009). It is a biologically meaningful, easily measurable and management sensitive fraction (de Araujo and de Melo 2010). The turnover time of microbial biomass varies from 0.5 to 5 years in comparison to >20 years for total SOM (Dalal 1998; Nyamadzawo et al. 2009). Based on dynamics of soil microbial biomass content long-term trends in SOM can be predicted (Truu et al. 2008). Changes in microbial biomass content due to change in land use practices can be used as early predictors of changes in SOM (Powlson et al. 1987; Yeates and Saggar 1998; Saggar et al. 2001; Melero et al. 2006; Brookes et al. 2008). Soil microbial biomass is affected by input of organic residues as high amounts of organic inputs result in higher microbial biomass (Peacock et al. 2001; de Araujo and de Melo 2010). Soils in No-tillage cropping systems have higher microbial biomass in comparison to soil under conventional tillage perhaps due to higher crop residue input and increased SOC content, or, higher soil moisture contents (Aslam et al. 1999; Alvarez and Alvarez 2000; Doyle et al. 2004; Nyamadzawo et al. 2009). During a field trial comparing conventional and organic farming practices, Melero et al. (2006) observed significantly higher microbial biomass under organic than conventional management practices due to higher input of residues. Similar results were observed by Tu et al. (2006); Araujo et al. (2008) and Okur et al. (2009) while comparing conventional and organic management practices. In general, microbial biomass C ranges between 0.8 and 7.0% of SOC (Wardle 1992). In agricultural top soils, microbial biomass C ranges between 0.3 and 4.0% depending on the soil texture and tillage practices (von Lutzow et al. 2007). There are different techniques to determine soil microbial biomass C which are listed in the upcoming section.

2.6.6 Biological approaches

Commonly used techniques to estimate soil microbial biomass are:

- * Direct microscopic counting (Jenkinson *et al.* 1976c)
- * Chloroform fumigation-Incubation (FI) (Jenkinson and Powlson 1976b)
- * Chloroform fumigation-Extraction (FE) (Vance *et al.* 1987)
- * Substrate induced respiration (Anderson and Domsch 1978; West and Sparling 1986)

- * Adenosine triphosphate (ATP) analyses (Jenkinson and Oades 1979; Jenkinson *et al.* 1979; Tate and Jenkinson 1982)
- * Phospholipids fatty acids (PLFA) (Frostegard and Baath 1996)
- * Microwave irradiation (MW) (Islam and Weil 1998)

Detailed descriptions of the methods and their advantages and limitations to estimate soil microbial biomass are given by Jenkinson (1988); Horwath and Paul (1994); Martens (1995); Dalal (1998); Brookes (2001); de Araujo (2010); Gonzalez-Quinones *et al.* (2011). Basic soil properties such as soil pH, soil moisture and organic matter level need to be considered while selecting a method to estimate soil microbial biomass. Among all the methods available, chloroform fumigation is the most commonly used method (de Araujo 2010; Gonzalez-Quinones *et al.* 2011) to estimate soil microbial biomass. In the chloroform fumigation method the fumigated soil is either incubated (Jenkinson and Powlson 1976b) or extracted (Vance *et al.* 1987). The chloroform fumigation extraction technique has been successfully used to determine N (Brookes *et al.* 1985), P (Hedley and Stewart 1982) and S (Saggar *et al.* 1981) contents of soil microbial biomass.

Literature suggests that soil microbial biomass is an integral component of SOM which regulates the storage and transformation of nutrients. Due to short turnover times (< 5 years) it is regarded as a major component of the active/labile C pool. It responds quickly to changes in land use practices, disturbance or restoration. Therefore, microbial biomass can be used in monitoring early changes in soil due to soil tillage, restoration and application of organic manures to the soil. However, the time consuming nature of microbial biomass measurement limits its usage in routine analysis. Moreover, soil microbial biomass values are affected by land use management and climate but it is difficult to compare these values across land uses, climate and soil types as no threshold values for comparison are currently available.

2.7 Conclusions

With increasing concerns about global climate change, the storage and dynamics of SOC under different land uses has received more attention due to the significant potential of soils to act as a source and sink for atmospheric CO_2 . Ploughing and conventional seedbed preparation results in loss of SOC as CO_2 from agricultural soils. Several researchers observed that adoption of No-tillage can significantly increase the SOC storage and reduce atmospheric CO_2 concentrations. However, positive change in SOC caused by adoption of

No-tillage depends upon the soil and climatic conditions of the area. Soil texture, temperature and moisture are key factors controlling CO_2 losses. Short term soil CO_2 fluxes may also affected by surface pressure fluctuations caused by wind speed and this needs to be accounted for when comparing tilled and untilled soils.

To date there is no standard technique to measure CO_2 emissions. Different techniques have been used to measure soil CO_2 emissions but chamber methods are the most common technique due to low input cost and direct measurement of CO_2 fluxes.

A wide range of physical, chemical and biological techniques have been applied to separate soil organic C into fractions, in an attempt to identify fractions with rapid and slow turnover rates. Several studies have been conducted to measure and characterize the young, labile fractions such as microbial biomass, light fraction, particulate organic C and the easily oxidisable fraction to relate their dynamics with various management practices. Currently, there is no standard fractionation technique to characterise a labile C fraction and develop a single relationship for a range of soils and relate it to the CO_2 loss after tillage. Further research on the characterisation of labile C fractions relating to CO_2 losses would be useful to evaluate the effect of land use practices.

Research on the reduction of CO_2 emissions with No-tillage practice in comparison to conventional tillage from agricultural soils in New Zealand is very limited. A single study conducted by Aslam *et al.* (2000) might not be sufficient to devise strategies to conserve soil C and reduce its loss as CO_2 . Therefore, monitoring CO_2 emissions from different tillage systems is important to understand the reductions in CO_2 emissions from No-tillage practice, which has been covered in this thesis.

2.8 References

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Chapter-3

Comparisons of methods to measure carbon dioxide emissions from soils and wind effects on soil carbon dioxide fluxes under controlled laboratory conditions

3.1 Introduction

There are several techniques i.e. micrometeorological, and the use of static and dynamic chambers methods for measuring CO_2 efflux, with large differences in accuracy, spatial and temporal variability, and applicability (Lund *et al.* 1999). The choice of a specific technique is often a trade off between requirements (accuracy) and feasibility (applicability and cost). To date, the use of chambers remains the most commonly used method for measuring the emissions of greenhouse gases (Denmead 2008). According to Livingston and Hutchinson (1995) chambers are grouped into steady-state (SS) and non-steady-state (NSS) chambers, and can be further divided into non-flow-through (NFT) and flow-through (FT), depending upon the air circulation through the chamber.

Based on the above classification the different chamber based approaches used for measurement of CO₂ fluxes are: (1) non-flow-through-non-steady-state chamber (NFT-NSS) also known as the static chamber flux gradient method in which air samples are taken at regular intervals by syringe and subsequently analyzed by gas chromatography, (2) flow-through-non-steady-state chamber (FT-NSS) also known as a closed dynamic chamber, (3) flow-through-steady-state chamber (FT-SS) also known as an open dynamic chamber and (4) non-flow-through-steady-state chamber (NFT-SS) also known as absorption chambers or alkali trap chambers as stated by Rochette and Hutchinson (2005). In steady-state chambers CO₂ concentration inside the chamber remains constant and ambient and CO₂ flux is calculated using the difference in CO₂ concentration between air leaving and entering the chamber. In non-steady-state chambers, the rate of change in CO₂ concentration inside the CO₂ flux (Livingston and Hutchinson 1995, Jensen *et al.* 1996, Alavoine *et al.* 2008). Each chamber method has its own strengths and weaknesses but, there is no single standard or reference to test the accuracy of these methods (Pumpanen *et al.* 2004).

Past literature comparisons showed differences between chamber techniques (Rochette *et al.* 1992, Kabwe *et al.* 2002, Janssens *et al.* 2000, Rochette and Hutchison

2005); non-steady-state chambers gave lower fluxes in comparison to steady-state chambers with underestimations ranging from 4% to 35% (Rayment 2000, Pumpanen *et al.* 2003, 2004). The observed differences between methods are due to a chamber's impact on the CO_2 concentration gradient at the soil-atmosphere interface and sensitivity to pressure differences between chamber headspace and the atmosphere.

Chambers based on alkali absorption overestimated lower (120 mg m⁻² hr⁻¹) CO₂ fluxes by 25% and underestimated higher (770 mg m⁻² hr⁻¹) CO₂ fluxes by 57% when compared with known CO₂ fluxes (Nay *et al.* 1994). Similar results were obtained by Jensen *et al.* (1996). They compared the static chamber alkali trap method with the closed dynamic chamber method on pasture as well as forest sites and suggested that the static method on average gave 12% higher flux rates below 100 mg CO₂-C m⁻² hr⁻¹, but 5 times lower flux rates above 100 mg CO₂-C m⁻² hr⁻¹. Therefore a comparison between chamber methods is essential if confidence with the measurement of CO₂ efflux is to be gained. The objective of this study was to compare and evaluate the three methods:

1) Static chamber using alkali traps,

2) Static chamber flux gradient method by periodic sampling and analysis of gas samples and
3) Portable Infra-Red Gas Analyser (IRGA): EGM-1connected to SRC-1 (PP systems)
for measuring a wide range of soil CO₂ emissions (Low vs. High) and variability to select the most appropriate method for measuring *in-situ* CO₂ fluxes.

3.2 Material and methods

The chamber methods were compared on low emission sub-surface soil and high emission surface soil in this study. The experiment was conducted in a closed room without any exhaust fan to maintain a constant temperature.

3.2.1 Description of soils

The soil used in this study was a Manawatu fine sandy loam (Typic Fluvial Recent Soil) under a permanent ryegrass pasture and had micaceous clay mineralogy (Saggar *et al.* 1999). Soils for the high and low emission studies were sampled from 0-6 cm and 2.5-7.5 cm depth. The reason for this was based on the root density differences reported by Kusumo (2009), that the surface soils would give 13.2 mg dry root g^{-1} dry soil to decompose compared with 6.1 mg dry root g^{-1} dry soil at 2.5-7.5 cm soil depth.

For the low emission study, a sub-surface soil sample (2.5-7.5 cm depth; total C= 30 g kg⁻¹, total N= 3.0 g kg⁻¹, pH= 5.4, Olsen P= 24.6 mg kg⁻¹ soil, exchangeable K= 249.6 mg kg⁻¹ soil, CEC= 16 cmol_c kg⁻¹) was collected from one spot at 2.5-7.5cm depth avoiding the

dense pasture root mass contribution to readily decomposable C in autumn (March 2009). The sub-surface soil sample was passed through a 5mm sieve and stored at 20°C for 2 weeks, allowing some decomposition of the readily decomposable C. This was a strategy to produce a low emission soil. It was assumed that two weeks storage would be sufficient to decompose the readily decomposable C. Moisture content of the soil was determined on the day when soil was put into each of the six closed-base replicate PVC chambers (23.5 cm diameter; 16 cm high). Two kg of field moist sub-surface soil (22.1 % gravimetric moisture content) was transferred into each of the six replicate chambers and firmly packed to obtain a final bulk density of 0.98 g cm⁻³. Moisture content in the replicate chambers was maintained throughout the experiment by weighing the chambers and spraying the required amount of deionised water onto the surface.

For the high emission study, surface soil samples (0-6.0 cm depth; total C= 32 g kg⁻¹, total N= 3.2 g kg⁻¹, pH= 5.3, Olsen P= 64.9 mg kg⁻¹ soil, exchangeable K= 292.5 mg kg⁻¹ soil, CEC= 17 cmol_c kg⁻¹) were collected from eight randomly selected areas in winter (June 2009). Three (6 cm depth and 10.5 cm diameter) cores were taken from each replicate area, sprayed with glyphosate and left in the glasshouse for 7 days to kill the surface grass. Each core was then cut into pieces and the soils from the three cores of each replicate were combined, weighed and then put in the chambers. Moisture content of the soil was determined at this time. Each closed-base chamber was filled with 2276 g of field moist surface soil (34.0 % gravimetric moisture content) and firmly packed to obtain a final bulk density of 0.86 g cm⁻³. Similar amounts of field moist soil were filled in the eight chambers representing eight replicates. Moisture content did not vary much between the eight replicate chambers but differences in root masses were not accounted for. The total C content given for the surface soil is of a 2 mm sieved ring ground sample and does not take roots present in the soil into account. Moisture content in the replicate chambers was maintained throughout the experiment by weighing the chambers and spraying the required amount of deionised water onto the surface.

3.2.2 Measurement of CO₂ flux

Low emission study: Static alkali trap chamber method and infrared gas analyser-EGM-1were compared on the low emission sub-surface soil.

3.2.2.1 Static alkali trap chamber method: Carbon dioxide flux was determined by absorbing CO₂ using 30 ml of NaOH solution of two concentrations (0.5M and 1.0M) each in a sealed chamber headspace for a specific period of time. A plastic petri dish covering 13.1% of a chamber's area was used for storing 30 ml of NaOH solution. The petri dish was elevated approximately 7 cm above the soil surface in the centre of the chamber. Two horizontal sticks were placed across the chamber to elevate the petri dish. For the six replicated chambers, 0.5M and 1.0M NaOH solutions were placed respectively in three chambers each. The enclosure times were 4, 8, 12, 16, 20 and 24 hours for absorbing CO_2 from each chamber. After every absorption interval the chambers were opened, NaOH solution from each chamber was taken out and stored in a plastic container for titration to calculate the amount of CO₂ emitted during that particular absorption interval. The amount of CO₂ emitted was further divided by the interval duration to calculate the hourly rate. The chambers were left open for 45 minutes to remove the existing CO₂ concentration built up in the chambers. Thereafter, the measurement of CO₂ flux from each chamber was taken using EGM-1. After taking the measurements with EGM-1, fresh alkali solutions were placed in each chamber for the next interval and the procedure was repeated after every absorption interval. Different absorption intervals were not compared simultaneously as it was assumed that low emission soil will give constant CO_2 flux throughout the study period. The total amount of CO_2 absorbed in NaOH solution was determined by back-titrating the excess NaOH with 0.2M HCl after precipitation of carbonates with BaCl₂. All the chemicals used were of analytical grade and solutions were prepared using deionised water. The solution of 0.2M HCl was standardised against Na_2CO_3 as per the method outlined by Lambert *et al.* (1949). The chamber headspace volume was 5282 ml. The seal between the chamber top and its permanently installed base was perfect. Each chamber had an internal half-turn locking system and a greased O-ring which formed a gas-tight seal when closed with a lid.

3.2.2.2 <u>Infrared (IR) gas analyser-Dynamic chamber method</u>: Carbon dioxide flux was measured by a dynamic chamber (10cm diameter; 15cm high) coupled to a portable infrared gas analyzer (IRGA) in a closed circuit (EGM-1 equipped with SRC-1, PP systems). Mixing of the air in the closed soil respiration chamber (SRC) during measurement was ensured by a small fan running inside the chamber. The fan speed was 0.5 m s⁻¹ and there was a mesh

screen between the fan and the soil surface to slow the air velocity at the soil surface. The mixed air was circulated (flow rate 200-400 ml min⁻¹) from the chamber into the IRGA sensor cell and back to the chamber by a pump in the EGM-1. The IRGA contained software to calculate CO_2 flux rates and each measurement took less than 2 minutes (PP systems 2010). Before each flux measurement, the SRC was moved away from the closed base chamber filled with the soil and the SRC mixing fan ran for 10s to restore the IRGA sensor cell to the ambient CO_2 concentration in the room. Three CO_2 flux measurements with EGM-1 were made from three different locations within each chamber at the end of every absorption interval i.e. 4, 8, 12, 16, 20, 24 hours of the alkali trap method. The mean of three flux values are stated as the flux value from each replicate chamber. In this study a soil collar for the placement and formation of a seal for the dynamic chamber was not used.

High emission study: Static alkali trap chamber, infrared gas analyser-EGM-1 and static chamber flux gradient methods were compared on high emission surface soils. Carbon dioxide measurements were made 2 hours after filling the chambers with soil.

3.2.2.3 <u>Static chamber flux gradient method</u>: Carbon dioxide gas samples (25 ml) were taken for a period of one hour with 60 ml polypropylene syringes fitted with 3-way stopcocks after sealing the chambers with a lid having one port. Five gas samples (25 ml each) were taken from each chamber at times t0, t10, t20, t30 and t60 (i.e. 0 min, 10 min, 20 min, 30 min and 60 min respectively, after closing the chamber). The gas samples were transferred to 12 ml evacuated vials and then analyzed using a Shimadzu GC- 17A gas chromatograph; CO₂ flux (mg m⁻² hr⁻¹) was estimated from the measurements made at different time intervals. The sample of the ambient air taken just after closing the chamber (t0) was used as a reference for calculating CO₂ fluxes. Carbon dioxide gas samples (25 ml) were taken by syringe from all the eight replicate chambers. After completion of a one hour measurement period, chambers were left open for 45 minutes to remove the existing CO₂ concentration built up in the chambers. Thereafter, 1.0 M NaOH traps were placed in the chambers to absorb CO₂ for 4 hours. The seal between the chamber top and its permanently installed base was perfect. Each chamber had an internal half-turn locking system and a greased O-ring which formed a gas-tight seal when closed with a lid. The chamber headspace volume was 4937 ml.

3.2.2.4 <u>Static chamber alkali trap method</u>: Carbon dioxide flux rates were determined by absorbing CO_2 in 1.0M NaOH placed in a plastic petri dish (13.1% of the chamber area) in all the eight replicates for 4 hours duration.

3.2.2.5 <u>Infrared (IR) gas analyser-Dynamic chamber method</u>: Three measurements from different locations within each chamber were made at the end of a 4 hour absorption interval, as for the alkali trap method.

3.2.3 Statistical analysis

An analysis of variance using SAS software (9.1) was performed on carbon dioxide fluxes measured by the three methods using the General Linear Model (GLM) procedure. Mean comparisons between CO_2 fluxes were done using Fisher's least significant difference (LSD) at 5% level of significance.

3.3 Results

3.3.1 Low emission study

Two chamber methods (static chamber (alkali trap) and dynamic chamber (EGM-1)) and two NaOH concentrations (0.5M and 1.0M) were compared on the low emission subsurface soil under controlled laboratory conditions. The CO₂ fluxes measured by 0.5M NaOH traps varied from 43.5 mg CO₂ m⁻² hr⁻¹ to 55.6 mg CO₂ m⁻² hr⁻¹, and by 1.0M NaOH traps varied from 52.4 mg CO₂ m⁻² hr⁻¹ to 69.9 mg CO₂ m⁻² hr⁻¹ (Table 3.1). The CO₂ fluxes measured by 0.5M and 1.0M NaOH traps over a 12 hour absorption interval were significantly higher than those measured over 4 and 8 hour intervals.

Table 3.1: Effect of absorption	intervals on the	CO ₂ fluxes usin	ng closed static	chambers	with
NaOH (alkali) traps of varying r	nolarities.				

Absorption interval	CO_2 flux (mg CO_2 m ⁻² hr ⁻¹)		
(hours)	0.5M NaOH	1.0M NaOH	
4	B 43.5±5.1 a	B 61.4±4.4 a	
8	B 44.8±2.5 a	C 52.4±1.2 a	
12	A 55.4±2.2 a	A 69.9±2.2 b	
16	A 55.6±2.2 a	BC 57.6±1.1 a	
20	BA 50.1±1.0 a	C 53.2±0.5 a	
24	BA 48.6±0.7 a	C 54.1±0.8 b	

Each value represents a mean of three replicates± SE

Capital letters represents absorption interval difference

Small letters represents absorption NaOH concentration difference

Means followed by same small letter in rows and same letter in columns are not significantly different

The CO_2 fluxes measured over a 20 and 24 hour absorption interval by both 0.5M and 1.0M NaOH trap solutions were lower than those measured over a 12 hour interval; however, significant differences were observed only in the case of 1.0M NaOH. No

significant difference was observed between the CO_2 fluxes measured by 0.5M and 1.0M NaOH except over the 12 and 24 hour intervals.

The CO₂ fluxes measured by EGM-1 equipped with SRC-1 in the chambers with 0.5M NaOH traps varied from 45.5 mg CO₂ m⁻² hr⁻¹ to 103.3 mg CO₂ m⁻² hr⁻¹, and in the chambers with 1.0M NaOH traps, varied from 43.3 mg CO₂ m⁻² hr⁻¹ to 91.1 mg CO₂ m⁻² hr⁻¹ (Tables 3.2 a, b). The CO₂ fluxes measured by EGM-1 over a 24 hour absorption interval (both in 0.5M and 1.0M NaOH chambers) were significantly higher than any other absorption interval.

Table 3.2: Effect of absorption intervals on CO_2 fluxes using closed static chambers with NaOH (alkali) traps 0.5M NaOH (A) and 1.0M NaOH (B) compared with spot measurements made by EGM-1 equipped with SRC-1.

Absorption interval	CO_2 flux (mg CO_2 m ⁻² hr ⁻¹)		
(hours)	0.5M NaOH	EGM-1	
4	B 43.5±5.1 a	B 45.5±4.0 a	
8	B 44.8±2.5 a	B 56.6±10.7 a	
12	A 55.4±2.2 a	B 50.0±5.7 a	
16	A 55.6±2.2 a	B 53.3±1.9 a	
20	BA 50.1±1.0 a	A 98.9±12.8 b	
24	BA 48.6±0.7 a	A 103.3±12.6 b	

(A)

(B)

Absorption interval	CO_2 flux (mg CO_2 m ⁻² hr ⁻¹)		
(hours)	1.0M NaOH	EGM-1	
4	B 61.4±4.4 a	B 54.4±4.4 a	
8	C 52.4±1.2 a	B 43.3±6.6 a	
12	A 69.9±2.2 a	B 46.6±3.3 b	
16	BC 57.6±1.1 a	B 60.0±16.7 a	
20	C 53.2±0.5 a	B 66.6±3.8 b	
24	C 54.1±0.8 a	A 91.1±2.2 b	

Each value represents a mean of three replicates \pm SE

Capital letters represents absorption interval difference

Small letters represents NaOH and EGM-1 method difference

Means followed by same small letter in rows and same letter in columns are not significantly different

A comparison between the CO_2 fluxes as measured by alkali traps (0.5M and 1.0M NaOH) with EGM-1 (Tables 3.2 a, b) shows similar CO_2 fluxes for the majority of the

absorption intervals. However, CO_2 fluxes measured by EGM-1 at 20 and 24 hour absorption intervals were significantly higher than for the alkali trap method.

In the present study the amount of residual alkali after a 24 hour absorption interval was 84.0% for 0.5M and 91.0% for 1.0M NaOH. Kirita and Hozumi (1966) stated that to obtain maximum absorption of CO_2 the amount of unused alkali should not be less than 80%, which suggested that both concentrations were capable of maximum CO_2 absorption but 1.0M NaOH had more capacity to absorb the higher CO_2 fluxes. Therefore, in the high emissions study 1.0M NaOH was used to measure CO_2 fluxes.

3.3.2 High emission study

Three chamber methods were investigated: static chamber alkali trap using 1.0M NaOH, dynamic chamber (EGM-1equipped with SRC-1), and the static chamber flux gradient method were compared on high emission surface soil under controlled laboratory conditions. Carbon dioxide fluxes for high emission study were measured for six weeks on days 1, 3, 5, 8, 10, 12, 15, 17, 19, 36, 38 and 40. The CO₂ fluxes measured by 1.0M NaOH traps varied from 553.5 to 1166.5 mg CO₂ m⁻² hr⁻¹, by EGM-1 they varied from 118.2 to 956.6 mg CO₂ m⁻² hr⁻¹ and by the static chamber flux gradient method they varied from 79.5 to 613.3 mg CO₂ m⁻² hr⁻¹ over the six week period (Figure 3.1). The patterns of CO₂ fluxes were similar for the three measurement methods; higher fluxes were observed in the beginning and these then decreased gradually.

The CO₂ fluxes measured by a 1.0M NaOH trap, EGM-1, and the closed chamber flux gradient method on any measurement day varied significantly from each other. CO₂ flux measurements by a 1.0M NaOH trap were significantly higher than the EGM-1 and the flux gradient method. The average absolute differences between the alkali trap and EGM-1measured fluxes during the first, second, third and sixth week were 264.6, 289.2, 348.0 and 439.6 mg CO₂ m⁻² hr⁻¹, which suggested that on average, the alkali trap method gave 30%, 85.4%, 121.1%, and 360 % higher fluxes than the EGM-1. Similarly, the average absolute differences between the alkali trap and flux gradient measured fluxes during the first, second, third and sixth week were 564.8, 467.1, 500.4, 472.3 mg CO₂ m⁻² hr⁻¹ suggesting 96.6%, 262.7%, 368.8% and 549.5% higher fluxes in the first, second, third and sixth week by the alkali trap method than by the closed static flux gradient method.



Figure.3.1: Comparison of CO₂ fluxes measured by 1.0M NaOH traps, EGM-1 and closed static flux gradient methods. Each value represents the mean of eight replicates with standard errors (\pm) shown by vertical bars. Vertical bars above represent LSD (P < 0.05) for comparison among the three methods where significant differences were found.

3.4 Discussion

3.4.1 Low emission study

Two major chamber approaches: the closed static chamber with an alkali trap and the closed dynamic chamber system (EGM-1) were compared on low emission sub-surface soil. The alkali trap provides integrated flux rates over time. Most of the alkali trap (concentration of NaOH varies from 0.5M to 2M depending upon the expected flux values) CO_2 flux values reported in the literature were obtained under long absorption times i.e. 24 hours (Buyanovsky *et al.* 1986; Rochette *et al.* 1992; Jensen *et al.* 1996; Aslam *et al.* 2000; Alvarez *et al.* 2001; Yim *et al.* 2002; Rottmann and Joergensen 2011). However, a long absorption time of 24 hours disturbs the natural conditions of the enclosed soil surface and provides either under- and over-estimation of CO_2 fluxes (Davidson *et al.* 2002). The present study verified that CO_2 fluxes measured by 1.0M NaOH over longer absorption intervals of 20 and 24 hours. During long absorption intervals, the absorption of CO_2 by the NaOH (alkali) trap is limiting, resulting in a build-up of CO_2 concentration inside the chamber (Jensen *et al.* 1996). In alkali trap absorption, CO_2 is first absorbed at the liquid

interface and then diffuses into the bulk solution. The NaOH solution placed in the chamber was not stirred, which may have resulted in a carbonate gradient which limited the capacity of NaOH to absorb CO₂. Moreover, long absorption intervals causes oxygen depletion, the build-up of CO₂ concentration and change in temperature & moisture content inside chambers, leading to low flux rates (Norman *et al.* 1997; Rochette and Hutchinson 2005).

In the present study, CO_2 fluxes measured by 1.0M NaOH were similar to the fluxes measured by 0.5M NaOH during the majority of absorption intervals, except for fluxes measured over 12 and 24 hour intervals. Carbon dioxide fluxes measured by 1.0M NaOH were significantly higher than those measured by 0.5M NaOH at 12 and 24 hour absorption intervals. There seems to be no effect of alkali concentration on CO_2 fluxes; any differences that occurred over 12 and 24 hour intervals were likely due to experimental variability.

Comparison of CO₂ fluxes measured by alkali traps (0.5M and 1.0M NaOH) and EGM-1 showed no differences for the majority of the absorption intervals except for the 20 and 24 hour intervals where fluxes measured by EGM-1were significantly higher than those of the alkali trap method. Higher CO₂ concentration build-up inside the chambers due to long absorption intervals of 20 and 24 hours could explain why significantly higher fluxes were measured by EGM-1 at 20 and 24 hours absorption intervals. The EGM-1 measurements were made on the chambers 45 minutes after removing the NaOH (alkali) traps. EGM-1 measured fluxes were actually the flush of CO₂ that was built-up inside the chambers due to the long absorption intervals. Davidson *et al.* (2002) suggested that short sampling duration of 2 minutes by EGM-1 minimizes the artefact caused by altering the CO₂ concentration gradient between the soil atmosphere and the chamber headspace. However, the spot measurements made by EGM-1 showed greater variability than those made from alkali traps.

The static alkali trap chamber method is the most commonly used method to measure soil CO₂ efflux (Yim *et al.* 2002). However, this method is greatly influenced by numerous methodological factors such as solution strengths, volumes, absorption areas and absorption times that in turn can affect the flux rates. Gupta and Singh (1977) compared different NaOH (alkali) concentrations (0.1M, 0.25M, 0.5M. 0.75M, 1.0M, 1.25M) under field conditions. The volume of alkali was kept at 10ml in each case and the alkali trap/chamber area ratio was 12.5%. They observed a linear increase in the amount of CO₂ absorbed with increasing concentration of NaOH from 0.1M ($45 \pm 1.8 \text{ mg CO}_2 \text{ m}^{-2} \text{ hr}^{-1}$) to 1.25M (282 ± 5.9 mg CO₂ m⁻² hr⁻¹), showing that increase in concentration of NaOH can affect the calculated flux with this technique. However, in a separate experiment using different volumes (10, 20, 30, 40 and 50 ml) and concentrations (0.1-1.25M) of NaOH under field conditions, Gupta and

Singh (1977) showed that additional increase in NaOH volume beyond 30ml had no effect on CO₂ fluxes at 0.75, 1.0 and 1.25M measured for a 24 hour interval. Similarly, Minderman and Vulto (1973) using different alkali (KOH) concentrations under field conditions (0.25-2.0M) showed that additional increase in alkali volume beyond 30ml had no effect on CO₂ fluxes as measured for a 24 hour interval. The size and shape of the alkali container is also important as it determines the ratio of the alkali surface area to ground enclosed by the chamber. For efficient CO₂ absorption, Kirita (1971) recommended a 13% alkali/chamber area ratio depending on their chamber designs and deployment conditions. The appropriate alkali (NaOH/KOH) concentration depends upon the ecosystem and expected CO₂ fluxes. The past literature and results of the present study suggest the use of a short absorption period of 4 hours for measuring CO₂ fluxes to minimize the chamber effects on CO₂ fluxes.

3.4.2 High emission study

In arable ecosystems flux rates obtained by the static chamber method have ranged between 0 and 1100 mg CO₂ m⁻² hr⁻¹ (Dao 1998; Aslam *et al.* 2000; Alvarez *et al.* 2001; Omonode *et al.* 2007; Ussiri and Lal 2009) and by the dynamic chamber method from 0 to 4500 mg CO₂ m⁻² hr⁻¹ (Reicosky and Lindstrom 1993; Reicosky *et al.* 1997; Alvaro-Fuentes *et al.* 2007; Bauer *et al.* 2006; Akbolat *et al.* 2009). The CO₂ fluxes measured in the high emission study by the static and dynamic methods are within the range reported for arable ecosystems.

The patterns of CO_2 fluxes measured during the high emission study were similar for the three measurement methods; higher fluxes were observed in the beginning and these then decreased gradually. This suggested an initial fast phase due to mineralization of easily degradable organic C in residues, followed by a slower phase in which more complex and recalcitrant forms or transformed metabolites are mineralized (Verma *et al.* 2010). The soil moisture and temperature were constant throughout the measurement period.

The results of a high emission study contradicted the findings of the previous comparative studies in which CO₂ flux estimates from the alkali traps were much lower than those from the EGM-1 and other dynamic methods (Ewel *et al.* 1987; Rochette *et al.* 1992). The lower fluxes obtained with EGM-1 compared to alkali trap fluxes in this study may either be caused by systematic under-recording due to poor sealing with the soil surface in the chamber leading to air leakage, or the NaOH trap method might be over-estimating the CO₂ fluxes. The flux rates determined by EGM-1 are also influenced by the number and frequency of measurements (King and Harrison 2002). In the present study CO₂ flux

measurements with EGM-1 were made once during the day of measurement. More measurements at different time intervals would have improved the accuracy and comparability with alkali trap measurements. The smaller flux values measured by the closed static chamber flux gradient method might have resulted from the reduced soil-atmospheric exchange of CO_2 due to concentration build up within the chamber (Conen and Smith 2000).

Nakadai et al. (1993) observed acceleration in soil CO₂ fluxes with decreasing CO₂ concentration in the chamber using the alkali trap static chamber method under laboratory conditions. In order to check whether the alkali trap (1.0M NaOH) was overestimating CO₂ fluxes, plastic petri dishes containing 30 ml of 1.0M of NaOH (13.1% of the chamber area) were placed in four replicate chambers to absorb CO₂ and simultaneous measurements of gas concentrations were done by syringe sampling. Each chamber had an internal half-turn locking system and a greased O-ring which formed a gas-tight seal when closed with a lid. Eight gas samples were taken at t0, t10, t20, t30, t60, t120, t180 and t240 (i.e. 0 min, 10 min, 20 min, 30 min, 60 min, 120 min, 180 min and 240 min, respectively from each chamber after closing the chamber with 1.0M NaOH trap). The chambers were sealed with air tight lids. The amount of increase and decrease in CO_2 concentration in the chamber headspace depends upon the efficiency of CO_2 absorption by the alkali trap (Bekku *et al.* 1997). The CO₂ concentration in the chambers decreased with time showing that the NaOH solution in the chamber was efficiently absorbing the CO₂ released from the soil. Mean head space CO₂ concentration in the four chambers decreased from 641 ppm at t0 to 353 ppm at t240 showing that the CO₂ concentration in the chambers did not go much below the ambient atmospheric CO₂ concentration during the four hour absorption period, thus overruling any overestimation of CO₂ flux by the alkali trap. When the headspace CO₂ concentration is equal to the ambient level, the absorption rate is equal to the surface flux (Hutchinson and Rochette 2003). The ambient CO₂ concentration of the room was 612 ppm. The ambient concentration of CO₂ was high because the experiment was conducted in a closed room without any exhaust fan to maintain the constant temperature; Nakadai et al. (1993) observed approximately 500 ppm CO₂ initial concentration under laboratory conditions using the alkali absorption method.

The ideal method for measuring CO_2 flux should have minimal influence on soil surface natural conditions and the concentration gradient, hence giving a reliable estimate of CO_2 efflux (Rochette and Hutchinson 2005). Theoretically, the dynamic method (EGM-1) fulfils this condition to a greater extent than the static method, with measurements over a short period of time minimizing chamber effects on soil temperature, moisture and the CO_2 concentration gradient (Le Dantec *et al.* 1999; Janssens *et al.* 2000). However, the dynamic method does not have the time-integrative power of the static method unless a very large number of measurements are taken during the measurement period. Such high daily sampling frequency poses constraints and is not easily accomplished with manual measurements (Jensen *et al.* 1996; King and Harrison 2002). This was the key reason why the alkali trap method was preferred over EGM-1. The flux gradient method was sensitive to large increases in the concentration of emitted CO_2 and expensive compared to the alkali trap method. Hence the alkali trap method was preferred over EGM-1 and the flux-gradient method to measure CO_2 fluxes in further studies.

3.5 Summary and conclusions

The flux of CO_2 emissions from soil may be needed in a suite of measurements required for monitoring short-term management effects on changes in soil C stocks. The three different chamber methods: i) static alkali traps, ii) flux gradient and iii) a portable infra-red gas analyser (IRGA): EGM-1connected to SRC-1 (PP systems) were compared for measuring CO_2 fluxes under controlled laboratory conditions using low (sub-surface) and high (surface) CO_2 -emitting soils, using a range of enclosure intervals (4, 8, 12, 16, 20 and 24 hours). Carbon dioxide fluxes measured by alkali trap and EGM-1 in the low emission study were similar during the majority of enclosure intervals except at 20 and 24 hours where the alkali traps measured significantly lower fluxes than EGM-1. This could be explained by the build-up of CO_2 concentrations inside the chamber as being closed for 20 and 24 hours affects measured CO_2 fluxes because soil flux rates depend upon the concentration gradient inside the chamber.

However, the fluxes measured over a six week period from the high emitting surface soil ranged between 553.5 and 1166.5 mg $CO_2 \text{ m}^{-2} \text{ hr}^{-1}$ for the NaOH traps, between 118.2 and 956.6 mg $CO_2 \text{ m}^{-2} \text{ hr}^{-1}$ for the EGM-1 and between 79.5 and 613.3 mg $CO_2 \text{ m}^{-2} \text{ hr}^{-1}$ for the flux gradient method. The lower flux values obtained with EGM-1 and the flux gradient method compared to alkali traps may be caused by systematic under-recording. Poor sealing of the EGM-1 with the soil surface, and decreasing atmospheric/soil CO_2 gradients under the flux gradient technique were suspected reasons. Overall results of this study show that EGM-1 measurements were highly variable in comparison to other methods. For extended periods of monitoring this method requires numerous replicated spot measurements. Thus, the measurement frequency with EGM-1 poses constraints and may not be easily accomplished. The static chamber flux gradient method seems sensitive to rapid and large increases in the concentration of emitted CO_2 and expensive compared to the alkali trap method. By contrast, the alkali trap method was not sensitive to different rates of CO_2 emission and has the advantages of simplicity, cheapness and integrating CO_2 flux over time (4 to 24 hours). Therefore, the alkali trap method was preferred over EGM-1 and the flux-gradient method to measure CO_2 fluxes in the following laboratory and field studies.

3.6 Effect of wind induced pressure fluctuations on carbon dioxide fluxes from simulated tillage and No-tillage treatments

3.6.1 Introduction

Generally, higher soil CO₂ fluxes are expected under high wind velocity due to the lower (negative) pressure created at the soil surface (Kimball and Lemon 1971; Takle *et al.* 2004; Xu *et al.* 2006) which pulls the CO₂ rich air from the soil. This phenomenon is known as the 'Venturi Effect' (Conen and Smith 1998; Reicosky and Archer 2007). However, pressure fluctuations only affect the CO₂ transport and not its production which suggests that the venturi effect may not have significant long term effect on CO₂ emissions from soils (Subke *et al.* 2003). In this section I report the results of the pressure fluctuation study conducted under controlled laboratory conditions comparing the differences in CO₂ fluxes from simulated tilled and No-tilled soils maintained at ambient and lower pressure conditions.

3.6.2 Material and methods

3.6.2.1 Soil characteristics

The experiment was conducted on Ohakea silt loam soil (0-10 cm) having 17% sand, 59% silt and 24% clay with the following characteristics: pH 5.9 (1:2.5 soil:water); bulk density of undisturbed soil was 1.07 g cm⁻³; total C & N contents were 39.7 g kg⁻¹ & 3.5 g kg⁻¹, Olsen P: 21.5 mg kg⁻¹ soil, exchangeable K: 163.8 mg kg⁻¹ soil, SO₄-S: 8.25 mg kg⁻¹ soil and CEC 19.8 cmol_c kg⁻¹; gravimetric soil water content at the time of sampling was 44.8 percent.

3.6.2.2 Chemical and physical analysis

Soils samples for chemical and physical analysis were collected in triplicate (0-10 cm soil depth) on the same day as soil cores were collected for CO_2 measurements. Each replicate was composed of 12 cores (2.5 cm diameter) randomly collected across the field. Soil samples were air dried and ground to pass through a 2mm sieve for further analysis. Soil pH (1:2.5), cation exchange capacity (CEC) and exchangeable K were analysed according to Blackmore *et al.* (1987). Olsen P and SO₄-S were determined according to Olsen *et al.* (1954) and Blackmore *et al.* (1987). A portion of each sample was ground to pass through a 0.2 mm sieve for total carbon and nitrogen analysis using a LECO induction furnace (Blackmore *et al.* 1987). Particle size distribution was determined by the pipette method of Claydon (1989). For soil bulk density determination, undisturbed soil cores (10 cm diameter,

10 cm depth) were taken and soil bulk density was calculated by dividing the mass of oven dried soil by the volume of the soil core. The gravimetric soil moisture content (SMC) was measured at 0-10 cm depth. Field moist samples were weighed, oven-dried (105° C) to constant mass, and weighed again. The final mass (*Ms*), and the difference between the field moist and dry masses (*Mw*), were used to calculate the gravimetric SMC = (*Mw/Ms*) X 100.

3.6.2.3 Collection of soil cores and simulation of tillage

A total of fifty four *in-situ* soil cores (10 cm diameter; 10 cm depth) were collected from the Sanson site (40°14'34.33"S and 175°21'27.07"E) during winter for simulated tillage (ST), No-tillage (NT) and non-disturbed (ND) control treatments. The site was sprayed with glyphosate herbicide prior to collection of the soil cores. Thirty six ND cores were collected after glyphosate application and before the start of tillage operations from three different locations on the farm, with twelve cores taken from each location. Immediately after the Cross Slot® No-tillage drilling eighteen NT cores were collected over the slots, with six cores collected from each location. Eighteen of the thirty six ND cores were broken up in the laboratory to simulate tillage treatment. The ST treatment was formulated from three *in-situ* ND soil cores by emptying the soil, breaking it into pieces, thoroughly mixing and packing it in a plastic container (19 cm diameter; 10 cm high) to obtain a final bulk density of 0.63 g cm⁻³. Each replicate of ST, NT and ND treatments were comprised of three intact cores collected from one sampling location. To calculate the CO₂ flux from the three treatments the amount of CO₂ emitted from each chamber was divided by the combined surface area of the three cores as the surface area of the plastic container used for the ST was comparable to the combined surface area of the three cores. Three replicates of each tillage treatment (ST, NT and ND) with no soil control were assessed both at ambient and lower pressures.

3.6.2.4 Application of different pressures to soils

The lower (negative) pressure of 20 millibars was obtained in each replicate chamber by using an individual bubble tower (Plate 3.1 A). A bubble tower column was constructed from a plastic tube (diameter=3.0cm; height=45.0cm) and sealed at the top by a two-hole rubber stopper. One hole was for the inlet tube (diameter=6.0mm; height=500mm) and the other hole for the outlet tube to the chamber. The lower end of the column also had a rubber stopper. All the connections were made with appropriate PVC tubing of 4.0 mm diameter. The bubble tower column was filled up to 35 cm with water and lower pressure was adjusted by lowering the inlet tube 20 cm below the surface of the water in the column. The flow of air was maintained constant in each replicate chamber by adjusting the bubble rates to approximately three bubbles per second (approximately a flow rate of 30 ml min⁻¹ allowing
chamber headspace turnover in 4 hours) in the bubble tower column and the exhaust gas scrubbing glass tube containing 50ml 1.0M NaOH solution.

For ambient pressure, chambers were set up identically to those set up at lower pressure but without a bubble column (Plate 3.1 B). The air flow was also adjusted to a bubbling rate approximately three bubbles per second in the exhaust gas scrubbing glass tube containing 50ml 1.0M NaOH solution so that equal air flows were obtained for ambient and lower pressure chambers.

3.6.2.5 Carbon dioxide measurements

Carbon dioxide was measured using the alkali trap static chamber method. Carbon dioxide measurements started on the same day of sampling, immediately after formulation of the simulated tillage treatment. Soil cores and containers filled with soil were placed in closed base static chambers along with a plastic petri dish containing 30 ml 1.0M NaOH to absorb CO₂ inside the chambers. Each chamber was sealed with an air tight lid having two ports. One port was connected to the bubble tower column and the other port was connected to the glass tube containing 50ml 1.0M NaOH solution sealed at the top with a two-hole rubber stopper. One hole was for the inlet tube (diameter=6.0mm; height=200mm) which was dipped in NaOH solution connected to the static chamber, and the other hole was for the short plastic tube which was connected to the suction hose. All the connections were made with appropriate PVC tubing of 4.0 mm diameter.

Carbon dioxide was absorbed for a duration of 16 hours which was split into 4 and 12 hour measurement periods. After completion of the 4 hour measurement period, NaOH solutions were immediately replaced with fresh NaOH solutions and then measurements were taken for another 12 hour period. One measurement of 16 hours was made in the first week after sampling of the soil cores and another measurement of 16 hours was made in the second week after sampling of the soil cores for all three (ST, NT and ND) treatments. The amounts of CO₂ absorbed in the NaOH solutions were determined by titrating them with dilute 0.2M HCl. The flux values were calculated by dividing the amounts of CO₂ absorbed by NaOH solutions in a plastic petri dish and glass tube were summed up to report as a lone flux value at a particular measurement period and pressure. Moisture contents in the soil cores and container were maintained throughout the experiment by weighing the soil cores and container and spraying the required amount of deionised water onto the surface to bring them to their original moisture at which they were sampled from the field.



Plate.3.1: Closed static chambers: lower pressure setup (A) and ambient pressure setup (B).

3.6.2.6 Statistical analysis

Mean carbon dioxide fluxes from the three tillage treatments at ambient and lower pressures were compared using the Kruskal-Wallis test followed by the Bonferroni (Dunn) t-test.

3.6.3 Results

Carbon dioxide fluxes measured over 4 and 12 hours under ambient and lower pressures during the two week study period are given in Table 3.3. Measurements made during the first week after sampling of soil cores suggest that CO₂-C fluxes for ST, NT and ND treatments with the onset of lower (negative) pressure were significantly higher than CO₂-C fluxes at ambient pressure by 12.0, 14.5, and 33.8%, respectively for a 4 hour measurement period. Thereafter, significantly higher CO₂-C fluxes were observed under lower pressure by 4.6 and 17.2 % for NT and ND treatments for the following 12 hour measurement period. No significant differences were observed between CO₂-C fluxes under ambient and lower pressures for the ST treatment during the 12 hour measurement period (Table 3.3). During the second week, no significant differences were observed between CO₂-C fluxes under ambient and lower pressures for the ST, NT and ND treatments for a 4 hour measurement period. However, during the 12 hour measurement period significant differences between CO₂ fluxes under ambient and lower pressures for the ST, NT and ND treatments for a 4 hour measurement period. However, during the 12 hour measurement period significant differences between CO₂ fluxes under ambient and lower pressures for the ST, NT and ND treatments for a 4 hour measurement period. However, during the 12 hour measurement period significant differences between CO₂ fluxes under ambient and lower pressures for the ST, NT and ND treatments for a 4 hour measurement period. However, during the 12 hour measurement period significant differences between CO₂ fluxes under ambient and lower pressures for the NT and ND treatments. At ambient pressure CO₂-C fluxes from ST were significantly higher than the NT treatment only for a 4 hour measurement period during the first week.

Thereafter, no significant differences between the three treatments were found at subsequent measurements. Under lower pressure, CO₂-C fluxes from the ST treatment during any measurement interval during the two week study period were not significantly higher than the NT treatment. However, CO₂-C fluxes from the ND treatment were consistently higher than the NT and ST treatments throughout the study period. Similar trends and statistical results were observed for the accumulated CO₂-C emissions over 4 and 12 hour measurement periods for the whole study duration (Table 3.4). The accumulated CO₂-C emissions were measured with the onset of lower pressure than those measured under ambient pressure (Table 3.4).

First week	mg CO ₂ -C m ⁻² hr ⁻¹				
4 hours	ST	NT	ND		
Lower pressure	A 358±9.8 a b	A 309±2.9 b	A 380±9.5 a		
Ambient pressure	B 320±7.9 a	B 270±12.0 b	B 284±11.0 a b		
		mg CO ₂ -C m ⁻² hr ⁻¹			
12 hours	ST	NT	ND		
Lower pressure	A 297±11.8 a b	A 273±2.7 b	A 339±2.3 a		
Ambient pressure	A 290±15.1 a	B 261±2.5 a	B 290±2.4 a		
Second week	mg CO ₂ -C m ⁻² hr ⁻¹				
4 hours	ST	NT	ND		
Lower pressure	A 226±19.4 a	A 200 ±6.2 a	A 226±10.5 a		
Ambient pressure	A 202±11.2 a	A 187 ±5.0 a	A 204±2.4 a		
	mg CO ₂ -C m ⁻² hr ⁻¹				
12 hours	ST	NT	ND		
Lower pressure	A 219±9.0 b	A 226±10.0 a b	A 280±4.8 a		
Ambient pressure	A 222±11.1 a	B 208±0.8 a	B 251± 5.3 a		

Table 3.3: Carbon dioxide fluxes (mg CO_2 -C m⁻² hr⁻¹) under lower (negative) and ambient pressures in laboratory conditions during 4 and 12 hour measurement period.

Table 3.4: Accumulated carbon dioxide emissions (mg CO_2 -C m⁻²) over 4, 12 and 16 hour measurement period under lower (negative) and ambient pressures in laboratory conditions.

First week	mg CO ₂ -C m ⁻²			
4 hours	ST	NT	ND	
Lower pressure	A 1433±39.3 a b	A 1235±11.5 b	A 1522±38.2 a	
Ambient pressure	B 1280±31.7 a	B 1079±48.0 b	B 1137±44.2 a b	
		mg CO ₂ -C m ⁻²		
12 hours	ST	NT	ND	
Lower pressure	A 3558±141.1 a b	A 3272±32.5 b	A 4072±27.2 a	
Ambient pressure	A 3477±181.7 a	B 3127±30.5 a	B 3474±28.3 a	
		mg CO ₂ -C m ⁻²		
16 hours	ST	NT	ND	
Lower pressure	A 4991±106.1 a	A 4507±41.7 b	A 5594±25.5 c	
Ambient pressure	A 4757±204.6 a	B 4207±78.5 a	B 4610±70.1 a	
Second week	mg CO ₂ -C m ⁻²			
4 hours	ST	NT	ND	
Lower pressure	A 904±77.7 a	A 799±24.6 a	A 902±42.1 a	
Ambient pressure	A 809±44.7 a	A 749±19.8 a	A 815±9.6 a	
	mg CO ₂ -C m ⁻²			
12 hours	ST	NT	ND	
Lower pressure	A 2633±108.4 b	A 2715±20.5 a b	A 3355±57.4 a	
Ambient pressure	A 2670±133.2 a	B 2491±9.5 b	B 3012±63.5 a	
		mg CO ₂ -C m ⁻²		
16 hours	ST	NT	ND	
Lower pressure	A 3537±184.5 a	A 3514±107.1 a	A 4257±79.3 a	
Ambient pressure	A 3479±177.8 a	B 3240±23.0 a	B 3827±72.9 a	

Each value in the table represents the mean of three replicates with standard errors (\pm) .

Capital letters represent pressure difference

Small letters represent tillage treatment difference

Means $(\pm S.E)$ followed by the same small letter in rows and same capital letter in column for the tested time intervals are not significantly different.

3.6.4 Discussion

Soil CO₂ flux measurements are very sensitive to pressure fluctuations (Fang and Moncrieff 1998). Most of the past studies reported how pressure differences between the outside and inside of the chamber (created due to air flow rate through the chamber and wind velocity) affects the soil CO₂ efflux (Hanson *et al.* 1993; Lund *et al.* 1999; Longdoz *et al.* 2000; Welles *et al.* 2001; Suleau *et al.* 2009). Results of the present study support the general perception that higher CO₂-C fluxes are observed under lower pressures due to high wind velocity. Irrespective of the three (ST, NT and ND) tillage treatments higher CO₂-C fluxes were measured with the onset of lower pressure than under ambient pressure throughout the two week study period.

A physical explanation of this could be that while a negative pressure gradient exists between air in the chamber and air contained in soil pores, soil air rich in CO₂ moves to equilibrate the pressure gradient. Also the lower CO_2 partial pressure in the chamber will result in a greater CO₂ diffusion gradient leading to higher initial fluxes. For the ST treatment significantly higher CO₂-C fluxes under lower pressure were observed only for the four hour measurement period during the first week. Thereafter, no significant differences were found between the CO₂-C fluxes at ambient and lower pressures. Loosening of soil resulted in the rapid decline in CO_2 concentration in the soil pores resulting in higher CO_2 -C fluxes only for the four hour measurement period. Similarly, in a field experiment, Reicosky et al. (2008) observed a rapid drop in the CO_2 concentration in a ploughed soil surface due to wind induced lower pressures and suggested the change in soil porosity following tillage results in rapid gas exchange leading to lower CO_2 concentrations in ploughed soils. Absence of a significant effect of lower pressure for the ST treatment during any other measurement period except for the four hour measurement period during the first week suggests that pressure fluctuations affect only the CO₂ transport but not production. In this experiment measurements were taken for 4 hour and 12 hour durations and no attempts were made to measure these losses on an hourly basis.

Subke *et al.* (2003) measured CO_2 fluxes in field conditions using an open dynamic chamber with an aim to model long term CO_2 fluxes, and emphasized that there was no need to include the effect of pressure fluctuations, as it affects only transport but not production. Moreover, averaged over time the net contribution of pressure related transport is zero as high fluxes due to negative pressure are followed by low fluxes due to positive pressure. Slow but continuous gas-exchange from the NT and ND treatments due to higher bulk densities and greater tortuosity of pore space in comparison to the ST treatment resulted in higher CO_2 -C fluxes under lower pressure. Results suggest that the effect of pressure fluctuations on soil CO_2 fluxes differ due to soil properties such as soil bulk density and porosity, with observed effects lasting for two weeks on NT and ND treatments.

3.6.5 Summary and conclusions

To better understand the impact of pressure fluctuations caused by wind velocity on soil CO₂ emissions from simulated tillage (ST) and No-tillage (NT) treatments, a two-week laboratory study was conducted. Carbon dioxide flux measurements were made using the alkali trap method from simulated tilled (ST) soils and No-tillage (NT) in-situ soil cores simultaneously maintained at ambient and lower pressures (a negative pressure of 20 millibars was created using an individual bubble tower for each chamber). Lower pressure had a significant effect on CO₂-C fluxes from the ST treatment only for the 4 hour measurement period of the first week. Thereafter, no significant differences between the two pressure treatments were observed. Tillage induced change in soil air porosity resulted in immediate exchange of CO_2 during the 4 hour measurement period of the first week, suggesting that pressure fluctuations affect CO₂ transport but not production. Compared to ST, NT and ND show significantly higher CO₂-C fluxes at lower pressure than ambient pressure for 4 and 12 hour measurement periods during the first week and for a 12 hour measurement period during the second week. The NT and ND treatments, which had higher bulk densities, lower air-filled pore spaces and greater tortuosity of pore space than the ST treatment, show an effect of pressure change (low pressure vs. ambient). This suggests that occasional change in wind induced pressure fluctuation can affect CO₂-C fluxes differently from soils that vary in soil bulk density and porosity.

The results of the present study suggest that atmospheric pressure fluctuations on soils may influence CO_2 fluxes from soils and the affect will vary with change in the soil bulk density and porosity. Changes in atmospheric pressure however, were found to have short term differential effects on CO_2 fluxes from simulated tilled soils. Therefore it was felt unnecessary to simulate the effects of occasional low pressure caused by the wind speed at the soil surface. Moreover, the focus of this project was to quantify the reductions in CO_2 emissions with Cross-slot No-tillage cultivation compared with full cultivation and develop either a soil test or decomposition model for quantitatively determining reductions in CO_2 losses with No-tillage. All further studies were conducted at ambient pressure conditions.

3.7 References

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Chapter-4

Carbon dioxide emissions from soils following rotary– and No-tillage seed bed preparation under field and controlled laboratory conditions

4.1 Introduction

No-tillage practice (NT) is globally recognised by scientists as a possible alternative to conventional tillage practice (CT) because it is highly beneficial in soil and water conservation and reduces production costs (Melero *et al.* 2011; Regina and Alakukku 2010; Moussa-Machraoui *et al.* 2010; Lal 2009; Christopher *et al.* 2009). Long-term No-tillage practice restores the soil organic carbon (SOC) lost as carbon dioxide due to conventional tillage practices thereby offsetting emissions by fossil fuel and alleviating concerns about projected global climate change (Blanco-Canqui and Lal 2008; Conant *et al.* 2007; West and Post 2002).

A vast international literature clearly demonstrates that CO_2 emissions after sowing are significantly less from No-tilled than conventionally tilled soils (Reicosky and Lindstrom 1993; Reicosky *et al.* 1997; Dao 1998; Rochette and Angers 1999; Alvarez *et al.* 2001; Al-Kaisi and Yin 2005; Bauer *et al.* 2006; Alvaro-Fuentes *et al.* 2008; Ussiri and Lal 2009; Morell *et al.* 2010). However, quantitative information on potential savings in soil CO_2 emissions with No-tillage compared to conventional tillage is limited in New Zealand. A single study conducted by Aslam *et al.* (2000) found no differences in CO_2 emissions from NT and CT soils throughout the crop growing period for summer maize and winter oats. The degree of soil disturbance by the 'Baker boot', a slot opener in the No-tillage drill used in that study, raises questions about the interpretation of the results. The 'Baker boot' slot opener underwent further development to become the current Cross Slot[®] opener. In the latest No-tillage drills, the 'Baker boot' has been replaced with an advanced Cross Slot[®] opener which causes negligible soil disturbance in comparison to the 'Baker boot' (Baker and Saxton 2007).

This necessitated the need to conduct a study to provide better scientific information on potential savings in soil CO_2 emissions with No-tillage under New Zealand conditions. Therefore this study was planned with the following objectives: 1) to quantify CO_2 emissions from simulated tillage (ST) and NT on a range of soils varying in physico-chemical properties under laboratory conditions and 2) to verify under field conditions the reliability of differences in CO₂ fluxes observed between ST and NT treatments from laboratory studies.

4.2 Material and methods

4.2.1 Laboratory study

4.2.1.1 Sites and soils

The five sites are located in the Manawatu region of North Island, New Zealand. These sites under annual crop rotation of pasture and cereals were established for Cross Slot[®] No-tillage cultivation over the last 2 to 15 years to reduce CO_2 losses from those that occur during conventional tillage practices. All five sites were on the farms using Cross Slot[®] No-tillage practice for seeding pasture and cereals. The location, classification and physico-chemical characteristics of the soils are given in Table 4.1.

	Glen Oroua	Tangimoana Kiwitea		Feilding	Sanson field site
location	40 ⁰ 18'58.33"S 175 ⁰ 21'46.40"E	40 ⁰ 17'33.25"S 175 ⁰ 17'16.95"E	40 ⁰ 3'56.79"S 175 ⁰ 43'10.87"E	40 ⁰ 15'50.88"S 175 ⁰ 37'23.82"E	40°14'34.33"S 175°21'27.07"E
рН (1:2.5)	5.8	5.7	6.1	6.0	6.0
CEC $(\operatorname{cmol}_c \operatorname{kg}^{-1})$	19.4	19.0	35.0	21.3	19.8
Olsen-P $(mg kg^{-1})$	33.9	45.8	14.7	14.2	21.5
SO_4-S (mg kg ⁻¹)	11.0	10.8	31.5	14.0	8.3
Total C (g kg ⁻¹)	43.5	43.3	85.0	30.3	39.7
Total N (g kg ⁻¹)	3.7	3.7	8.7	2.8	3.5
Bulk Density (Mg m ⁻³)	1.02	0.98	0.76	1.18	1.07
Soil type	Carnarvon black sandy loam	Carnarvon black sandy loam	Dannevirke silt loam	Ohakea stony silt loam	Ohakea silt loam
Soil Classification USDA	Typic Humaquept	Typic Humaquept	Typic Hapludand	Typic Epiaqualf	Typic Epiaqualf
NZ	Concretionary Sandy Gley soil	Concretionary Sandy Gley soil	Typic Orthic Allophanic soil	Typic Orthic Gley soil	Typic Orthic Gley soil
Sand (%)	71	57	15	21	17
Silt (%)	18	31	55	51	59
Clay (%)	11	12	30	28	24
No-tillage duration (years)	2	15	7	8	11
Clay mineralogy*	Micaceous	Micaceous	Allophanic	Smectitic	Smectitic

Table 4.1: Physico-chemical characteristics of the five soil sites (0-10cm).

Clay mineralogy* Claridge (1961); Saggar et al. (1996; 1999) and Hedley et al. (2000)

4.2.1.2 Collection of soil cores and simulation of tillage

Soil cores from all the sites were sampled for CO_2 measurements during December 2009 and January 2010 following the cultivation of pasture in late spring/early summer. All the four soil sites were sprayed with glyphosate herbicide prior to seeding. Two tillage treatments: simulated-tillage (ST) and No-tillage (NT) were compared; the third treatment was a non-disturbed (ND) control. The control treatment had no soil disturbance by tillage.

Thirty six *in-situ* soil cores (10 cm diameter, 10 cm depth) were collected at a soil depth of 0-10 cm from each site. Twenty four ND cores were collected before the start of field tillage operations from 4 different locations on each farm, six from each location. Twelve NT treatment cores were collected over the slots immediately after the Cross Slot[®] No-tillage drilling, three cores from each location. Each treatment comprised four replicates and each replicate was composed of three intact cores collected from one sampling location. Twelve out of twenty four ND cores were broken up in the laboratory to simulate tillage treatment. The ST treatment was formulated from three *in-situ* soil cores by emptying the soil, breaking it into pieces, thoroughly mixing and packing it in a plastic container (19 cm diameter; 10 cm high).

Carbon dioxide measurements for all the three treatments started after 2 hours of mixing the soils for the ST treatment. Moisture contents in the cores and container as sampled from the field were maintained throughout the experiment by weighing the soil cores and spraying the required amount of deionised water onto the surface. Measurements continued for 92 days (Glen Oroua), 83 days (Tangimoana), 81 days (Kiwitea), and 54 days (Feilding) until the emissions subsided and the differences between the treatments became negligible.

4.2.1.3 Carbon dioxide measurements

Carbon dioxide flux production was measured using the alkali trap method. Soil *insitu* cores and plastic containers filled with soil were placed in closed base static chambers and CO₂ measurements were taken on a daily basis at constant temperature (23° C) by placing a plastic petri dish containing 30 ml of 1M NaOH within each chamber. Chambers were sealed with air tight lids to control the leakage of carbon dioxide. After a 4 hour absorption period, the plastic petri dishes containing NaOH were removed and the total amount of CO₂ absorbed in each solution was determined by titrating NaOH with dilute 0.2M HCl. All the chemicals used were of analytical grade and the solutions were prepared using deionised water. The solution of 0.2M HCl was standardised against Na₂CO₃ as per the method outlined by Lambert *et al.* (1949). A working solution of 1.0M NaOH was prepared and the molarity was checked against standardised acid.

4.2.1.4 Calculation of CO₂ flux for NT cores

Carbon dioxide fluxes measured from the ND cores were used to calculate CO_2 fluxes from the actual area disturbed by the Cross Slot[®] opener in NT treatment cores. To calculate CO_2 flux from ST, NT and ND treatments the amount of CO_2 emitted from each chamber was divided by the surface areas of 3 *in-situ* cores. Further, CO_2 flux from the actual area disturbed by the Cross Slot[®] opener in NT treatment cores was calculated by determining the area of the portion disturbed by the Cross Slot[®] opener and then by subtracting that area from the total surface area of the core, giving the undisturbed surface area.

Total surface area of the core= 78.57 cm^2

Area of the core disturbed by the Cross Slot[®] opener=43.44 cm²

(Calculated from the Cross Slot[®] width=4.5cm)

Area of the core left undisturbed by Cross Slot[®] opener=35.13 cm²



Figure.4.1: Portion of the soil core disturbed by the Cross Slot[®] opener shown by the width of the Cross Slot[®].

Ratio of the core area disturbed and area left undisturbed by the Cross Slot[®] opener was determined by dividing the area disturbed by the Cross Slot[®] opener by the total surface area of the core (43.44/78.57=0.55).

Similarly, the ratio of the area left undisturbed was calculated (35.13/78.57=0.45).

The actual CO_2 flux contributed by the disturbed portion of the NT treatment core was then calculated from the measured CO_2 fluxes from the NT and ND treatments as:

 $NT_C \text{ flux} = \text{Calculated } NT_C \text{ flux } x \text{ } 0.55 + \text{ND}_C \text{ flux } x \text{ } 0.45$ $Calculated NT_C \text{ flux} = ((NT_C \text{ flux } - (ND_C \text{ flux } x \text{ } 0.45))/0.55)..... (Eq. 4.1)$ where

 NT_C flux = CO_2 flux from the NT treatment core

 ND_C flux = CO_2 flux from ND treatment core

The field CO_2 fluxes from the NT treatment were estimated by taking into account the proportions of disturbed and undisturbed field areas by the Cross Slot[®] No-tillage drill system.

Drill width=450 cm

Number of Cross Slot[®] openers in the drill=29

Portion disturbed by Cross Slot[®] openers= Number of Cross Slots[®] × Width of Cross Slot[®]

Ratios of the portion disturbed and undisturbed by Cross Slot® openers with one pass of drill: NT = (130.5/450) = 0.29; ND = (1-0.29) = 0.71 NT (0.29): ND (0.71).

Field NT CO₂ flux= Calculated NT_C flux×0.29+NDc flux×0.71..... (Eq. 4.2)

4.2.1.5 Soil moisture content (SMC)

The gravimetric SMC was measured at 0-10 cm depth. Replicated soil samples were collected from respective sites on the same date when soil cores were collected for carbon dioxide measurements. Field moist samples were weighed, oven-dried (105° C) to constant mass, and weighed again. The final mass (*Ms*), and the difference between the field moist and dry masses (*Mw*), were used to calculate the gravimetric SMC = (*Mw/Ms*) X 100. Gravimetric SMC (%) for the four soil sites were: Glen Oroua (46.1%); Tangimoana (41.9%); Kiwitea (60.6%); Feilding (30.8%) at the time of sampling the soil cores.

4.2.1.6 Cumulative CO₂ emissions

Cumulative CO_2 -C emissions for the whole measurement period were calculated by linear interpolations of the measured fluxes and integrated over time for all the soil sites.

4.2.2 Field study

Following CO_2 measurements from the intact soil cores collected from four different soil sites during the summer season (December 2009 and January 2010), CO_2 measurements were made in the field during the autumn (April to July 2010) and summer (November 2010 to February 2011) seasons.

4.2.2.1 Site and soil

The field experiment was carried out at a site near Sanson planted in barley (*Hordeum vulgare* L.) during summer and ryegrass (*Lolium multiflorum* L.) pasture during the autumn season. The site was sprayed with glyphosate herbicide prior to seeding. The location, classification and physico-chemical characteristics of the soil are given in Table 4.1.

4.2.2.2 Establishment of field experiment

The three tillage treatments compared during the field trial were: 1) tillage with a rotary tiller (RT) to a depth of 10 cm followed by bar harrow, 2) direct seeding with a Cross Slot[®] No-tillage drill (NT), and 3) a non-disturbed (ND) control. The control treatment had no soil disturbance by tillage.

A portion (length 90m; width 18m) of a 1 hectare paddock was selected to conduct the trial. To compare three treatments this experimental area was equally divided into three segments of 90m x 6m each. For gas measurements, PVC chambers (diameter 23.5 cm; height 21 cm) were randomly installed in each segment, immediately after the completion of tillage operations. Each chamber had an internal half-turn locking system and a greased Oring which forms a gas-tight seal when closed with a lid (Saggar *et al.* 2004). Each chamber was inserted 5 cm into the soil to avoid any leakage of CO₂ out of the chamber.

During the autumn (April to July 2010) seeding of pasture, the number of PVC chambers differed between the three treatments. A number of twelve, four and eight replicate chambers were installed in RT, NT and ND treatments, respectively. Chambers were installed over the slots after Cross Slot[®] No-tillage seeding. Carbon dioxide measurements were made for 110 days during the autumn season. To measure the CO₂ flux due to disturbance from rotary tilled soils, NaOH trap solutions were placed immediately after tillage operations (i.e. 0-4 hours) in four of the twelve chambers. In the remaining eight chambers NaOH trap solutions were placed two hours after the completion of tillage operations (i.e. 2-6 hours). In the NT and ND treatments, NaOH trap solutions were placed

two hours after the completion of tillage operations. No changes were made to the existing field set-up during the summer season.

During the summer seeding of barley the same three (RT, NT and ND) tillage treatments were compared and the number of PVC chambers did not differ between the three treatments. Eight replicate chambers were installed in each RT, NT and ND treatment to measure CO_2 losses and measurements were taken intermittently for 99 days. The NaOH trap solutions were placed two hours after the completion of tillage operations in all three treatments. The soil surface in and 1.0m around the chambers was always kept free of vegetation to avoid the influence of plants on CO_2 measurements as the objective was to quantify the short term C loss due to the decomposition of crop or pastures residues after tillage. Although the plant-free surface does not simulate standard agronomic practice, it does avoid the complication of plant and root respiration being confused with microbial respiration during the decomposition of residues. Measurements were normally taken between 9 am and 1pm.

A two hour period is required after the chamber installation for all the greenhouse gas fluxes to come to equilibrium (Field protocol, Personal Communication Surinder Saggar).

A parallel simulated tillage trial (i.e. taking CO₂ measurements from soil *in-situ* cores similar to the study conducted under laboratory conditions in December 2009 and January 2010 as in section 4.2.1) was conducted along with the field CO₂ measurements. During the autumn season, closed base chambers enclosing the soil cores were placed in the field. During the summer season chambers were placed in the laboratory at 23° C to compare the CO₂ emissions under controlled conditions with the field emissions. Moisture content in the soil cores was maintained at original levels at which they were sampled from the field i.e.15.8 per cent throughout the measurement period.

4.2.2.3 Carbon dioxide measurements

Carbon dioxide was measured using an alkali trap method similar to that used in the laboratory trial described in Section 4.2.1.3.

4.2.2.4 Soil temperature and moisture content

Soil temperature was measured with iButton data loggers (DS1921Z) which can measure a temperature range -5° C to $+26^{\circ}$ C with $\pm 1^{\circ}$ C accuracy. Temperature buttons were buried at 10 cm depth in the soil adjacent to the chambers for all three treatments. Buttons were programmed to measure soil temperature bi-hourly. Gravimetric SMC was measured at 0-10 cm depth on the replicated soil samples collected from the three tillage treatments on the day of CO₂ measurement as described in section 4.2.1.5.

4.2.2.5 Cumulative CO₂ emissions

Cumulative CO_2 -C emissions for each season were calculated by linear interpolations of the measured fluxes and integrated over time.

4.2.3 Collection of soil samples for physical and chemical analysis

Soils samples for physical and chemical analysis were collected in triplicate (0-10 cm soil depth) from all five sites on the same day as soil cores were collected for CO_2 measurements. Each replicate was composed of 12 cores (2.5 cm diameter) randomly collected across the field. Soil samples were air dried and ground to pass through a 2mm sieve for further analysis.

4.2.3.1 Physical analysis

Particle size distribution was determined by the pipette method of Claydon (1989). For soil bulk density determination, undisturbed soil cores (10 cm diameter, 10 cm depth) were taken and soil bulk density was calculated by dividing the mass of oven dried soil by the volume of the soil core.

4.2.3.2 Chemical analysis

Soil pH (1:2.5) and cation exchange capacity (CEC) was analysed according to Blackmore *et al.* (1987). Olsen P and SO₄-S were determined according to Olsen *et al.* (1954) and Blackmore *et al.* (1987). A portion of each sample was ground to pass through a 0.2 mm sieve for total carbon and nitrogen analysis using a LECO induction furnace (Blackmore *et al.* 1987).

4.2.4 Collection of climate data

Weather data was obtained from the National Climate Database interface (<u>http://cliflo.niwa.co.nz/</u>). For the study period, April 2010 to July 2010 and November 2010 to February 2011, daily air temperature and precipitation data was collected.



Figure.4.2: Air temperature and precipitation at the Sanson field site from April 2010 to February 2011.

4.2.5 Measurements of above- and below-ground residue inputs

Pasture residues were collected from all five soil sites. At each site three replicate plots measuring 1m x 1m were marked. Pasture grass was sprayed with glyphosate and after 10 days five soils cores (10 cm diameter) were taken to a depth of 10 cm from all the three replicates. To simulate the grazed conditions above-ground residues were cut to 2cm height. Roots and above ground parts of the pasture grass were then separated by gentle shaking and wet sieving. Oven dried (65 0 C) samples of above ground pasture and root biomass from each core were used to calculate the residue yield.

4.2.6 Statistical analysis

An analysis of variance using SAS software (9.1) was performed on CO_2 fluxes from the three tillage treatments using the General Linear Model (GLM) procedure. Mean comparisons between CO_2 fluxes were done using Fisher's least significant difference (LSD) at 5% level of significance. Pearson correlation and regression coefficients were determined using SAS software (9.1) to determine the relationship between the total amount of CO_2 lost till day-54 under laboratory conditions and different soil properties. All the soil replicates were used in correlation and regression analysis

4.3 Results

4.3.1 Laboratory study

In the laboratory study, the trends of the CO₂-C fluxes were similar in both tillage treatments and non-disturbed control in all the soil sites, with high peaks of CO₂-C observed for the first few days and low rather constant CO₂-C fluxes afterwards. The daily CO₂-C fluxes (kg CO₂-C ha⁻¹ d⁻¹) for the ST, NT and ND treatments from all the four soil sites are shown in Figures 4.3 a, b, c, d. During the 92 days measurement period for the Glen Oroua site (Figure.4.3.a), significantly higher CO₂-C fluxes from the ST than the NT and ND treatments were observed for the first 10 days; thereafter the differences in the three treatments narrowed, with CO₂-C fluxes from NT and ND gradually becoming larger than ST. Significantly higher CO₂-C fluxes from NT than ND were observed only on day-1 of the measurement period. From day-33 of the measurement period CO₂-C fluxes from the NT and ND treatments were either similar or higher than the ST treatment. For the Tangimoana site, during the 83 days of the measurement significantly higher CO₂-C fluxes from the ST than the NT and ND treatments were observed for the first 24 days and thereafter the differences in CO₂-C fluxes narrowed, with CO₂-C fluxes from ND gradually becoming larger than NT and ST from day-41 (Figure.4.3.b). No significant differences were observed between CO₂-C fluxes from NT and ND throughout the measurement period. During the 81 days of measurements for the Kiwitea site, the ST treatment emitted significantly higher CO₂-C than the NT treatment, but only for the first 6 days. From day-20 onwards CO₂-C fluxes from the ND treatment become larger than from the NT and ST treatments (Figure 4.3.c). For the Feilding site (Figure.4.3.d), the CO₂-C fluxes were significantly higher from ST than NT and ND for only the first 5 days of the 54 day measurement period. From day-37 of the measurement period, fluxes from ND were either similar or higher than from the ST treatment.

Comparing all the soil sites, the Kiwitea soil site produced higher initial CO_2 -C fluxes and the Feilding site produced lowest initial CO_2 -C fluxes from all the treatments in comparison to the other soil sites. The accumulated CO_2 -C emissions from the Glen Oroua, Tangimoana, Kiwitea and Feilding soil sites from ST were 2303, 3264, 2905, 1086 kg CO_2 -C ha⁻¹; NT were 2349, 2871, 2789, 973 kg CO_2 -C ha⁻¹ and ND were 2490, 3023, 3008, 953 kg CO_2 -C ha⁻¹ (Figure. 4.4). None of the soils from the four sites showed any significant difference in the accumulated CO_2 -C emissions between the ST, NT and ND treatments. The

amounts of residue input were highest at Tangimoana (8.3 Mg ha⁻¹) followed by Kiwitea (8.0 Mg ha⁻¹), Glen Oroua (7.5 Mg ha⁻¹) and Feilding (3.1 Mg ha⁻¹) sites.

Amongst sites, the amount of CO_2 -C lost was maximum from the Tangimoana site and minimum from the Feilding site. One can argue that it might be due to a shorter measurement period of 54 days but when the total CO_2 -C lost from all the soils till day-54 were compared, even then the CO_2 -C loss was lowest from the Feilding site (Table 4.2).

Table 4.2: Cumulative average CO_2 -C emissions from simulated tillage (ST), No-tillage (NT) and Non-Disturbed (ND) treatments from Glen Oroua, Tangimoana, Kiwitea and Feilding site soils until day-54.

Sites	Cumulated CO ₂	lated CO_2 -C emissions (kg CO_2 -C ha ⁻¹)			
	ST	NT	ND		
Glen Oroua	1822	1730	1821		
Tangimoana	2708	2330	2452		
Kiwitea	2468	2318	2521		
Feilding	1086	973	953		

Three soil types were used in the laboratory study; they varied in texture, total carbon content, gravimetric soil moisture content (GMC) and the amount of plant and root residues incorporated. A correlation analysis was conducted to relate these factors to the total amount of CO_2 -C lost till day-54 from the ST treatment for all the four soil sites. As the amount of residue incorporated and moisture contents were similar in all three tillage treatments in each soil, only the ST treatment was considered for the correlation analysis. For the correlation analysis soil types were converted into indicator variables.

Correlation analysis showed that except for soil type, all other factors were significantly correlated with the total amount of CO_2 -C lost until day-54 from the ST treatment (Table 4.3). Therefore soil type was not included in multiple regression analysis which was conducted to find out which of these three factors i.e. total carbon content, gravimetric soil moisture content (GMC) and amount of plant and root residue incorporated have a significant effect on CO_2 -C emissions. The multiple regression (Table 4.4) showed that out of the three factors, residues incorporated had a significant effect on the CO_2 -C lost from the ST treatment.

Table 4.3: Pearson correlation coefficients (R) of the total amount of CO_2 -C lost until day 54 during laboratory incubation from the ST treatment and different soil factors of the four test soils.

Soil factors	R
Gravimetric moisture content (%)	0.57*
Total carbon content (g kg ⁻¹)	0.57*
Residues incorporated (Mg ha ⁻¹)	0.83**
Soil type	0.38

R: Correlation coefficient

*Correlation coefficient significant at p < 0.05

**Correlation coefficient significant at p < 0.001

Table 4.4: Multiple linear regression model between soil CO₂-C emissions till day 54 during laboratory incubation and gravimetric moisture content (GMC), total carbon content (TC) and residues incorporated (RES) for the ST treatment on the four test soils.

Tillage treatment	Multiple linear regression	\mathbf{R}^2
ST	668 - 18.6GMC + 12.3TC + 232RES**	0.72

R²: Coefficient of determination

**Coefficient of determination significant at p < 0.001



Figure.4.3: Daily average CO₂-C fluxes as influenced by tillage (ST, Simulated tillage; NT, No-tillage and ND, Non-Disturbed) from (a) Glen Oroua, (b) Tangimoana, (c) Kiwitea and (d) Feilding site soils. Each value represents the mean of four replicates and vertical bars above represent LSD (P < 0.05) for comparison among tillage treatments where significant differences were found.



Figure.4.4: Cumulative average CO_2 -C emissions from simulated tillage (ST), No-tillage (NT) and Non-Disturbed (ND) treatments from Glen Oroua, Tangimoana, Kiwitea and Feilding site soils along with standard errors (±) shown by vertical bars. N.S. stands for not significant

4.3.2 Field study

During the autumn season CO₂-C fluxes were measured continuously for the first 27 days and intermittently on days 36, 43, 49, 76 and 110 (Figure.4.5.a). Daily CO₂-C fluxes ranged from 17.5-44.0 kg CO₂-C ha⁻¹d⁻¹ for RT; 15.6-38.0 kg CO₂-C ha⁻¹d⁻¹ for NT and 15.1-35.4 kg CO₂-C ha⁻¹d⁻¹ for the ND treatment. During the 110-day measurement period in the autumn season RT gave higher daily CO₂-C fluxes than NT and ND treatments. Significantly higher CO₂-C fluxes from RT than NT and ND treatments were observed in 25 out of 32 daily CO₂-C measurement events. No significant differences were observed between CO₂-C fluxes from NT and ND treatments. During the autumn season measurement period the cumulative amount of rainfall received was 288mm and soil temperature ranged between 4.7 and 16.2°C. An early burst of CO₂ emission was measured during the autumn season only in the RT treatment. Carbon dioxide flux immediately after tillage (i.e. 0-2 hours/ t-0 hours) (56.8 kg CO₂-C ha⁻¹d⁻¹) was significantly higher than flux measured after 2 hours (i.e. 2-6 hours/ t-2 hours) (41.5 kg CO₂-C ha⁻¹d⁻¹) (insert in Figure.4.5.a). The disturbance caused by tillage in the field significantly elevated the CO₂ fluxes for the first four hours only (0-4

hours). Daily rates estimated from the amount of CO_2 trapped during this period tends to overestimate daily CO_2 fluxes because the CO_2 trapped between 2-6 hours after tillage was considerably lower and more similar to the rates observed at 24 and 48 hours after tillage. The CO_2 flux rates from all twelve replicate chambers were used to estimate the average daily for the remainder of the experiment.

During the summer season, CO_2 -C fluxes were measured on alternate days for the first 4 weeks and the frequency of subsequent measurements was reduced to 3, 4 and 7 days for the following 2 months (Figure.4.5.b). Daily CO₂-C fluxes during the summer season ranged from 13.3-109.4 kg CO₂-C ha⁻¹d⁻¹ for RT; 10.9-93.2 kg CO₂-C ha⁻¹d⁻¹ for NT and 10.5-89.0 kg CO₂-C ha⁻¹d⁻¹ for ND treatment. Out of 30 CO₂-C measurement events during the summer season, significantly higher CO₂-C fluxes from the RT than the NT treatment were observed for 17 measurement events. The majority of these significant differences were observed during the first 35 days after tillage. Significant differences between CO₂-C fluxes from NT and ND treatments were observed only on day one. Carbon dioxide fluxes during the summer season showed variation depending on the GMC's of the soil (Figure. 4.6).

Gravimetric moisture content (GMC) ranged between 19.5% and 38.7% during the autumn season and varied from 9.5% to 39.8% during the summer season across all three RT, NT and ND treatments (Figures. 4.7 a, b). On average the GMC's during both seasons were in the order ND > NT > RT. Such an effect could be related to easier water percolation in rotary tilled soils than No-tillage and more compact soil conditions in the non-disturbed control treatment. Additionally, presence of the surface crop residues in the NT and ND treatments decreases the evaporative loss of water from the NT and ND treatments compared to the RT treatment (Jarecki and Lal 2003). Average GMC was higher during the autumn season in comparison to the summer season. High GMC's in the summer season were observed only in the event of rainfall (Figures.4.2 & 4.7. b). Soil temperatures ranged between 4.7 and 16.2°C during autumn and 16.5 and 23.6°C during the summer season (Figures. 4.8 a, b). The soil temperatures in autumn were lower than those observed in summer. However, there were no differences in soil temperature among the tillage treatments during both seasons.

The total amounts of CO₂-C emitted from RT, NT and ND treatments during the autumn and summer seasons are shown in Figures. 4.10. a, b. Accumulated CO₂-C emissions from RT, NT and ND treatments during the autumn season were 2580, 2215 & 2405 kg CO₂-C ha⁻¹ and 3330, 2877 & 2807 kg CO₂-C ha⁻¹ during the summer season. The total amount of CO₂-C emitted from RT was significantly higher than for NT and ND treatments during the

autumn and summer seasons. The amounts of CO_2 -C emitted from NT and ND treatments did not differ significantly during both the autumn and summer seasons. The residue inputs were higher in the summer (7.2 Mg ha⁻¹) than in the autumn cultivation (5.3 Mg ha⁻¹).

The linear regression analysis relating CO_2 -C fluxes to soil temperature and moisture contents in respective tillage treatments during the summer and autumn seasons are given in Table 4.5. No relationship was observed between the CO_2 -C fluxes and soil temperature during both seasons. No relationship was observed between the CO_2 -C fluxes and soil moisture during the autumn season, but a weak and significant relationship was observed during the summer season.

Table 4.5: Linear regression coefficients of determination (\mathbb{R}^2) comparing the CO₂-C fluxes from the RT, NT and ND treatments with gravimetric moisture content (GMC %) and soil temperature during the autumn and summer seasons.

Soil factors	Coefficient of determination Autumn season			Coefficient of determination Summer season		
	RT	NT	ND	RT	NT	ND
Soil temperature	0.02	0.01	0.01	0.003	0.003	0.004
GMC (%)	0.18	0.01	0.02	0.25*	0.29*	0.16*

R²: Coefficient of determination

*Coefficient of determination significant at p < 0.05



Figure.4.5: Daily average CO₂-C fluxes as influenced by tillage (RT, Rotary tillage; NT, No-tillage and ND, Non-Disturbed) during (a) autumn and (b) summer season from the Sanson field site. Each value represents the mean of twelve replicates for ST, four replicates for NT, eight replicates for ND treatment for autumn season and eight replicates for all three treatments during the summer season with standard errors (\pm) shown by vertical bars. Vertical bars above represent LSD (P < 0.05) for comparison among tillage treatments where significant differences were found. Insert in Figure.4.5.a shows the CO₂-C flux immediately after tillage (t-0 hours) and after 2 hours (t-2 hours) of the tillage event.



Figure.4.6: Daily average CO₂-C fluxes from the rotary and No-tilled soils during the summer season from the Sanson field site affected by soil GMC.



Figure.4.7: Gravimetric moisture content (GMC %) at 0-10 cm depth during 4 hour CO_2 measurements (a) autumn and (b) summer season.



Figure.4.8: Soil temperature at 0-10 cm depth for 4 hour CO_2 measurement period during (a) autumn season and (b) summer season.

In conjunction with field measurements, CO_2 -C measurements were repeated with *in*situ soil cores placed under field conditions during the autumn season and under laboratory conditions during the summer season with simulated tillage treatments as applied in earlier laboratory studies during the summer season (December 2009 and January 2010 as in section 4.2.1). Measurements continued for 110 days during the autumn season and 99 days for the summer season.

The daily CO₂-C fluxes from ST, NT and NT treatments during the autumn and summer seasons are shown in Figures 4.9.a, b. During the autumn season CO₂-C fluxes ranged from 18.4-55.1 kg CO₂-C ha⁻¹d⁻¹ for ST treatment; 13.3-44.2 kg CO₂-C ha⁻¹d⁻¹ for NT treatment and 12.4-40.1 kg CO₂-C ha⁻¹d⁻¹ for ND treatment. The total amount of CO₂-C lost during the autumn season was significantly higher in ST (2960 kg CO₂-C ha⁻¹) than NT (2149 kg CO₂-C ha⁻¹) and ND (2027 kg CO₂-C ha⁻¹) treatments (Figure.4.10.a). No significant differences between NT and ND treatments were observed.

During the summer season CO₂-C fluxes ranged from 11.5-121.0 kg CO₂-C ha⁻¹d⁻¹ for ST treatment; 12.0-75.7 kg CO₂-C ha⁻¹d⁻¹ for NT treatment and 11.1-52.8 kg CO₂-C ha⁻¹d⁻¹ for ND treatment. The cumulated CO₂-C emissions from ST (2253 kg CO₂-C ha⁻¹) were higher than NT (2023 kg CO₂-C ha⁻¹) and ND (1860 kg CO₂-C ha⁻¹) treatments during the summer season. However, no significant differences were observed between ST, NT and ND treatments (Figure.4.10.b).

The CO₂-C emissions measured from *in-situ* soil cores during the autumn season were similar to those reported from the field measurements (~2500 kg CO₂-C ha⁻¹); however, a similar trend was not observed during the summer season. Under field conditions CO₂-C emissions (Figure 4.10.b) were ~3000 kg CO₂-C ha⁻¹ compared with ~ 2000 kg CO₂-C ha⁻¹ for laboratory simulated conditions during the summer season.



Figure.4.9: Daily average CO₂-C fluxes as influenced by tillage (ST, Simulated tillage; NT, No-tillage and ND, Non-Disturbed) during (a) autumn under field conditions and (b) summer season under laboratory conditions. Each value represents the mean of four replicates and vertical bars above represent LSD (P < 0.05) for comparison among tillage treatments where significant differences were found.



Figure.4.10: Cumulative average CO₂-C emissions as influenced by tillage (ST, Simulated tillage; NT, No-tillage and ND, Non-Disturbed) during (a) autumn and (b) summer season with standard errors (\pm) shown by vertical bars. Letters indicate statistically significant differences (p < 0.05). N.S. stands for not significant.

4.4 Discussion

4.4.1 Laboratory study

Carbon dioxide measurements from the soils collected from the four different sites were carried out at constant laboratory temperature (23°C). Soil moisture contents of the respective soils were maintained at their original level at which they were sampled from the field. Significantly higher initial CO₂-C fluxes from ST than NT and ND treatments could be attributed to physical release of the CO₂ entrapped in the soil pores followed by microbial decomposition of easily decomposable organic matter (Curtin et al. 2000; Elder and Lal 2008). Differences in the CO₂-C fluxes between ST and NT treatments ceased when tillage-stimulated decomposition diminished, likely due to the exhaustion of easily decomposable residue substrates. Irrespective of the soil sites higher CO₂-C fluxes from ND than ST and NT treatments were observed after a few days of experiment initiation. This could be due to decrease in CO₂ concentrations in ST and NT soil pores due to rapid gas exchange or cessation of microbial decomposition due to exhaustion of easily decomposable residue substrates. Reicosky et al. (2008) compared CO₂ concentrations in mould board ploughed and NT soils under field conditions and observed higher CO₂ concentrations in NT soils (about 3.3%) than mould board ploughed soils (about 1.4%) suggesting higher CO_2 concentrations in least disturbed soils.

As the experiment was conducted under controlled conditions, the differences in the total amount of CO₂-C lost during the measurement periods for the four test soils were attributed to differences in soil texture, gravimetric moisture content, total carbon content, and the amount of plant and root residues incorporated, respectively. The total amounts of CO₂-C lost during the entire measurement periods from different soils irrespective of the tillage treatments are affected by the amounts of residue incorporated. A multiple regression of cumulative CO₂-C emissions over days-54 from the ST treatment for the four soils with gravimetric soil moisture content, total carbon content and amount of residues incorporated also confirmed that amount of residues incorporated was an important factor that affected the total amount of CO₂-C lost from soils (Table. 4.4). The total amount of CO₂-C lost from the three tillage treatments did not show any significant differences in all the four soils. Lack of variation in soil moisture and temperature conditions in the laboratory could explain why the differences in total amount of CO₂-C lost from different tillage treatments were not significant.

Although the differences between cumulative CO_2 -C emissions from ST, NT and ND treatments were not significant, the amount of CO_2 -C lost from the ND treatment was either similar or higher than the NT treatment in all four soil sites (Glen Oroua, Tangimoana, Kiwitea and Feilding) and higher even than the ST treatment in the Glen Oroua and Kiwitea sites (Figure 4.4). Higher CO_2 -C fluxes from the ND than ST and NT treatments after (few days) experiment initiation were the cause of the higher total CO_2 -C emissions from ND than NT and ST treatments.

4.4.2 Field study

The early burst of CO₂-C after rotary tillage (insert in Figure.4.5.a) measured during the autumn season was likely due to an increase in the air transport coefficient resulting from soil loosening. Rochette and Angers (1999) measured significantly higher CO₂ fluxes immediately after mould board ploughing than in undisturbed soils and used the term *degassing* to explain the increase in CO₂ fluxes immediately following tillage resulting from changes in the physical characteristics of tilled soils. *Degassing* means the loss of CO₂ from loosened soils due to change in bulk density and air filled porosity. Reicosky & Lindstrom (1993); Reicosky *et al.* (1997) and Kessavalou *et al.* (1998) concluded that a sharp increase in CO₂ flux immediately after tillage was due to the physical release of the CO₂ entrapped and accumulated in soil pores from previous microbial activity. Comparing different tillage treatments, Al-Kaisi and Yin (2005) observed a decrease of 52 to 68% in CO₂ fluxes within the first two hours after tillage operations which was greater than the reductions of 26.9% observed within 2 hours of the tillage operation in this study.

Significantly higher CO₂ emissions were observed from rotary tilled soils than the Cross Slot No-tillage seeded soils during the summer and autumn seasons. These results were in accordance with the previous studies conducted on comparison of contrasting tillage systems on CO₂ emissions from a range of cropping soils (Table 2.2 Chapter-2). The average difference between the CO₂-C emissions from the RT and NT treatments during autumn and summer were 3.4 and 4.7 kg CO₂-C ha⁻¹d⁻¹ (calculated by dividing the cumulated CO₂-C emissions during the summer and autumn seasons by the number of days for which measurements were made), which are similar to the differences reported by the following studies reported in Table 2.2 Chapter-2; Dao (1998); Ellert and Janzen (1999); Al-Kaisi and Yin (2005); Morell *et al.* (2010). Significantly higher daily CO₂-C fluxes from RT than NT and ND treatments during the summer and autumn seasons (Figures 4.5.a & b) could be attributed to accelerated soil organic matter decomposition due to tillage management.

Tillage incorporates the crop residues into the soil profile, breaks soil aggregates and modifies the soil environment i.e. soil temperature, moisture and aeration, resulting in enhanced soil organic matter decomposition (Alvarez *et al.* 2001; Six *et al.* 2002; Bauer *et al.* 2006). The CO₂ fluxes measured during this study period were similar to those reported by Aslam *et al.* (2000) under contrasting tillage systems in New Zealand conditions also using the absorption technique.

Daily CO₂-C fluxes during the summer season were limited by soil moisture status. Irrespective of the tillage treatments, higher flux rates were measured on day-3 compared to the fluxes measured on day-1 because of an increase in soil moisture content (Figure 4.6) following rainfall on day-2 of the measurement period (Figure 4.2). Similar peaks in CO₂-C fluxes were observed on day-30 and day-68 of the measurement period with increase in soil moisture content due to rainfall events. Bauer *et al.* (2006); Alvaro-Fuentes *et al.* (2008); Akbolat *et al.* (2009) and Morell *et al.* (2010) reported similar increases in CO₂-C fluxes after rainfall events. Akinremi *et al.* (1999) suggested that higher CO₂-C fluxes after a rainfall event was due to displacement of CO₂ from soil pores to the atmosphere due to water filling the soil pores, followed by an increase in microbial activity. In this study, rates of CO₂-C fluxes remained high as long as the soil remained moist due to rainfall events, and fell as soil dried.

The linear regression relating daily CO₂-C fluxes from RT, NT and ND treatments with soil temperature and moisture content during the autumn and summer seasons showed that only soil moisture had a significant effect on daily CO₂-C fluxes (Table 4.5) but only during the summer season. However, other researchers concluded soil temperature was a major factor influencing soil CO₂-C fluxes (Buyanovsky *et al.* 1986; Franzluebbers *et al.* 1995; Frank *et al.* 2002; Al-Kaisi and Yin 2005). After reviewing the interactive effects of soil physical factors and biological processes on soil atmosphere gas exchange, Smith *et al.* (2003) concluded that CO₂ production by aerobic respiration is temperature driven but becomes moisture dependent as soil dries. This could explain why a significant relationship was observed between daily CO₂-C fluxes and soil moisture during the summer season.

During the autumn season, the CO_2 -C flux measurements were made during April to July, soil temperatures were relatively low i.e. varied between 4.7 and 16.2°C and soil moisture was generally stable over that period. This could explain why no significant effect was found between CO_2 -C fluxes and soil temperature and moisture.
However, the complex process of CO_2 production and release from soils could not be explained without taking into account the decline of crop residues with time. This complex relationship is explained in Chapter-6.

For the entire study period, irrespective of the tillage treatments, higher peaks of CO_2 -C fluxes were observed during the summer season in comparison to the autumn season. Bauer *et al.* (2006) observed a two fold increase in CO_2 fluxes from conventional and conservative tilled soils during summer compared to spring and autumn seasons. Results of the present study are consistent with the previous seasonal studies (Omonode *et al.* 2007; Akbolat *et al.* 2009; Iqbal *et al.* 2008; Ussiri and Lal 2009). Generally, air and soil temperature controlled the seasonal variations in CO_2 fluxes across all tillage treatments with high fluxes during summer when temperatures were high resulting in high biological activity, and low fluxes during autumn/winter when temperatures are low resulting in low biological activity.

4.4.3 Comparisons of CO₂ emissions during laboratory and field studies

The magnitude of CO₂-C emissions from RT/ST, NT and ND treatments both in field and *in-situ* soil cores were similar (~2.5 Mg CO₂-C ha⁻¹) (Figure 4.10.a) during the autumn season, but during the summer season the magnitude of CO₂-C emissions from RT/ST, NT and ND treatments both in the field and under laboratory conditions were not similar. Under field conditions CO₂-C emissions (Figure 4.10.b) were ~3.0 Mg CO₂-C ha⁻¹ compared with ~ 2.0 Mg CO₂-C ha⁻¹ for laboratory simulated conditions.

During the autumn season *in-situ* soil cores were placed in the field and were exposed to similar variable moisture and temperature conditions that might have resulted in similar magnitudes of CO₂-C emission. During the summer season *in-situ* soil cores were placed under laboratory conditions. The lower CO₂-C emissions measured under laboratory conditions than by field chambers may have been due to: a) lower soil moisture content (15.8% GMC) at the time of collection of the soil cores used in the laboratory compared with variable moistures in the field or b) contribution of CO₂ from the deeper soil layers (> 10 cm) in field chambers may have resulted in higher emissions in the field chambers.

4.5 Summary and conclusions

Carbon dioxide (CO₂) emissions were measured for up to 3 months in the laboratory from four soils varying in physico-chemical properties and one soil in the field (autumn and summer seasons) using the alkali trap method following Cross Slot[®] No-tillage cultivation, simulated conventional tillage (ST) and non-disturbed (ND) control. The total amount of

 CO_2 emitted from the four soils under laboratory conditions ranged between 1086 and 3264 kg CO_2 -C ha⁻¹ for ST, 973 and 2871 kg CO_2 -C ha⁻¹ for NT and 953 and 3023 kg CO_2 -C ha⁻¹ for the ND treatment. In general, three out of four soils lost more CO_2 from the ST treatment (between 113 and 393 kg CO_2 -C ha⁻¹) than they did for the NT treatment.

Similarly, in the field the total CO₂-C emissions were significantly higher from RT (2580 kg CO₂-C ha⁻¹) than NT (2215 kg CO₂-C ha⁻¹) treatment during the autumn season, and from RT(3330 kg CO₂-C ha⁻¹) compared with NT (2877 kg CO₂-C ha⁻¹) during the summer season. Results of the field study suggests that conversion of pasture to cropping and cropping to pasture resulted in a net annual conservation of 818 kg C ha⁻¹ for NT soils.

The average differences between the CO_2 -C emissions from the RT and NT treatments during autumn and summer were comparable to the differences reported by the previous studies comparing the CO_2 -C emissions from tilled and No-tilled soils. Carbon dioxide fluxes during the summer season showed variation depending upon the rainfall and GMC's of the soil. Carbon dioxide measurements made from both the field chambers and *in-situ* soil cores following RT/ST or NT treatments were similar in magnitude (~2.5 Mg CO₂-C ha⁻¹) during the autumn season. During the autumn season *in-situ* soil cores were placed in the field and were exposed to similar variable moisture and temperature conditions which resulted in the similar magnitude of CO₂-C emissions from both field and *in-situ* soil cores.

However, during the summer season the magnitude of CO_2 emissions from RT/ST, NT and ND treatments both in field and laboratory conditions were not similar. Under field conditions carbon dioxide emissions were ~3.0 Mg CO₂-C ha⁻¹ compared with ~ 2.0 Mg CO₂-C ha⁻¹ for laboratory simulated conditions during the summer season. During the summer season soil cores were placed under laboratory conditions and were maintained at constant temperature and moisture content. Lack of variation in soil moisture and temperature in laboratory incubated soil cores resulted in the difference in magnitude of CO₂ emissions between field and soil cores. Therefore the concept that soil cores from the field being brought into the laboratory and tillage-treatment simulated to give reliable estimates of CO₂ emissions under field conditions appears to be unworkable.

The present study aimed to quantify CO_2 loss due to microbial decomposition of crop and pasture residues after tillage, which was the key reason why the soils under study in both the field and laboratory were left bare without root activity for the measurement periods.

Overall, Cross Slot No-tillage seeding of barley and pasture resulted in a net annual conservation of 818 kg C ha⁻¹ in soil which otherwise would have contributed 3.0 Mg CO_2 to the atmosphere.

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Chapter-5

Influence of soil type and rates of residue addition on decomposition of residues in soils under controlled laboratory conditions and the relationship of CO₂ emitted during the entire incubation period with labile soil C fractions

5.1 Introduction

Soil organic carbon (SOC) is the traditional measure of soil organic matter (SOM) (Skjemstad *et al.* 2006), and an important indicator of soil quality due to its influence on soil physical, chemical and biological properties (Saggar *et al.* 2001; Chen *et al.* 2009) The SOC pool is comprised of labile and recalcitrant/passive C pools with varying turnover rates (Haynes 2005).

The labile C pools, which turn over rapidly, respond quickly to land use change and soil management practices (Benbi *et al.* 2012; Toosi *et al.* 2012); and it has been suggested that they are the early indicators of SOC change (Xu *et al.* 2011). For this reason some C pools which respond quickly to agricultural management practices (e.g. microbial biomass-C, hot water extractable-C and oxidisable organic-C) may be able to be used to detect changes in SOC and therefore be used to predict CO_2 losses from soils.

In the earlier laboratory and field studies (Chapter-4), CO_2 measurements were made from soils varying in texture (silt loam and sandy loam), moisture contents, and containing different amounts of residue. However it was difficult to ascertain the effect of texture and residue inputs on CO_2 emissions due to the confounding effects of soil texture, moisture and amount of residue in these soils. Therefore, a laboratory incubation study was planned:

- To evaluate the effect of soil type and residue input rates on residue decomposition; in order to establish a relationship between soil texture, residue input and CO₂ losses, and
- To identify the labile C pools contributing to these losses and establish a relationship between the oxidisable-C pool and CO₂ losses.

5.2 Material and methods

5.2.1 Soils

The four soils used were the Carnarvon black sandy loam (Carnarvon SL), Ohakea stony silt loam (Ohakea stony SiL), Ohakea silt loam (Ohakea SiL), and Horotiu silt loam (Horotiu SiL). Soil classes and relevant chemical properties are given in Table 5.1; chemical analysis was carried out by the procedures outlined in section 4.2.3.2 (Chapter-4). Three soils (Carnarvon SL, Ohakea stony SiL, Ohakea SiL) came from three different field sites in the Manawatu region and one soil (Horotiu SiL) came from the Waikato region in the North Island of New Zealand. Soils from the Manawatu region were used in the laboratory study (Chapter-4) for measuring CO_2 fluxes. From each field site, four replicate surface soil (0-10 cm) samples were taken, and for each replicate 100 soil cores (2.5 cm diameter) were taken and pooled (sampling avoided recent dung and urine patches). Soil samples were collected during September 2010 i.e. early spring. The samples were sieved (< 2 mm) in the field moist state and stored at 4°C for about 10 to 15 days. These fresh, field-moist, sieved samples were used in the decomposition studies.

			-	-
	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL
pH	5.7	6.0	6.0	5.8
CEC $(\text{cmol}c \text{ kg}^{-1})$	19.0	21.3	19.8	35.0
Olsen-P (mg kg ⁻¹)	45.8	14.2	21.5	16.7
SO_4-S (mg kg ⁻¹)	10.8	14.0	8.3	10.3
Total C (mg C g^{-1} soil)	43.8	28.8	38.6	50.3
Total N (mg C g ⁻¹ soil)	3.7	2.8	3.5	5.2
Soil type	Carnarvon black sandy loam	Ohakea stony silt loam	Ohakea silt loam	Horotiu silt loam
Soil classification USDA	Typic Humaquept	Typic Epiaqualf	Typic Epiaqualf	Typic Hapludand
NZ	Concretionary Sandy Gley soil	Typic Orthic Gley soil	Typic Orthic Gley soil	Typic Orthic Allophanic soil
Particle size distribution Sand (%)	57	21	17	31
Silt (%)	31	51	59	42
Clay (%)	12	28	24	27
Field capacity moisture (%)	49.5	40.5	50.7	66.4
Clay mineralogy*	Micaceous	Smectitic	Smectitic	Amorphic

Table 5.1: Chemical characteristics of the four soils (0-10cm) used in the incubation study.

Clay mineralogy* Claridge (1961); Saggar et al. (1996; 1999) and Hedley et al. (2000)

5.2.2 Plant material

Ryegrass (*Lolium multiflorum* L.) pasture residues were collected from the three sites in the Manawatu region. At each site, three replicate plots measuring 1m x 1m were marked. Pasture grass was sprayed with glyphosate herbicide and after 10 days five soils cores (10 cm diameter) were taken to a depth of 10 cm from all three replicate plots. To simulate the grazed conditions above-ground, plant residues were cut to 2 cm in height. Roots and aboveground residues of the pasture grass were then separated by gentle sieving and stored at 4°C. Residues collected from all three sites were thoroughly mixed and chopped (< 2 cm in length) using a pair of scissors. Residues on dry weight basis contain 42.6%C, 1.35%N, 32.2% cellulose, 32.5% hemi cellulose, and 7.9 % lignin. Cellulose, hemicellulose and lignin contents were determined by the procedure outlined by Robertson and van Soest (1981).

5.2.3 Decomposition study

The soils were adjusted to 80% of their field capacities. Four replicate soil samples equivalent to 25 g (oven dry) were mixed thoroughly by hand with 2.0, 2.6, 3.25 g fresh pasture residues in a 55 ml plastic container, amounting to a concentration of 5.11, 6.81 and 8.51mg C g⁻¹ oven-dried soil or 12, 16 and 20 Mg dry matter residue ha⁻¹ to 10 cm depth, respectively. A control treatment with no residue was also included. All the containers were placed in 1.8L Agee jars containing vials with 10ml 1M NaOH to absorb CO₂ and 10 ml CO₂ free water to avoid any loss of moisture during incubation. The jars were placed in a room where temperature ranged between 18.5 and 24.3° C during the measurement period (see Figures 5.1 & 5.2). The incubations were carried out for 129 days during which vials of 1M NaOH were removed at 2-4 day intervals for measuring trapped CO₂, the jars were flushed with ambient air, a fresh vial containing 10 ml of 1M NaOH was placed back inside the jar for CO₂ absorption, and the jars were resealed. The weight of the container + soil + residues was recorded and adjusted for moisture in order to maintain 80% field capacity. Moisture was adjusted weekly to correct the moisture loss from soils during venting of jars.

The percentage of C decomposition of the residue was calculated as follows:

net CO_2 - $C_{emitted} = CO_2$ - $C_{amended}$ - CO_2 - $C_{control}$(Eq. 5.1)

Net CO_2 - $C_{emitted}$ represents the amount of C emitted from ryegrass residue amended soil (CO_2 - $C_{amended}$) minus C emitted from unamended soil (CO_2 - $C_{control}$), which represents background soil organic matter decomposition.

% residue C _{decomposition} = [net CO₂-C_{emitted}/C _{added}] X 100..... (Eq. 5.2) C _{added} is the amount of C added as residues to soil.

It is acknowledged that this calculation does not consider any priming effect caused by addition of the residue. The estimates of residue CO₂-C emissions could be overestimated if priming occurs.

5.2.4 Soil fractionation

At the end of the decomposition study the control and residue amended soils were then subjected to physical fractionation to see if there were any changes in the fractions due to added residues.

5.2.4.1 Light (LF) and heavy fraction (HF) density separation

Two replicates of each treatment (12, 16 and 20 Mg dry matter residue ha⁻¹ and unamended control soil) were separated using density fractionation. Density fractionation was conducted using air-dried samples. Soil subsamples (equivalent to 5 g on an oven dry basis) were weighed into 40 ml centrifuge tubes and 30 ml of NaI (density= 1.8 g cm^{-3}) was added. The tubes were shaken end over end for 5 minutes and were left to stand for 2 hours. After standing, the top half of the suspended soil and floating roots were sucked out of the tube with a rubber hose attached to a side arm flask (under vacuum). A glass tube with 54 µm nylon mesh fitted at the top was placed inside the flask for collection of the light fraction. The light fraction material collected on the mesh was washed thoroughly to remove any soil particles and NaI solution. The light and heavy fractions were washed once with 0.01M CaCl₂ and about 10 times with deionised water as stated by Zhang *et al.* (2007) to ensure complete removal of NaI. Both light and heavy fractions were dried at 70°C and analysed for total-C by LECO.

5.2.5 Labile carbon fractions

5.2.5.1 Hot water extractable-C

Hot water extractable-C (HWC) was determined on field moist soil samples maintained at 80% of field capacity according to the method of Ghani *et al.* (2003). Soil samples (equivalent 3 g on oven dry basis) were weighed into 50 ml polypropylene centrifuge tubes, and 30 ml of distilled deionised water was added into the centrifuge tubes. These tubes were shaken on a vortex shaker for 10 seconds to suspend the soil in water. The tubes were capped and left for 16 hours in a hot-water bath at 80°C. At the end of the 16 hour period, each tube was shaken for 10 seconds on a vortex shaker to ensure that the HWC released from the soil organic matter was fully suspended in the extraction medium. These tubes were then centrifuged for 20 minutes at 3500rpm (2200g). Supernatants were filtered

through cellulose nitrate membrane filters and were analysed for C by acid dichromate digestion.

5.2.5.2 Microbial biomass-C

Field moist soil samples maintained at 80% of field capacity were analysed for microbial biomass-C by the fumigation extraction method (Vance *et al.* 1987). Non-fumigated soil samples and samples that had been fumigated with ethanol free CHCl₃ were extracted with 0.5M K₂SO₄ for 30 min (1:5 soil: extractant ratio), filtered and an aliquot was analysed for C by acid dichromate digestion. The C from the fumigated samples minus non-fumigated samples was taken to represent the microbial-C flush and converted to microbial biomass C using the relationship: microbial $C = \frac{1}{0.41}$ C flush (Saggar *et al.* 2001).

5.2.5.3 Hydrogen peroxide oxidisable-C

Five grams of ring-ground, air-dried soil was weighed into a 110 ml plastic container, a magnetic bar placed in it, and the plastic container placed in a 1L Agee jar, set on a magnetic stirrer. An ambient air sample (t0) was collected immediately after sealing the jars with the lid fitted with rubber septum. Thereafter, 25 ml of 2% H_2O_2 was added into the plastic container with a 50 ml polypropylene syringe, and the magnetic stirrer was started to stir the soil-peroxide mixture. Gas samples were collected over a period of four hours (at 0, 1, 2, 3 and 4 hours) using a 25 ml polypropylene syringe fitted with a 3-way stopcock, and transferred into 12 ml evacuated vials. These over-pressured gas samples were analysed using a Shimadzu GC- 17A gas chromatograph (GC). Prior to the calculations it was confirmed that there was a linear production of CO_2 during the 4 hour period.

The amount of C oxidised from 5 g soil by 2.0% hydrogen peroxide was calculated by subtracting the CO_2 concentration at t (0) from t (4hour). The resultant CO_2 concentration was used for further calculations:

Concentration of CO_2 obtained from $GC = \mu L CO_2 L^{-1}$

Density of CO₂= 1.8 μ g μ L⁻¹ at 25°C temperature and 1 atmosphere pressure

Volume of Jar= 1L

Weight of soil taken= 5 g

0.2727 is the factor to convert CO_2 to C

The amount of C oxidised after 4 hours was correlated with the amount of CO_2 -C respired during the 129 day incubation period.

5.2.5.4 Potassium dichromate oxidisable-C

This fraction was determined through a method modified from Walkley and Black (1934) (Chan *et al.* 2001). The method involves oxidation of total organic carbon by 0.4N $K_2Cr_2O_7$ using 18 N H₂SO₄. In the standard Walkley and Black method 36N H₂SO₄ and 1N $K_2Cr_2O_7$ are used. The organic carbon oxidisable by 0.4N $K_2Cr_2O_7$ using 18 N H₂SO₄ was considered to be a very labile C fraction.

5.2.5.5 Potassium sulphate extractable-C (Soluble-C)

Field moist soil samples maintained at 80% of field capacity were extracted with 0.5M K₂SO₄ for 30 min (1:5 soil:extractant ratio), filtered, and an aliquot was analysed for C by acid dichromate digestion.

5.2.6 Recovery percentage of the added residue-C

The recovery percentage of the added residue-C was calculated as follows:

% recovery = $\frac{(LF-C+HF-C) \text{ after incubation} + CO_2-C \text{ emitted}}{(LF-C+HF-C) \text{ before incubation} + \text{ added residue-C}} x100.....(Eq. 5.4)$

(LF-C+HF-C) after incubation is the sum of total-C determined on LF and HF of control and residue applied treatments of the four test soils after incubation study i.e. on day 129.

CO₂-C emitted is the amount of residue-C emitted as CO₂ during the incubation period.

(LF-C+HF-C) before incubation is the sum of total-C determined on LF and HF of control treatment of the four test soils before starting the incubation study i.e. on day zero.

Added residue-C is the rate of residue-C addition to the soil i.e. $5.11-8.51 \text{ mg C g}^{-1}$ soil.

5.2.7 Statistical analysis

An analysis of variance using SAS software (9.1) was performed on the results of cumulative CO_2 -C emissions, light and heavy fraction C recovered at end of incubation, and percentage of added residues decomposed using a General Linear Model (GLM) procedure. Percentage data was log transformed and mean comparisons were made on transformed data. Mean comparisons were done using Fisher's least significant difference (LSD) at 5% level of significance. The regression coefficients were determined using SAS software (9.1) to determine relationships between total amounts of CO_2 respired during the incubation study and different labile-C fractions extracted. All soil replicates were used in the regression analysis. An analysis of covariance (ANCOVA) was performed using Minitab-16 with soil

type as covariate, in order to answer the question of whether the extracted labile-C fractions used to predict the CO_2 emissions are affected by soil type. Similar to the linear regressions, the ANCOVA used the response variable on the y-axis and the covariate as the independent variable on the x-axis. Full models that included an interaction of covariate with labile-C fraction were performed. The interaction term allows the regression lines to have different slopes. All soil replicates and residue treatments were used in the analysis. An analysis of variance (ANOVA) was performed using Minitab-16 to analyse the effect of residue rates and soil types on labile-C fractions and CO_2 respired.

5.3 Results

5.3.1 Soils

Soils used in this study significantly varied in total-C and N contents but there were minor differences in texture and pH.

5.3.2 Decomposition of added ryegrass pasture residues

The CO_2 -C emission rates from all four test soils responded rapidly to the residue application. The first measurements (on day-2) showed that the CO_2 -C emission rates from residue incorporated soils were four to five times than that observed from control soils. In all the four test soils, residue treatments showed large pulses of CO_2 in the initial weeks and then decreased gradually over time (Figures.5.1 and 5.2). It is noted that the correction of moisture loss on day-57 resulted in an increase in CO_2 emissions for the next measurement.

The amounts of CO₂-C emitted from Carnarvon SL, Ohakea stony SiL, Ohakea SiL, and Horotiu SiL were 2.57, 5.02, 6.19 & 7.32 mg C g⁻¹ soil; 2.76, 5.40, 5.72 & 7.10 mg C g⁻¹ soil; 3.59, 6.66, 7.60 & 8.31mg C g⁻¹ soil and 3.32, 5.73, 6.62 & 7.39 mg C g⁻¹ soil at 0, 5.11, 6.81 & 8.51 mg C g⁻¹ soil of residue application rates (Figure 5.3) during the 129 days incubation period. Irrespective of the soil type, CO₂-C emissions from the residue treated soils were significantly higher than control soils. Despite the different rates of residue addition, the surface soil conditions were similar in all jars and there was no evidence of excessive fungal growth caused by soil type or residue addition.

The proportion of applied residue-C emitted as CO_2 during 129 day incubation period varied between 43.3 and 60.0%. No significant differences were observed between the amount/proportion of residue-C decomposed as CO_2 at each level of residue applied in all the four test soils (Table 5.3). Irrespective of the residue input rates about 25.0-37.7% of the residue-C decomposed during the first 30 days in all the four test soils, followed by slow

decomposition (11.1-14.2%) in the next 30 days (31-60 days). Subsequent residue decomposition was very slow with only 6-11% residue-C decomposed in the following 69 days (Table 5.2).

There were no clear effects of soil type or texture on the residue-C decomposition during the incubation period (Table 5.3). There were no clear effects of the total-C contents on the amounts of CO_2 -C emitted from the control treatment of the four test soils (Figure 5.3).



Figure.5.1: The CO₂-C emission rates from (a) Carnarvon SL and (b) Ohakea stony SiL as influenced by residue application during 129 days of laboratory incubation.



Figure.5.2: The CO₂-C emission rates from (a) Ohakea SiL and (b) Horotiu SiL as influenced by residue application during 129 days of laboratory incubation.



Figure.5.3: Cumulative CO₂-C emissions from Carnarvon SL, Ohakea stony SiL, Ohakea SiL and Horotiu SiL. Each bar represents a mean of four replicates with standard error. Letters indicate statistically significant differences (p < 0.05).

	Ra	Rates of residue addition			
SOILS	5.11	6.81	8.51		
SOILS	mg C g ⁻¹ soil	mg C g ⁻¹ soil	mg C g ⁻¹ soil		
	0-30 days				
Carnarvon SL	28.0	33.6	32.1		
Ohakea stony SiL	32.5	25.0	30.7		
Ohakea SiL	37.7	34.4	33.9		
Horotiu SiL	27.8	27.7	26.2		
31-60 days					
Carnarvon SL	11.1	12.6	12.5		
Ohakea stony SiL	12.6	11.9	13.3		
Ohakea SiL	14.2	13.3	13.4		
Horotiu SiL	12.1	12.3	13.3		
	61-12	29 days			
Carnarvon SL	8.7	6.9	11.0		
Ohakea stony SiL	6.4	6.4	6.9		
Ohakea SiL	8.0	11.0	8.0		
Horotiu SiL	7.2	8.2	8.2		

Table 5.2: Percentage of added residue-C respired at different times of the incubation in the four different soils amended with different rates of ryegrass pasture residues.

Each value represents a mean of four replicates.

Table 5.3: Percentage of added residue-C decomposed after	129	days i	in the	four	different
soils amended with different rates of ryegrass pasture residue.					

	Rates of residue addition				
SOILS	5.11	6.81	8.51		
	mg C g ⁻¹ soil	mg C g ⁻¹ soil	mg C g ⁻¹ soil		
Carnarvon SL	B47.9a	A53.2a	A55.8a		
Ohakea stony SiL	BA51.6a	B43.3a	B50.9a		
Ohakea SiL	A60.0a	A58.8a	A55.4a		
Horotiu SiL	B47.2a	BA48.4a	B47.8a		

Each value represents a mean of four replicates. Capital letters (A, B, C, D) stands for differences between soils and small letters (a, b, c, d) stand for differences between residue rates.

5.3.3 Residue-C recovery

Total-C determined on the light (LF) and heavy fractions (HF), plus the CO₂-C emitted during the incubation period (Table 5.4), accounted for 97 to 110% of the total C in residue treatments (at 5.11, 6.81 and 8.51 mg C g⁻¹ soil) and 99 to 105% of C in control treatments. Addition of ryegrass pasture residues had no significant effect on the amounts of C in the heavy fraction in all the test soils during the incubation period, except Carnarvon SL in which the amount of C in the heavy fraction at the 5.11 and 6.81 mg C g⁻¹ soil residue addition significantly decreased in comparison to the control. However, no differences were observed between the control and the 8.51 mg C g⁻¹ soil residue level. In contrast, LF-C showed significant differences between the treatments in all the four test soils (Table 5.4).

The total-C determined on the light (LF) and heavy fractions (HF) on the control treatment of the four test soils at the beginning (on day-0) and after 129 days incubation study are given in Table 5.5. No significant differences were observed for LF and HF-C determined at the beginning and after the incubation for all the soils tested. However, over the duration of the incubation study, LF-C of the four test soils declined.

Carnarvon SL						
Treatment	LF-C (mg C g ⁻¹ soil)	HF-C (mg C g ⁻¹ soil)	CO ₂ -C respired (mg C g ⁻¹ soil)	% recovery		
Control	3.68 ± 0.29	38.53 ± 0.10	2.57 ± 0.09	102.3		
5.11 mg C g ⁻¹ soil	7.44 ± 0.15	35.31 ± 0.42	5.02 ± 0.10	97.7		
6.81 mg C g ⁻¹ soil	9.21 ± 0.70	35.53 ± 0.54	6.19 ± 0.11	100.7		
8.51 mg C g ⁻¹ soil	10.55 ± 2.16	39.88 ± 1.31	7.32 ± 0.10	110.4		
LSD (p< 0.05)	4.51	2.91	0.30			
Ohakea stony SiL						
Treatment	LF-C (mg C g ⁻¹ soil)	HF-C (mg C g ⁻¹ soil)	CO ₂ -C respired (mg C g ⁻¹ soil)	% recovery		
Control	0.84 ± 0.05	25.14 ± 0.30	2.76 ± 0.08	99.9		
5.11 mg C g ⁻¹ soil	3.69 ± 0.50	26.27 ± 0.71	5.40 ± 0.08	104.3		
6.81 mg C g ⁻¹ soil	2.63 ± 0.82	26.59 ± 0.77	5.72 ± 0.24	98.2		
8.51 mg C g ⁻¹ soil	4.27 ± 1.04	26.10 ± 0.79	7.10 ± 0.03	100.5		
LSD (p< 0.05)	2.78	2.64	0.41			
Ohakea SiL						
Treatment	LF-C (mg C g ⁻¹ soil)	HF-C (mg C g ⁻¹ soil)	CO ₂ -C respired (mg C g ⁻¹ soil)	% recovery		
Control	2.62 ± 0.48	33.23 ± 0.80	3.59 ± 0.14	102.1		
5.11 mg C g ⁻¹ soil	5.57 ± 0.55	34.02 ± 0.64	6.66 ± 0.14	105.8		
6.81 mg C g ⁻¹ soil	6.39 ± 1.20	33.04 ± 1.29	7.60 ± 0.03	103.5		
8.51 mg C g ⁻¹ soil	6.84 ± 0.15	32.92 ± 1.08	8.31 ± 0.08	102.0		
LSD (p< 0.05)	2.76	3.86	0.34			
		Horotiu SiL				
Treatment	LF-C (mg C g ⁻¹ soil)	HF-C (mg C g ⁻¹ soil)	CO ₂ -C respired (mg C g ⁻¹ soil)	% recovery		
Control	1.94 ± 0.42	47.81 ± 0.97	3.32 ± 0.06	105.5		
5.11 mg C g^{-1} soil	5.95 ± 0.60	46.22 ± 0.92	5.73 ± 0.11	104.5		
6.81 mg C g ⁻¹ soil	6.95 ± 0.78	46.86 ± 1.63	6.62 ± 0.08	105.8		
8.51 mg C g^{-1} soil	7.64 ± 0.11	45.86 ± 1.80	7.39 ± 0.04	103.5		
LSD (p< 0.05)	2.12	5.45	0.23			

Table 5.4: Light and heavy fraction-C as influenced by ryegrass pasture residue addition and recovery (%age) of added C after 129 days of the incubation study.

Each value of LF-C and HF-C represents a mean of two replicates \pm SE. Each value of CO₂-C represents a mean of four replicates \pm SE. Table 5.5: Light and heavy fraction-C at the beginning (day-0) and after 129 days incubation study in control treatments of the four soils.

:

Carnarvon SI	von SI	.]	Ohakea s	tony SiL	Ohake	a SiL	Horoti	iu SiL
$ \begin{array}{c c} LF-C \\ (mg \ C \ g^{-1} \ soil) \end{array} \begin{array}{c c} HF-C \\ (mg \ C \ g^{-1} \ soil) \end{array} \begin{array}{c c} LF-C \\ (mg \ C \ g^{-1} \ soil) \end{array} \end{array} $	$\begin{array}{c c} HF-C \\ (mg \ C \ g^{-1} \ soil) \end{array} (mg \ C \ g^{-1} \ soil) \end{array}$	LF-C (mg C g ⁻¹ soil)		HF-C (mg C g ⁻¹ soil)	LF-C (mg C g ⁻¹ soil)	HF-C (mg C g ⁻¹ soil)	LF-C (mg C g ⁻¹ soil)	HF-C (mg C g ^{-l} soil)
5.18±0.36 38.60±1.72 1.90±0.80	38.60±1.72 1.90±0.80	1.90 ± 0.80		26.88±1.02	4.10 ± 1.06	34.52±1.64	3.23±0.50	47.09±3.52
3.68±0.29 38.53±0.10 0.84±0.05	38.53±0.10 0.84±0.05	0.84 ± 0.05		25.14 ± 0.30	2.62 ± 0.48	33.23±0.80	1.94 ± 0.42	47.81±0.97
0.06 0.97 0.28	0.97 0.28	0.28		0.19	0.27	0.52	0.09	0.86

Each value represents a mean of four replicates \pm SE for Day-0 Each value represents a mean of two replicates \pm SE for Day-129

5.3.4 Labile carbon fractions

The purpose of the decomposition study was to assess background decomposition and that of the added pasture residues through emissions of CO_2 ; and relate these emissions to measured labile C fractions. The five labile C fractions determined on the soils used in the study were hot water extractable-C (HWC), microbial biomass-C (MBC), 2% hydrogen peroxide oxidisable-C (H₂O₂-C), 0.4N potassium dichromate oxidisable-C (K₂Cr₂O₇-C) and 0.5M K₂SO₄ extractable-C (K₂SO₄-C). The labile C fractions determined on the control treatments of the four soils used in the study on day-0 i.e. before starting the incubation study are given in Table 5.6. The MBC ranged between 0.35 and 0.93 mg CO₂-C g⁻¹soil, HWC between 0.93 and 1.36 mg CO₂-C g⁻¹soil, H₂O₂-C between 0.12 and 0.82 mg CO₂-C g⁻¹soil, K₂Cr₂O₇-C between 2.62 and 4.21 mg CO₂-C g⁻¹soil and K₂SO₄-C between 0.10 and 0.21 mg CO₂-C g⁻¹soil for the four soils used in the incubation study.

Coefficients of determination comparing the CO₂-C respired during the 129 days incubation period with the labile C fractions (MBC, HWC, H₂O₂-C, K₂Cr₂O₇-C and K₂SO₄-C are given in Table 5.7 A. None of the labile C fractions, except MBC (R^2 = 0.70**) showed any significant relationship with the CO₂-C respired during the incubation period. This suggested an influence of soil type on the extracted labile C fractions used for predicting CO₂-C emissions from a range of soils.

Listed in Table 5.7 B are the p-values for the slope comparisons of different labile C fractions extracted from the four soil types. Statistically significant differences between the slopes of the four soil types suggested the effect of soil type on the extracted labile C fractions used to predict CO_2 -C emissions.

Labila C fractions	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL		
Lablie C fractions	$(mg C g^{-1} soil)$					
MBC	0.35±0.02	0.48 ± 0.01	0.93±0.03	0.58 ± 0.01		
HWC	1.31±0.01	0.93±0.01	1.36±0.02	1.05 ± 0.02		
H ₂ O ₂ -C	0.82±0.045	0.20±0.003	0.61±0.024	0.12±0.003		
K ₂ Cr ₂ O ₇ -C	3.97±0.03	2.62±0.05	3.52±0.03	4.21±0.03		
K ₂ SO ₄ -C	0.16±0.01	0.11±0.00	0.10±0.01	0.21±0.00		

Table 5.6: Labile C fractions (mg C g^{-1} soil) determined on the control treatments of the four soils at the beginning (day-0) of the incubation study.

Each value represents a mean of four replicates± SE.

Labile C fractions vs. CO ₂ -C respired				
MBC	$R^2 = 0.70 * *$	p<0.001		
HWC	$R^2 = 0.06$	p=0.34		
H ₂ O ₂ -C	$R^2 = 0.04$	p=0.44		
K ₂ Cr ₂ O ₇ -C	$R^2 = 0.05$	p=0.42		
K ₂ SO ₄ -C	$R^2 = 0.01$	p=0.75		

Table 5.7 A: Linear regression coefficients of determination comparing the CO_2 -C respired during 129 days incubation period with the labile C fractions determined on the control treatments of the four soils at the beginning (day-0) of the incubation study.

Table 5.7 B: P-values comparing the slopes for different labile-C fractions of Carnarvon SL, Ohakea stony SiL, Ohakea SiL with the slope of Horotiu SiL.

		Horotiu	SiL (p-value f	for slope)		
Soil type	Labile-C fractions					
	MBC	HWC	H ₂ O ₂ -C	K ₂ Cr ₂ O ₇ -C	K ₂ SO ₄ -C	
Carnarvon SL	0.023	0.242	0.041	0.044	0.012	
Ohakea stony SiL	0.315	0.018	0.995	0.720	0.818	
Ohakea SiL	1.000	0.049	0.029	0.032	0.059	

MBC= Microbial biomass-C

HWC= Hot water extractable-C

 H_2O_2 -C= Carbon oxidised by 2% hydrogen peroxide during 4 hour duration

K₂Cr₂O₇-C= Carbon oxidised by 0.4N potassium dichromate

 K_2SO_4 -C= 0.5M K_2SO_4 extractable-C

Of all the labile C fractions determined on the control soils, only MBC and HWC were determined on residue treated soils (residues applied at 0, 5.11, 6.81 and 8.51 mg C g⁻¹ soil) before and after the incubation study to find a relationship between CO₂-C evolved during the incubation and MBC & HWC. The MBC and HWC were determined immediately after the addition of residues to the soils. The HWC in the four soils at different residue input rates before and after the incubation study are given in Tables 5.8 and 5.9. Addition of residues significantly increased the HWC content at every level in all the soils before incubation, suggesting that addition of residues also contribute to HWC fraction. There was a decrease in HWC content after 129 days of incubation at every level of residue addition in comparison to the content present in the beginning. Similar results were observed for MBC in all the four soils at different residue input rates before incubation, MBC significantly increased in comparison to the control treatment with addition of residues in all the soils. After 129 days of incubation no significant difference was observed between any rates of added residues.

HWC (mg C g ⁻¹ soil)							
Treatment	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL			
Control	1.31 ± 0.01	0.93 ± 0.01	1.36 ± 0.02	1.05 ± 0.02			
$5.11 \text{ mg C g}^{-1} \text{ soil}$	1.58 ± 0.01	1.17 ± 0.01	1.48 ± 0.02	1.17 ± 0.02			
6.81 mg C g ⁻¹ soil	1.66 ± 0.02	1.24 ± 0.02	1.53 ± 0.01	1.24 ± 0.02			
8.51 mg C g ⁻¹ soil	1.70 ± 0.01	1.29 ± 0.01	1.59 ± 0.01	1.26 ± 0.02			
LSD (p< 0.05)	0.04	0.04	0.05	0.06			

Table 5.8: Hot water extractable-C at the beginning (day-0) of the incubation study in the four soils as influenced by pasture residue additions.

Each value represents a mean of four replicates± SE.

Table 5.9: Hot water extractable-C after 129 days of the incubation study in the four soils as influenced by pasture residue additions.

HWC (mg C g^{-1} soil)							
Treatment	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL			
Control	1.09 ± 0.02	0.78 ± 0.01	1.09 ± 0.01	0.80 ± 0.02			
5.11 mg C g ⁻¹ soil	1.17 ± 0.01	0.84 ± 0.01	1.10 ± 0.01	0.82 ± 0.02			
6.81 mg C g ⁻¹ soil	1.19 ± 0.02	0.82 ± 0.01	1.12 ± 0.01	0.83 ± 0.02			
8.51 mg C g ⁻¹ soil	1.22 ± 0.04	0.88 ± 0.02	1.14 ± 0.03	0.83 ± 0.03			
LSD (p< 0.05)	0.09	0.06	0.07	0.09			

Each value represents a mean of two replicates ± SE.

MBC (mg C g ⁻¹ soil)							
Treatment	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL			
Control	0.35±0.02	0.48±0.01	0.93±0.03	0.58 ± 0.01			
$5.10 \text{ mg C g}^{-1} \text{ soil}$	0.50 ± 0.04	0.65±0.01	$1.04{\pm}0.00$	0.71±0.02			
6.81 mg C g ⁻¹ soil	0.53±0.03	0.73±0.02	1.11±0.02	$0.74{\pm}0.02$			
8.51 mg C g ⁻¹ soil	0.59±0.04	0.75±0.04	1.11±0.02	0.75±0.02			
LSD (p< 0.05)	0.11	0.07	0.06	0.05			

Table 5.10: Microbial biomass-C at the beginning (day-0) of the incubation study in the four soils as influenced by pasture residue additions.

Each value represents a mean of four replicates± SE.

Table 5.11: Microbial biomass-C after 129 days of the incubation study in the four soils as influenced by pasture residue additions.

MBC (mg C g ⁻¹ soil)						
Treatment	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL		
Control	0.23±0.01	0.37±0.01	0.38±0.00	0.33±0.03		
5.10 mg C g^{-1} soil	0.24±0.03	0.38±0.00	0.37±0.02	0.35±0.06		
6.81 mg C g ⁻¹ soil	0.27±0.01	$0.40{\pm}0.00$	0.40 ± 0.00	0.35±0.03		
8.51 mg C g^{-1} soil	0.27±0.03	$0.47{\pm}0.05$	$0.40{\pm}0.01$	0.39±0.00		
LSD (p< 0.05)	0.09	0.11	0.04	0.16		

Each value represents a mean of two replicates ± SE.

The plots of the relationship between cumulative amount of CO_2 -C emitted and MBC and HWC measured at the beginning (day-0) of the incubation study using ANCOVA are given in Figures 5.4 (a, b) and p-values comparing the slopes for four soil types in Table 5.12.

Analysis of covariance takes different soil types into account and explains more variation in the data i.e. MBC ($R^2= 0.75$) and HWC ($R^2= 0.89$) and showed a reasonable relationship with cumulative amount of CO₂-C evolved during the incubation study. However, the slopes of the four soil types (Table 5.12) differ significantly, which suggested the effect of soil type on the extracted labile C fractions.



Figure.5.4: Relationships between cumulative CO_2 -C respired during 129 days incubation from the four soil types at different residue rates with (a) microbial biomass-C and (b) hot water extractable-C measured at the beginning (day-0) of the incubation study using ANCOVA model.

Table 5.12: P-values comparing the slopes of the Carnarvon SL, Ohakea stony SiL, Ohakea SiL with the slope of Horotiu SiL for microbial biomass-C (MBC) and hot water extractable-C (HWC) determined at different residue rates.

Soil trma	Horotiu SiL (p-value for slope)		
Son type	MBC	HWC	
Carnarvon SL	0.164	0.000	
Ohakea stony SiL	0.042	0.001	
Ohakea SiL	0.074	0.000	

Table 5.13: Summary of the analysis of variance of the amounts of microbial biomass-C (MBC) and hot water extractable-C (HWC) determined on day-0 when four soils were incubated with different ryegrass residue rates and the total CO_2 -C evolved during the incubation.

Source	DF	Sum of squares		
		MBC	HWC	Total CO ₂ -C evolved
Soil type	3	2.6408	2.1042	17.534
Residue rate	3	0.4542	0.8285	176.054
Soil type X Residue rate	9	0.0223	0.0671	2.193
Error	48	0.1162	0.0439	2.196
Total	63	3.2337	3.0438	197.978

In order to check the capability of H_2O_2 to oxidise the labile-C fraction from the soils used in this study, all the four test soils (5 g ring ground, air dried soil) were mixed with glucose (a highly labile C source) equivalent to 44.12 mg C (the amount was calculated on the basis of the amount completely oxidised by 25 ml of 1% H_2O_2), and ryegrass pasture residues equivalent to 35.31 mg C (amount equivalent to the 12 Mg residues per hectare treatment used in the laboratory decomposition). Glucose and ryegrass pasture residue mixed soil samples (in triplicate) and were then oxidised with 2% H_2O_2 at 35°C for 2 hours (Table 5.14). The amounts of C oxidised from soils with addition of glucose and residues did not show any significant difference in comparison to the amounts oxidised from the control soils. The amount of carbon oxidised by 2% H_2O_2 showed a similar trend as observed when soils alone were oxidised, suggesting that the oxidation behaviour of H_2O_2 is affected by the soil type and might not be able to differentiate between residue inputs.

Table 5.14: Amount of carbon oxidised by 2% H₂O₂ during a 2 hour oxidation period at 35° C from the four soils.

Treatment	Carnarvon SL	Ohakea stony SiL	Ohakea SiL	Horotiu SiL
	(mg C g ⁻¹ soil)			
Soil alone	0.66 ± 0.06	0.27±0.04	$0.60{\pm}0.05$	0.22±0.07
Soil+glucose	0.69±0.11	0.29 ± 0.07	0.59±0.12	0.18±0.06
Soil+roots	0.67 ± 0.07	0.32±0.04	0.64 ± 0.09	0.29±0.07
LSD (p< 0.05)	0.30	0.19	0.33	0.24

Each value represents a mean of three replicates \pm SE.

5.4 Discussion

5.4.1 Decomposition of added ryegrass pasture residues

Carbon dioxide evolution from soils responded rapidly to residue incorporation. The pattern of decomposition suggested an initial fast phase due to mineralization of easily degradable organic C in residues, followed by a slower phase in which more complex and recalcitrant forms or transformed metabolites are mineralized (Table 5.2). This pattern is supported by many previous studies (Saggar et al. 1996; Curtin et al. 1998; Lu et al. 2003; Nourbakhsh 2006; Verma et al. 2010). During the 129 days of incubation, the proportion of residues decomposed did not differ between differences in soil texture/type as no consistent trend was observed (Table 5.3). Numerous studies conducted in the past to determine the influence of soil texture (clay content) on organic residue decomposition (Sorensen 1983; Ladd et al. 1981, 1985, 1992; Sharkov and Bukreeva 2004) have showed that in comparison to sandy soils, fine textured soils (with higher clay content) have lower rates of residue decomposition and retain higher proportions of residues by complexing with decomposition products, thereby reducing the losses of residue C from soil. The results of the present study are not in accordance with the studies reported in the literature where greater differences in soil textures (sand/clay soils) have been compared, contrasting with the narrow range of soil textures (sandy loam/silt loam) and the small number of soils used in this study. Results of this study showed that the proportion of added residue C decomposed during the entire incubation was independent of the rates of residue addition. These results are in accordance with the results obtained by Jenkinson (1977), who suggested that 'the proportion of the C evolved from residue amended soil above from the control soil has always been independent of the quantity added if the C addition does not exceed 1.5% of the dry weight of soil and if decomposition was allowed to continue for at least 3 to 6 months'. These conditions are applicable to our study as the experiment was conducted over 4 months and residue C additions did not exceed 1.5% of the dry weight of soil.

5.4.2 Residue-C recovery

Christensen (1992) reported that the LF is composed of mineral free organic residues at various degrees of decomposition with high C concentration, whereas the heavy fraction (HF) comprises high density organo-mineral complexes with low C concentrations that are more stable than the light fraction. Separation of the whole soil into a light (< 1.8 g cm⁻³) and heavy fraction (> 1.8 g cm⁻³) showed that the HF-C was completely unaffected by the application of residue (Table 5.4). This suggested that the HF consisted of stable/native C

with a very low decomposition rate. The significant decrease of the HF-C at 5.11 and 6.81 mg C g^{-1} soil residue rates in Carnarvon SL in comparison to the control could be due to experimental variability.

However, the LF-C showed clear differences between residue application rates, and all the applied residue rates showed a typical decomposition pattern - initially rapid followed by slower decay rates. The results of the present study are similar to that reported by Magid *et al.* (1997). They separated the residues remaining in the soils into light and heavy fractions after a 20 month incubation period and observed that application of residues made no difference to HF-C, whereas LF-C increased with application of residues.

The amounts of LF-C in the control treatment of the four soils at the beginning and after 129 days incubation did not vary significantly (Table 5.5). However, over the duration of the incubation study, LF-C of the four test soils decreased. The large replicate variability due to presence of the background root residues in the 2 mm sieved soils used in the study could explain why significant differences were not observed.

5.4.3 Labile carbon fractions

Many chemical and biological techniques have been proposed to measure the labile fraction of soil organic matter in a hope to provide an index of loss of organic carbon due to land use changes. Among the various labile C fractions, hot water extractable-C (HWC), microbial biomass-C (MBC), 2% hydrogen peroxide oxidisable-C (H_2O_2 -C), 0.4N potassium dichromate oxidisable-C ($K_2Cr_2O_7$ -C) and 0.5M K_2SO_4 extractable-C (K_2SO_4 -C) were used to predict the CO₂-C evolved during the laboratory incubation. Except for MBC, all other labile C fractions (i.e. HWC, H_2O_2 -C, $K_2Cr_2O_7$ -C and K_2SO_4 -C) had poor and insignificant relationships with the amount of CO₂-C respired during the incubation study from the control soils (Table 5.7A). The significant differences between the slopes suggest that the labile C fractions were strongly influenced by soil type (Table 5.7B).

Soil treatment with hydrogen peroxide was introduced by Robinson (1922) to remove organic matter prior to particle size analysis and before soil mineralogy analysis (Feller *et al.* 1992). The SOC fraction resistant to H_2O_2 treatment is considered to be a stable C fraction (Leifeld and Kogel-Knabner 2001). Many past studies (von Lutzow *et al.* 2007; Helfrich *et al.* 2007; Favilli *et al.* 2008) using concentrated H_2O_2 (10-30% (wt/wt)) suggested wet oxidation with H_2O_2 selectively removes the active soil C pool, leaving behind the stable C pool. In the present study a very mild concentration of H_2O_2 (2% (wt/wt)) was used to oxidise the labile-C fraction. Among the four soils, the amount of carbon oxidised by H_2O_2 was highest in Carnarvon SL followed by Ohakea SiL, Ohakea stony SiL, and lowest in Horotiu SiL (Table 5.6), which could be due the influence of clay content on the oxidation of soil C by H₂O₂. Hosking (1932) observed that soils having a higher clay content showed resistance to oxidation by H₂O₂, signifying the protection of organic matter by interaction with clay minerals. During H₂O₂ oxidation, Fe and Al associated with organic matter are released and precipitate as hydroxides on organic and inorganic components. These precipitates alter the specific surface area and charge of the mineral phase and can protect organic matter from further degradation (Sequi and Aringhieri 1977). This could explain why the lowest amount of C oxidised was observed in the allophanic Horotiu SiL soil (rich in clay minerals of short-range order namely allophane, ferrihydrite and imogolite). Eusterhues et al. (2005) observed a positive correlation between the peroxide resistant organic C with clay content and iron oxides in two forest soils. Petigara et al. (2002) observed that in surface soils with high organic matter and manganese content, H_2O_2 decomposes to water, di-oxygen and hydroxyl ion instantly, with water and di-oxygen dominating the decomposition, and the formation of free hydroxyl ion represents <10% of the total H_2O_2 decomposed. The consumption of H_2O_2 in metal oxidation reduces organic matter oxidation.

Hot water extractable-C consists of a labile pool of SOM which includes microbial biomass C, carbohydrates, and amines (Sparling *et al.* 1998). Previous studies (Ghani *et al.* 2003; Schulz 2004) suggested hot water extractable-C as one of the sensitive indicators to reflect changes in soil organic matter caused by different management practices. Ghani *et al.* (1999) observed that 45-60% of C extractable with hot water was carbohydrates. Shepherd *et al.* (2001) and Haynes *et al.* (1991) showed a decrease in hot-water extractable carbohydrate in pasture soils on conversion to continuous cropping using conventional cultivation. Significant increases in the HWC fraction, with addition of pasture residues in comparison to the control treatment in all the four test soils before the incubation, suggest the contribution of residues to the HWC fraction.

Soil microbial biomass C is an active C fraction used as an early predictor of change in soil organic matter due to land use practices (Powlson *et al.* 1987; Yeates and Saggar 1998; Saggar *et al.* 2001; Melero *et al.* 2006; Brookes *et al.* 2008; Nyamadzawo *et al.* 2009). Addition of pasture residues significantly increased the MBC in comparison to the control treatment in all the test soils before the incubation study. These results accord with the previous studies which suggest that soil MBC is affected by the input of organic residues, with high amounts of organic inputs resulting in higher microbial biomass carbon (Peacock *et al.* 2001; de Araujo and de Melo 2010). In the present study, MBC measurements were made immediately after the addition of residues. However, previous studies (Ocio and Brookes 1990; Ocio *et al.* 1991a, b) suggest that, to have a valid estimate of the biomass C in residue-amended soils, measurements should be made 5-20 days after the addition of residues. Due to its rapid turnover rate, microbial biomass responds quickly to land use changes in comparison to total SOM, but some researchers regard it as a poor predictor of SOM change in the longer term because the amount of MBC in soil is affected by the amount of crop residues, soil moisture, and temperature (Wardle and Parkinson 1990; Sarathchandra *et al.* 1989; He *et al.* 1997; Murphy *et al.* 2007).

Soil C oxidised by 0.4N $K_2Cr_2O_7$ was assumed to be the labile C fraction. The amount of C oxidised by 0.4N $K_2Cr_2O_7$ ranged between 8.3-9.2% of the total-C content present in the four test soils used in the study. This suggested that 0.4N $K_2Cr_2O_7$ oxidises a constant proportion of total-C depending upon the soil type, therefore 0.4N $K_2Cr_2O_7$ oxidisable-C fraction cannot be regarded as a labile C fraction. This could explain why a poor relationship was observed between $K_2Cr_2O_7$ -C and the amount of CO₂ respired from the control soils. Soil C extracted by 0.5M K_2SO_4 was assumed to be a soluble C fraction, however, a weak and insignificant relationship (R^2 =0.01) was observed between K_2SO_4 -C and the amount of CO₂ respired from the control soils during the 129 days of laboratory incubation.

Results of the regression analysis in Table 5.7 A and Figures 5.4 a and b show that neither MBC or HWC labile-C fractions could be used to predict CO_2 emissions from different soils due to management practices, or in experimental incubation of four soils with different ryegrass residue additions. This result contrasts with other studies (Ghani *et al.* 2003; Melero *et al.* 2006), which have shown that these labile-C pools could be used as sensitive indicators of soil-C change due to contrasting tillage practices and/or different cropping systems over significant periods of time on the same soil.

To explain why the labile-C tests fail to predict the soil CO_2 -C emissions due to management practices it is instructive to consider the analysis of variance results for both the amounts of carbon extracted by the two tests, MBC and HWC, and for the total CO_2 -C evolved from the experimental incubation of ryegrass residues in four different soil types. In this study the amounts of labile-C measured by MBC and HWC were strongly influenced by soil type, which explained 69.1 and 81.6% of the variation respectively, and were less influenced by the amount of ryegrass residues, which only accounted for 14.0 and 27.2% of the variance, respectively (Table 5.13). However, emissions of CO_2 -C from these soils were

more strongly affected by the amount of residue added (Figure 5.3) explaining 88.9 % of the variation in total CO₂-C emissions, whilst soil type only explained 8.8% (Table 5.13). This difference in the relative influence of soil type and residue rates on labile-C fractions and CO₂-C emissions explains why the labile-C fractions were poor indicators of CO₂-C emissions.

5.5 Summary and conclusions

Laboratory incubation was conducted to assess the differences in the decomposition of ryegrass pasture residues in different soil types. Four soils (Carnarvon SL, Ohakea stony SiL, Ohakea SiL and an allophanic Horotiu SiL) received ryegrass pasture residues at 0, 5.11, 6.81, and 8.51 mg C g^{-1} oven dried soil, equivalent to 0, 12, 16 and 20 Mg oven-dried residues ha⁻¹ and incubated in 1.8L Agee jars at 80% of field capacity for about a 4 month period. The amount of ryegrass pasture residues decomposed was estimated from the amount of CO₂ emitted during the decomposition. Over the first 60 days of incubation, ryegrass pasture residues were rapidly mineralised in all the soils and subsequent decomposition was slow. In this study, the proportion of ryegrass residues decomposed did not differ between differences in the soil texture/type and were independent of the rates of residue addition.

An investigation to correlate the labile C fractions with the CO₂-C respired was conducted during this incubation period. Labile C fractions (HWC, MBC, H₂O₂-C, K₂Cr₂O₇-C and K₂SO₄-C) were determined on day zero before beginning the incubation study to correlate these fractions with the amount of CO₂-C evolved from the soils during 129 days' incubation period. None of the labile C fractions except MBC showed any significant relationship with the CO₂-C respired during incubation of control soils. Results of the present study suggest that it is not possible to derive a single relationship using any of the labile-C fractions to predict CO₂ loss from different soil types due to a change in soil management practices.

5.6 References

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Chapter-6

Short term test plus development of predictive decomposition simulation model

6.1 Introduction

As discussed in Chapter-5, the measured labile soil C fractions (hot water extractable-C, microbial biomass-C, hydrogen peroxide oxidisable-C, potassium dichromate oxidisable-C and potassium sulphate extractable-C) were not accurate predictors of the amount of CO₂-C respired from soils during the four month laboratory incubation study. Therefore, it was considered that CO₂ emitted during a short period of incubation may be related to the total amount of CO₂ emitted in both the laboratory incubations and in turn related to the amount of CO₂ emitted in the field. This principle is the same as used to predict the quantity of mineralisable N present in cropped soils from short-term laboratory incubation (Gianello and Bremner 1986). To extend this concept further to predict CO₂ loss from conventional and No-tillage cultivation from laboratory CO₂ emission data a soil C decomposition simulation model is required.

Two most commonly used soil organic matter (SOM) decomposition simulation models are Roth-C (Jenkinson 1990) and Century (Parton et al. 1987). These models are designed for large spatial and temporal scales to estimate soil-C dynamics on regional and global scales (year time steps). The concepts used in these models to simulate the decomposition of SOM pools having different rates of decomposition has proved effective to model the long term dynamics of soil C (Smith et al. 1997). It is necessary that shorter time step models of SOM decomposition are developed and validated under field conditions (Leite et al. 2009; Lei et al. 2006; Bruun et al. 2003) to simulate short term (hourly or daily) CO₂ variations. In this study, periods of study did not exceed 99 days for a season and the variable that was used to valid the model was CO₂-C flux from a four hour measurement period. It was beyond the scope of this study to attempt to modify either the Roth-C or Century models to simulate pasture and crop residue C decomposition within a season. For this purpose, the carbon pool concepts used in the longer-term models were used to develop a three compartment model for a short-term simulation of the daily CO₂ emissions observed in the laboratory and field. La Scala Jr et al. (2009) developed a single compartment model to describe short-term CO₂ losses after tillage, however it was clear at a very early stage of data simulation that a single compartment model could not simulate the very rapid emission

of CO_2 in the first 15 days, the slightly reduced rates from 16 to 60 days (Figure 6.3) and the slow rates of emission beyond 60 days.

The hypothesis is that the amount of CO_2 emitted from short term laboratory incubation can be used to predict the field soil C loss either by simple regression analysis or by a more complex relationship involving the key factors influencing C loss within the context of a soil C decomposition simulation model.

6.2 Methodology

6.2.1 Simple regression model

Carbon dioxide emissions measured under laboratory conditions from simulated tillage (ST) and No-tillage (NT) *in-situ* soil cores (Chapter-4 section 4.3.1) were used to predict soil-C loss in field during the summer season. All replicates used to measure CO_2 fluxes under laboratory conditions and in the field were used in the regression analysis. For field comparisons, each replicate used in the laboratory measurements represents the position of the static chamber installed in the field for both the RT and NT treatments.

In brief, to compare two tillage treatments: simulated-tillage (ST) and No-tillage (NT) and a non-disturbed (ND) control, *in-situ* soil cores (10 cm diameter, 10 cm depth) were collected at a soil depth of 0-10 cm from 4 different locations on each field site. Nondisturbed (ND) cores were collected before the start of field tillage operations and the NT treatment cores were collected over the slots immediately after the Cross Slot® No-tillage drilling. Each treatment comprised four replicates and each replicate was composed of three intact cores collected from one sampling location. Half of the total numbers of ND cores collected from each field site were broken up in the laboratory to simulate tillage treatment. The ST treatment was formulated from three ND soil cores by emptying the soil, breaking it into pieces, thoroughly mixing and packing it in a plastic container. Moisture contents in the cores and container as sampled from the field were maintained throughout the experiment by weighing the soil cores and spraying the required amount of deionised water onto the surface. Soil *in-situ* cores and plastic containers filled with soil were placed in closed base static chambers and CO₂ measurements were taken on a daily basis at constant temperature (23[°]C) by placing a plastic petri dish containing 30 ml of 1M NaOH within each chamber for a 4 hour period. Carbon dioxide measurements continued for 92 days (Glen Oroua), 83 days (Tangimoana), 81 days (Kiwitea), 54 days (Feilding) and 99 days (Sanson) until the emissions subsided and the differences between the treatments became negligible.

Least squares' fitting of the CO_2 -C evolved until days 2, 4, 6, 8, 10 and 12 with total amounts of CO_2 -C evolved during the full incubation period were used to determine the duration of incubation.

6.2.2 Two and three compartment decomposition models

Tillage induced CO_2 -C fluxes have been shown to be important over short durations (Reicosky and Lindstrom 1993, Rochette and Angers 1999, La Scala *et al.* 2001, 2006) and are primarily related to the decay of the labile C fraction which has more rapid turnover than the total soil-C (La Scala Jr *et al.* 2009). Tillage also changes the soil conditions i.e. improved oxygen, temperature and moisture contents required for rapid decomposition (Six *et al.* 1998). Therefore, to build models that simulate CO_2 emissions in field soils with varying amounts of C in crop residues and more mature SOM, and to eventually accommodate the environmental effects of varying soil temperature and moisture, compartmented model structures based on the principle of the classical five compartment Roth-C model (Jenkinson 1990) were constructed.

6.2.2.1 Development of a temporally dynamic two compartment model using laboratory data

In the proposed two compartment dynamic model CO_2 fluxes are expressed in terms of C so they can be directly related to C present in the soil. In the two compartment model, the two compartments are an active crop or pasture residue pool and a more stable, soil C pool. It was assumed that the size of the active pool was negligible in comparison to the stable-C pool; therefore, the initial stable-C pool in the two compartment model was sized based on the measured total soil-C. Values for decay constants (for active and stable-C pools) were found by iteratively varying each value to maximize the coefficient of determination between the predicted (modelled) and observed 12 days daily flux values during laboratory incubations. After several iterations using Microsoft Excel 2010 for the five different soil types, standard size of active pool and decay constants for active and stable-C pools were fixed as described:

The two compartment model to describe the daily flux rate of CO₂ from tilled soil:

 $F_n = (C_n x k_c) + (A_n x k_a)....(Eq. 6.1)$ Where:

 F_n = Flux rate on any day n (Mg C ha⁻¹ d⁻¹)

Stable-C pool (Mg C ha⁻¹) is represented as C (pool-C), the initial stable-C pool value was determined by multiplying total-C concentration by soil bulk density and its subsequent values i.e. C_n on any day n was determined using the following formula:

$$C_n = [C_{(n-1)} - (C_{(n-1)} \times k_c)]$$

The most labile or active-C pool (crop or pasture residues Mg C ha⁻¹) is represented as A (pool-A). A_n on any day n was determined using the following formula:

$$A_n = [A_{(n-1)} - (A_{(n-1)} \times k_a)]$$

The decay constants k_c and k_a were estimated experimentally based on the best fit of the model to measured CO₂-C fluxes during the 12 day laboratory incubation of the simulated tillage treatment of each of the five soils used in the laboratory incubation study (Chapter-4). The model was then used to predict emissions for the full term incubations.

The initial assumptions were that the pool-C decay constant (k_c) was 0.0005 per cent of the initial size of pool-C per day and was assumed to be constant for all the soils irrespective of soil type. For example, if the initial size of the stable-C pool for a particular soil is 44.4 Mg ha⁻¹, it will decay at 0.00022 d⁻¹.

The size of pool-A was based on the amount of C lost as CO_2 during 12 days incubation from disturbed/simulated tilled soils under controlled laboratory conditions. For pool-A, the decay constant (k_a) was fixed as 0.10 d⁻¹ and was assumed to be constant for all the soils irrespective of the soil type. For the No-tillage treatment the decay constant for pool-A was allowed to vary in the two and three compartment models.

The daily decay constant values equate to < 0.2 per cent of the stable-C pool which is similar to the values used in the models calculating annual C change (Bayer *et al.* 2006; Leite *et al.* 2009). The fast pool decomposes very quickly and cannot be equated with annual models.

The two compartment model does not allow the transfer of C from the stable pool to the labile pool for the period of decomposition. To predict the CO_2 emissions from the decomposition of crop residues over a short period of time it may not be necessary to include the C transfer between pools.

Variations to a three compartment model with and without temperature and moisture are discussed in the Results and Discussion section 6.3 of this chapter.

6.2.2.2 Model efficiency (ME) also known as one of the expressions of R^2 (coefficient of determination) in non-linear fitting evaluations, was calculated by the following formula as stated by La Scala Jr *et al.* (2009):

$$ME = 1 - \frac{\sum_{t=1}^{n} (F_t^{obs} - F_t^{pred})^2}{\sum_{t=1}^{n} (F_t^{obs} - \overline{F}_t^{obs})^2}$$

Where:

 F_t^{obs} is the observed CO₂-C flux

 \bar{F}_t^{obs} is the mean of observed CO₂-C fluxes throughout the measurement period

 F_t^{pred} is the predicted CO₂-C flux

Model efficiency/ \mathbb{R}^2 will vary between minus infinity and 1 with higher values (closer to 1) indicative of superior performance.

6.3 Results and discussion

6.3.1 Simple regression model

The least squares regression analysis (Table 6.1) showed a significant relationship between the total amounts of CO₂-C respired during the full incubation period (54 to 99 days) with the CO₂-C evolved until days 2, 4, 6, 8, 10 and 12. Amongst the days, day-12 showed the highest R-square value and lowest residual standard error compared to day-2, 4, 6, 8 and 10. Therefore, the 12 day laboratory incubation (i.e. the total amount of CO₂-C evolved to day-12) was proposed to be a good predictor of C lost from the ST and NT soils of the five soil sites used during laboratory incubation.

McLauchlan and Hobbie (2004) also suggested 12 days of laboratory incubation as the most accurate technique to estimate labile-C because the majority of the material being mineralized during this period is labile and it closely resembles natural decomposition under field conditions. Moreover, a commercial test cannot be longer because it has to provide timely advice to farmers after spraying glyphosate herbicide.

Table 6.1: Linear regression coefficients of deermination comparing the CO_2 -C evolved during full laboratory incubation period (54 to 99 days) with CO_2 -C evolved; until day 2, 4, 6, 8, 10 and 12 using soils from five soil sites.

Days	Simulated	tillage (ST)	No-till	age (NT)
Day-2	$R^2 = 0.46*$	RSE=0.61	$R^2 = 0.68*$	RSE=0.42
Day-4	$R^2 = 0.73^*$	RSE=0.43	$R^2 = 0.81*$	RSE=0.32
Day-6	$R^2 = 0.85*$	RSE=0.32	$R^2 = 0.88*$	RSE=0.26
Day-8	$R^2 = 0.90*$	RSE=0.26	$R^2 = 0.91*$	RSE=0.22
Day-10	$R^2 = 0.93*$	RSE=0.22	$R^2 = 0.93*$	RSE=0.19
Day-12	$R^2 = 0.96*$	RSE=0.17	$R^2 = 0.94*$	RSE=0.18

*p<0.001 RSE= residual standard error

The linear regression (Figures 6.1 and 6.2) showed a strong relationship ($R^2=0.87$) between the total CO₂-C evolved from simulated tillage (ST) and No-tillage (NT) soils until day-12 with the full laboratory incubation period (54 to 99 days) for the five soil sites. However, a weak relationship ($R^2=0.35$) was observed when the total CO₂-C evolved from simulated tillage (ST) and No-tillage (NT) soils until day-12 under laboratory conditions was compared with CO₂-C evolved from rotary tillage (RT) and No-tillage (NT) soils for the full measurement period (99 days) during the summer season from the Sanson field site.

Separate linear regression models were fitted for the ST and NT treatments for both the laboratory (Figure 6.1) and field (Figure 6.2) comparisons and were analysed for the differences of slopes and intercepts between treatments using analysis of covariance. No significant differences were observed for the slopes (p=0.88 for the laboratory (Figure 6.1); p=0.77 for the field comparison (Figure 6.2)) and intercepts (p=0.06 for the laboratory (Figure 6.1); p=0.35 for the field comparison (Figure 6.2)) between the NT and ST treatments. Therefore it was decided to use a single regression model for both the treatments.

Due to variations in field soil moisture and temperature, CO_2 -C emissions observed in the field were higher than observed in laboratory. This resulted in a weak relationship between laboratory and field measurements. The complex process of CO_2 production and release from soils could not be explained without taking into account the soil temperature and moisture content (Wildung *et al.* 1975). Wagner *et al.* (1997) observed that a linear regression model underestimates the CO_2 flux rates and suggested developing a model based on the complex physical mechanisms involved because the production of CO_2 in soils is often nonlinear. Therefore, the concept that field soils being brought into the laboratory, cultivation treatment simulated and the CO_2 -C evolved during 12 day incubation being used to predict field soil C loss appears to be unworkable if field soil climate conditions cannot be simulated. For this reason, attention was turned to testing two or three compartment time dependent models that could be made to respond to temporal climate change.



Figure.6.1: The relationship between the total CO₂-C evolved from simulated tillage and No-tillage soils until day-12 with CO₂-C evolved during full incubation period (54 to 99 days) from the five soil sites during laboratory incubation study.



Figure.6.2: The relationship between the total CO_2 -C evolved from simulated tillage and No-tillage soils until day-12 under laboratory conditions with total CO_2 -C evolved from rotary tillage (RT) and No-tillage (NT) soils in field for the full measurement period (99 days) during summer for the Sanson field site.

6.3.2 Application of two compartment model to predict CO₂ fluxes in laboratory for full measurement period

Results of developing a CO_2 -C flux model by fitting the 12 day laboratory incubation data and then predicting CO_2 -C fluxes for full measurement period (54-99 days) for the five soil sites for simulated tillage (ST) treatment with a two compartment model are presented in Tables 6.2 and 6.3, respectively.

The coefficient of determination (\mathbb{R}^2) for simulated tillage treatment showed good agreement between observed and predicted (modelled) CO₂-C fluxes for the full measurement period for most of the soil sites (Table 6.3). However, fitting for the full measurement period CO₂-C fluxes from simulated tillage treatment in the laboratory study showed that the model starts under-estimating the fluxes after a few days suggesting that the simulated flux rates needs to be increased. The predicted (modelled) CO₂-C fluxes when plotted with observed fluxes for the Glen Oroua site (Figure 6.3c) shows the underestimation of predicted fluxes by the two compartment model for the simulated tillage treatment; figures for the remaining sites are not shown. Therefore the third pool i.e. the intermediate pool was included in the two compartment model resulting in the three compartment model.

The three compartment model to describe the daily flux rate of CO_2 from tilled soil: $F_n = (C_n x k_c) + (A_n x k_a) + (B_n x k_b)..... (Eq. 6.2)$ Where:

 F_n = Flux rate on any day n (Mg C ha⁻¹ d⁻¹)

Pools C & A and their respective decay constants are same as described in equation 6.1.

Intermediate-C pool (Mg C ha⁻¹) is represented as B (pool-B); initial size was set as:

Pool-B (Initial) = Pool-A (Initial) x 1.5 and its subsequent values i.e. B_n on any day n were determined using the following formula:

 $B_n = [B_{(n-1)} - (B_{(n-1)} \times k_b)]$

The size and decay constant (k_b) of pool-B was estimated experimentally based on the best fit of the model to measured CO₂-C fluxes during the 12 days incubation of the simulated tillage treatment of each of the five soils used in laboratory incubation study.

For pool-B, decay constant (k_b) was assumed to be 1.0 per cent of initial size of pool-B per day. For example, if the initial size of pool-B for particular soil is 1.22 Mg ha⁻¹ it will decay at 0.0122 d⁻¹ and is assumed to be constant for all the soils irrespective of soil type.

A factor of 1.5 used to estimate pool-B from pool-A gave the best fit to the 12 days observed laboratory data.

The decay constants were set on the initial pool sizes.

Figure 6.4 shows the contribution of pools A, B and C to CO_2 fluxes in two and three compartment models. Inclusion of intermediate pool (pool-B) in the two compartment model (i.e. the three compartment model) better fitted the laboratory data for ST treatment (Figure 6.3 d) and (Tables 6.4 and 6.5).

The model was further adjusted by changing the decay constant of pool-A to predict the Notillage laboratory data.

The three compartment model to describe the daily flux rate of CO_2 from No-tilled soil:

 $F_n = (C_n x k_c) + (A_n x k_a/2) + (B_n x k_b)....(Eq. 6.3)$

The model parameters described in equation 6.2 for tilled soils were used to predict CO_2 -C fluxes from No-tilled soils only by varying the value of decay constant (k_a) of pool-A. The decay constant value (k_a) of pool-A was estimated experimentally based on the best fit of the model to measured CO_2 -C fluxes during the 12 days' incubation of the No-tillage treatment of each of the five soils used in the laboratory incubation study. The value of the decay constant (k_a) of pool-A to predict the CO_2 -C fluxes from the No-tillage treatment was half the value used to fit the 12 days' CO_2 -C fluxes observed from simulated tillage treatment in the laboratory.

The adjusted three compartment model very well fitted the 12 days and full measurement laboratory data for the NT treatment (Tables 6.4 and 6.5).

measured and predicted (modelled) CO₂-C fluxes for 12 days of the five soils used in laboratory incubation study for developing the two Table 6.2: Two carbon pools (Pool-C and A), their respective decay constants (k_c and k_a) and coefficient of determination (R²) of the daily compartment model.

	I			I	
\mathbb{R}^2	0.63	0.17	0.39	0.76	0.63
k_{a}	0.10	0.10	0.10	0.10	0.10
kc	0.00022	0.00021	0.00032	0.00018	0.00021
Pool-A (Mg C ha ⁻¹)	0.81	0.97	0.80	0.45	0.76
Pool-C (Mg C ha ⁻¹)	44.40	42.40	64.60	35.80	42.40
Tillage treatment	ST	LS	LS	ST	ST
Soil sites	Glen Oroua	Tangimoana	Kiwitea	Feilding	Sanson

Table 6.3: Coefficient of determination (R²) of the daily measured and predicted (modelled) CO₂-C fluxes for the full measurement period of the five soils used in laboratory incubation study after application of the two compartment model.

Coil aitee	Tillage	Pool-C	Pool-A	4	4	D ²	
	treatment	(Mg C ha ⁻¹)	(Mg C ha ⁻¹)	N c	Кa	2	
Glen Oroua	LS	UV VV	0.81	0.0007	0.10	0.83	
(92 days)	10	44.40	10.01	77000.0	01.0	0.07	
Tangimoana	L'N	UV CV	0.07	1 0000 0	0.10	0.13	
(83 days)	10	42.40	16.0	17000.0	01.0	C1.U	
Kiwitea	ЦIJ	כע כט	0 00		0.10	92.0	
(81 days)	10	04.00	0.00	700000	01.0	00	
Feilding	сT	35 00	0.45	0.00010	0.10	0 71	
(54 days)	10	00.00	0.4.0	01000.0	0.10	0.71	
Sanson	сT	UV CV			0.10	0 05	
(99 days)	10	42.40	0.70	0.00021	01.0	0.0	
ST= Simulated tilla	lge						

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Figure.6.3: Comparison of observed and predicted (modelled) CO_2 -C fluxes by two and three compartment models for 12 days (a, b) and full 92 days (c, d) measurement period for the Glen Oroua site for the simulated tillage (ST) treatment.



Figure.6.4: Carbon dioxide contribution from pools-A & C of (a) two compartment model and pools-A, B and C of (b) three compartment models for the simulated tillage (ST) treatment for the Glen Oroua soil site.

Table 6.4: Three carbon pools (Pool-C, A and B), their respective decay constants (k_c , k_a and k_b) and coefficient of determination (\mathbb{R}^2) of the daily measured and predicted (modelled) CO₂-C fluxes for 12 days of the five soils used in laboratory incubation study for developing the three compartment model.

Soil sites	Tillage treatment	Pool-C (Mg C ha ^{-l})	Pool-A (Mg C ha ⁻¹)	Pool-B (Mg C ha ⁻¹)	kc	ka	k _b	\mathbb{R}^2
	ST	44.40	0.81	1.22	0.00022	0.10	0.0122	0.82
OIGII OLOUG	NT	44.40	0.81	1.22	0.00022	0.05	0.0122	0.46
Toncino	ST	42.40	26.0	1.46	0.00021	0.10	0.0146	0.58
1 aligililualia	NT	42.40	26.0	1.46	0.00021	0.05	0.0146	0.44
Vt	ST	64.60	08.0	1.21	0.00032	0.10	0.0121	0.61
NIWIICa	NT	64.60	08.0	1.21	0.00032	0.05	0.0121	0.27
Eo:14:6 %	ST	35.80	0.45	89.0	0.00018	0.10	0.0068	0.91
renuing	NT	35.80	0.45	0.68	0.00018	0.05	0.0068	0.44
Concon	ST	42.40	0.76	1.14	0.00021	0.10	0.0114	0.76
IIUGIIBC	LΝ	42.40	0.76	1.14	0.00021	0.05	0.0114	0.48

ST= Simulated tillage NT= No-tillage

Table 6.5: Coefficient of determination (\mathbb{R}^2) of the daily measured and predicted (modelled) CO₂-C fluxes for full measurement period of the five soils used in laboratory incubation study after application of the three compartment model.

Soil sites	Tillage treatment	Pool-C (Mg C ha ⁻¹)	Pool-A (Mg C ha ⁻¹)	Pool-B (Mg C ha ⁻¹)	kc	ka	k _b	\mathbb{R}^{2}
Glen Oroua	ST	44.40	0.81	1.22	0.00022	0.10	0.0122	0.95
(92 days)	NT	44.40	0.81	1.22	0.00022	0.05	0.0122	0.89
Tangimoana	ST	42.40	0.97	1.46	0.00021	0.10	0.0146	0.76
(83 days)	NT	42.40	0.97	1.46	0.00021	0.05	0.0146	0.88
Kiwitea	ST	64.60	0.80	1.21	0.00032	0.10	0.0121	0.84
(81 days)	NT	64.60	0.80	1.21	0.00032	0.05	0.0121	0.72
Feilding	ST	35.80	0.45	0.68	0.00018	0.10	0.0068	0.91
(54 days)	NT	35.80	0.45	0.68	0.00018	0.05	0.0068	0.76
Sanson	ST	42.40	0.76	1.14	0.00021	0.10	0.0114	0.92
(99 days)	NT	42.40	0.76	1.14	0.00021	0.05	0.0114	0.64

ST= Simulated tillage NT= No-tillage

6.3.3 Application of temporally dynamic three compartment model to field data

After application of the three compartment model to predict CO_2 -C fluxes under laboratory conditions, the model was applied to predict CO_2 -C fluxes under field conditions. Carbon dioxide measurements were made during autumn and summer seasons from the Sanson field site. During the summer season simultaneous CO_2 -C measurements were made on *in-situ* soil cores in the laboratory along with field measurements. The model parameters derived from the summer laboratory incubation were used to predict summer season CO_2 -C fluxes. Laboratory CO_2 -C measurements were not conducted for the *in-situ* soil cores collected during the autumn season as the *in-situ* soil cores were placed in the field (Chapter-4 section 4.3.2). Therefore the model was tested only for the summer field measurements.

Under laboratory conditions of constant temperature and soil moisture, CO₂-C fluxes measured over 4 hours were extrapolated to 24 hour daily CO₂-C fluxes, therefore, the decay constants were set on 24 hour daily flux measurements. The three compartment model was restructured to predict the CO₂ evolved during the 4 hour measurement period i.e. the decay constants were adjusted according to the 4 hour measurement period rather than the 24 hour daily flux in the laboratory to model CO₂-C fluxes in field. This change in model time step to 4 hour was made to accommodate a large diurnal change in temperature (section 6.3.4), when daily measurements were made at specific times and known temperatures and moistures. There were variations in soil temperature and moisture with rainfall in the field. Flux measurements were made between 9 am to 1pm in field and soil temperature and moisture were monitored on a regular basis.

When temperature and moisture changes were not accounted for by the three compartment model, the coefficient of determination (R^2) showed a poor agreement between observed and predicted (modelled) CO₂-C fluxes for both rotary tillage (RT) and No-tillage (NT) treatments (Figures 6.5 a, b) when applied to full length summer field measurements. In the field, CO₂-C fluxes were affected by soil moisture and temperature as large diurnal variations in soil temperature and moisture with rainfall were observed (Chapter-4, Figures 4.6b and 4.7b). This could explain the poor agreement between observed and predicted (modelled) CO₂-C fluxes for both rotary tillage (RT) and No-tillage (NT) treatments under field conditions. The observed C loss from RT and NT treatments for the 99 days measurement period in the field was 3.33 and 2.87 Mg C ha⁻¹ and varied significantly between the tillage treatments. However, the predicted (modelled) C loss from RT and NT treatments for 99 days measurement period in the field was 2.50 and 2.49 Mg C ha⁻¹ (Figures 6.6 a, b). Therefore, temperature and moisture scalars were added to the predictive model.



Figure.6.5: Comparison of observed and predicted (modelled) CO_2 -C fluxes by the three compartment model for 99 days measurement period for the Sanson site from (a) rotary tillage (RT) and (b) No-tillage (NT) treatments.



Figure.6.6: Comparison of observed and predicted (modelled) cumulative CO₂-C emissions by the three compartment model for 99 days measurement period for the Sanson site from (a) rotary tillage (RT) and (b) No-tillage (NT) treatments.

6.3.4 Development of temporally and climatically dynamic three compartment model6.3.4.1 Development of temperature scalar

A range of literature based on field studies (Raich and Schlesinger 1992; Lloyd and Taylor 1994; Lenton and Huntingford 2003) and laboratory incubation studies at controlled temperature and soil moistures excluding roots (Kirschbaum 1995, 2000) was reviewed to determine an appropriate temperature response function to be used in the three compartment model so that predicted CO_2 fluxes from laboratory incubations could be made responsive to field temperature. Soil respiration measurements under natural field conditions cannot provide an unbiased estimate of the temperature dependence of organic matter decomposition due to the confounding effects of soil moisture and seasonal substrate availability; moreover the contribution of root respiration was not separated from that of soil microorganisms and fauna in field studies (Kirschbaum 2006). Therefore, it was decided to use the temperature response function obtained (Kirschbaum 2000) under highly controlled laboratory conditions to predict soil respiration under field conditions.

The temperature dependence of decomposition of carbon pools following equation 6.4 was used in the three compartment model. The effect of temperature on decomposition rate of each carbon pool in equation 6.4 is modelled by the response function of Kirschbaum (2000), normalised at a reference temperature of 30° C as stated by Corbeels *et al.* (2005).

$$r_T = exp\left[\frac{3.90(Ts-30)}{Ts+31.79}\right]$$
....(Eq. 6.4)

Where: T_s (°C) represents the soil temperature and 31.79 represents a constant derived by fitting the laboratory incubation data by Kirschbaum (2000).



Figure.6.7: Temperature scalar for the three compartment model showing relative change in CO_2 fluxes (r_T) normalised to account for laboratory CO_2 -C flux measurements being made at a constant temperature of 23°C.

The temperature response function (r_T) was interpolated over the range of soil temperatures (averaged over 4 hours) measured in the field during which 4 hour CO₂-C flux measurements were made (Figure 6.7). The decay constants (Table 6.5) used in the three compartment model accurately described CO₂ evolution in the laboratory at 23°C constant temperature. Therefore, these r_T values need normalising about 23°C. A temperature scaling factor (T_{sl}) was derived by equating the r_T values calculated from equation 6.4 for 23°C to 1 and is presented as secondary axis in Figure 6.7. The equation 6.4 used for calculating r_T values does not take into account the soil temperature and moisture interaction.

6.3.4.2 Development of moisture scalar

Soil moisture is an important abiotic driver of soil C dynamics (O' Brien *et al.* 2010). Importance of moisture content has been demonstrated in temperature controlled laboratory studies where roots are excluded (Davidson *et al.* 1998). Variations in soil moisture affect the diffusion of soluble substrates at lower moisture content (Kechavarzi *et al.* 2010) whereas high moisture content limits the diffusion of oxygen (Hashimoto and Komatsu 2006; Skopp *et al.* 1990; Linn and Doran 1984). Owing to confounding effects of soil

moisture and temperature on soil respiration, application of laboratory based understanding of the effects of soil moisture on soil respiration to field studies has not been fully achieved (Reichstein *et al.* 2005; Davidson *et al.* 2000, 1998). Several empirical relationships have been established between field soil respiration and soil moisture and temperature (Almagro *et al.* 2009; Yuste *et al.* 2007; Xu *et al.* 2004). However, these relationships were site specific hence no appropriate soil moisture function was found from the literature. Therefore, the field data was scrutinised and a period where soil temperature was approximately constant but moisture varied was selected. The first step was to find a relationship between CO_2 -C fluxes and moisture content. The CO_2 -C flux values measured from rotary and Notillage treatments during that period were plotted against the observed gravimetric moisture content (GMC) of soil in the field and polynomial regression equations were fitted to the data (Figures 6.8 a, b):

$$y = -2E-05x^{2} + 0.0011x - 0.0074$$
 (Figure 6.8 a)
 $y = -5E-07x^{2} + 0.0003x - 0.0014$ (Figure 6.8 b)

The second step was to normalise the data to produce a moisture scalar. The decay constants used in the three compartment model accurately described CO_2 evolution in the laboratory at constant ~15% GMC for Sanson site soil. Therefore, the predicted CO_2 -C flux values derived from these polynomial equations were normalised by equating for 15.0% GMC to 1 and is present as secondary axis in Figures 6.8 a, b. The relationship for the scalar values for different moisture contents are given in the equation 6.5 and 6.6 and also shown in Figures 6.8 a, b.

Moisture factor (w_s) = -0.0043 x^2 + 0.2391x - 1.6087 (Figure 6.8 a)...... (Eq. 6.5) Moisture factor (w_s) = -0.0002 x^2 + 0.1004x - 0.4686 (Figure 6.8 b)...... (Eq. 6.6)

A small decrease in the soil CO_2 -C flux values (Figure. 6.8 a, b) may be attributed to the development of anaerobic conditions in the soil which would have decreased the aerobic microbial activity. Linn and Doran (1984) also observed a decrease in the aerobic microbial activity with increasing soil moisture content.

These temperature and moisture functions were constrained within the observed (measured) field soil temperature and moisture values.



Figure.6.8: Moisture scalars shown by dotted lines for the three compartment model derived from field observations of CO₂-C fluxes at a range of soil moisture contents in (a) rotary tillage (RT) and (b) No-tillage (NT) treatments. Solid lines represent the relationship between the measured CO₂ fluxes and GMC's of soil in the field.

The three compartment model including moisture and temperature factors to describe the flux rates of CO_2 from tilled soil:

 $F_n = (T_{sl} x w_s x ((C_n x k_c) + (A_n x k_a) + (B_n x k_b)))....(Eq. 6.7)$ Where:

 F_n = Flux rate for 4 hour period n (Mg C ha⁻¹ 4hr⁻¹)

Stable-C pool (Mg C ha⁻¹) is represented as C (pool-C), initial stable-C pool value was determined by multiplying total-C concentration by bulk density and its subsequent values i.e. C_n for 4 hour period n were determined using the following formula:

 $C_n = [C_{(n-1)} - (T_{s1} x w_s x (C_{(n-1)} x k_c))]$

The most labile or active-C pool (Mg C ha⁻¹) is represented as A (pool-A), initial size was set to be the amount of C lost as CO_2 during 12 days incubation from disturbed/simulated tilled soils under controlled laboratory conditions and its subsequent values i.e. A_n for 4 hour period n were determined using the following formula:

 $A_{n} = [A_{(n-1)} - (T_{s1} x w_{s} x (A_{(n-1)} x k_{a}))]$

Intermediate-C pool (Mg C ha⁻¹) is represented as B (pool-B); initial size was set as;

Pool-B (Initial) = Pool-A (Initial) x 1.5 and its subsequent values i.e. B_n for 4 hour period n were determined using the following formula:

 $B_{n} = [B_{(n-1)} - (T_{s1} x w_{s} x (B_{(n-1)} x k_{b}))]$

The three compartment model including moisture and temperature factors to describe the flux rates of CO_2 from No-tilled soil:

 $\mathbf{F_n} = (\mathbf{T_{sl} x w_s x} ((\mathbf{C_n x k_c}) + (\mathbf{A_n x k_a/2}) + (\mathbf{B_n x k_b}))).....(Eq. 6.8)$ The model parameters described in equation 6.7 for the tillage treatment were used to predict CO₂-C fluxes from No-tillage treatment only by changing the value of decay constant (k_a) of pool-A and moisture factor. The value of decay constant (k_a) of pool-A to predict the CO₂-C fluxes from No-tillage was half the value used to fit the CO₂-C fluxes measured from the tillage treatment.

T_{sl} = Temperature factor	(Eq.	6.4)
w _s = Moisture factor	. (Eq.	6.6)

6.3.4.3 Application of temporally and climatically dynamic three compartment model to field data

The three compartment model including temperature and moisture factors, when applied to the full length summer field measurements showed improvement in the coefficient of determination (\mathbb{R}^2) between observed and predicted (modelled) CO₂-C flux values for both rotary tillage ($\mathbb{R}T$) and No-tillage ($\mathbb{N}T$) treatments (Figures 6.9 a, b) in comparison to when the model was applied without these factors (Figures 6.5 a, b).

The observed C loss from RT and NT treatments during the 99 days measurement period in field was 3.33 and 2.87 Mg C ha⁻¹, and varied significantly between the tillage treatments. However, the predicted (modelled) C loss from RT and NT treatments was 2.30 and 2.80 Mg C ha⁻¹ (Figures 6.10 a, b). The model accurately predicted the amount of C lost from NT treatment but the predicted amount for the RT treatment was 30 per cent less than the amount that was lost from the RT treatment in field. Moreover, the predicted amounts of C lost were higher for NT than RT treatment which was contrary to the findings from the field study. For the RT treatment, the model underestimated the higher peaks of CO₂-C fluxes observed after rainfall events thus explaining why the predicted C loss for the RT treatment was less than the observed C loss. High CO₂-C fluxes after rainfall events were thought to be the result of increased mineralization of the lysed microbial cells caused by desiccation or due to rapid rewetting (Magid et al. 1999; Fierer and Schimel 2003) and moisture stimulated microbial activity (Kessavalou et al. 1998). However, the model predicted the CO₂-C fluxes well in the NT treatment. Since moisture content remained higher in the NT than RT treatment throughout the measurement period (Chapter-4, Figure 4.7b), wetting and drying may not have caused severe microbial lyses and enhanced microbial activity and CO₂-C loss in the NT treatment. Moreover the moisture scalar in the model predicted higher CO₂-C fluxes due to higher moisture contents in the NT treatment. Thus the model is unable to predict the CO₂-C loss from the RT soil.

The current three compartment model selects the carbon pools on a chemical analysis basis and do not take the dynamic nature of microbial biomass into account. Further research work is required if the three compartment decomposition model is to be improved to simulate short term C loss from NT and RT soils within the season of tillage. It may be advantageous to estimate the crop or pasture residue carbon and use that to determine rapidly decomposing pool sizes. In addition to accommodating wetting and drying it will be necessary to incorporate a microbial biomass pool in the model which can increase as residue carbon is decomposed but decrease when soil moisture drops below wilting point and when

the soil subsequently wets up and becomes a pool of rapidly decomposing C. Therefore in future studies, an improvement in model development would arise if laboratory incubations are conducted at a range of soil temperatures and moistures and microbial biomass measurements are made on a frequent basis to quantify the microbial biomass C pool. This will produce independent scalars for temperature and moisture that could be validated on the field data.

The data obtained during this Ph.D. study was not sufficient to provide or develop a model that could be used to predict CO_2 loss from conventional and No-tillage cultivation in New Zealand soils based on a 12-day laboratory incubation.



Figure.6.9: Comparison of observed and predicted (modelled) CO₂-C fluxes by the three compartment model including temperature and moisture factors for the Sanson site from (a) rotary tillage (RT) and (b) No-tillage (NT) treatments for 99 days measurement period.



Figure.6.10: Comparison of observed and predicted (modelled) cumulative CO_2 -C emissions by the three compartment model including temperature and moisture factors for the Sanson site from (a) rotary tillage (RT) and (b) No-tillage (NT) treatments for 99 days measurement period.

6.4 Summary and conclusions

The concept was to use the amount of CO_2 emitted from the 12 days laboratory incubation to predict the field soil C loss due to conventional and No-tillage practices. Firstly a simple regression model was used and then complex two and three compartment models were used to predict the soil C loss. The coefficient of determination (R^2) using regression model showed a strong relationship between CO_2 -C emitted in 12 days and full laboratory incubation period for both simulated tillage (ST) and No-tillage (NT) treatments but a poor relationship was observed for CO_2 -C emitted during the full measurement period in the field for rotary (RT) and No-tillage (NT) treatment, suggesting the need for complex two and three compartment models to simulate CO_2 -C emissions in field varying in soil temperature and moisture.

The two compartment model comprising active and stable-C pools, developed from 12 days laboratory incubation data of ST treatment when applied to predict full incubation laboratory fluxes for ST treatment underestimated the CO_2 -C fluxes after few days. Therefore, to increase the simulated fluxes, an intermediate C pool was included in the two compartment model resulting in the three compartment model. The decay constant of the active C pool in the three compartment model was adjusted to predict CO_2 -C fluxes for the No-tillage treatment.

The coefficient of determination (\mathbb{R}^2) using the three compartment model showed a good agreement between predicted (modelled) and observed CO₂-C fluxes from ST and NT treatments under laboratory conditions but a poor relationship was found for CO₂-C fluxes measured in field from RT and NT treatments.

To make the laboratory fluxes align to field fluxes, temperature and moisture functions were included in the three compartment model. The three compartment model including temperature and moisture functions when applied to predict CO_2 -C fluxes in field improved the coefficient of determination (\mathbb{R}^2) values for both RT and NT treatments in comparison to when the three compartment model without temperature and moisture functions was applied. The model accurately predicted the amount of C lost from the NT treatment; however, the predicted amount for the RT treatment was 30 per cent less than the amount that was lost from the RT treatment in field. The predicted amounts of C lost were higher for NT than RT treatment which was in contrast to the findings from the field study. The high peaks of CO_2 -C fluxes observed after rainfall events for RT treatment were underestimated by the applied model which resulted in lower predicted C loss than the

6.5 References

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Chapter-7

Conclusions and recommendations for future work

7.1 Conclusions

Review of the literature suggested that No-tillage seeding conserves soil C by reducing CO₂ losses compared with conventional tillage practices. Quantitative information on the reduction of CO₂ emissions with No-tillage practice in comparison to conventional tillage is limited in New Zealand. Moreover, the effects of soil and climatic conditions and amounts of plant residue at cultivation on these emissions from cropping/pasture systems under a temperate climate, such as for New Zealand, are not well documented, nor are they well quantified. Therefore, information on the reductions of CO₂ emissions with Cross Slot[®] No-tillage cultivation in comparison to the conventional cultivation operations practised in New Zealand and a process-based understanding of the soil and climatic factors controlling CO₂ losses is required. A series of laboratory and field experiments were conducted to test the hypothesis that Cross Slot[®] No-tillage cultivation is a management strategy for conserving and perhaps increasing soil C in cropping and pasture soils.

7.1.1 Selection of appropriate method for measuring CO₂ fluxes

The objective was to establish a suitable method for measuring CO_2 emissions from seedbeds. Carbon dioxide fluxes are measured by a number of techniques i.e. micrometeorological and static and dynamic chambers, with large differences in accuracy, spatial and temporal variability, and applicability. To date, chambers are the most commonly used for measuring the emissions of greenhouse gases. To establish a suitable chamber method for collecting and analysing CO_2 , three different approaches were investigated. Static alkali traps, flux gradient and a portable infra-red gas analyser (IRGA), were compared under laboratory conditions using low (sub-surface) and high (surface) CO_2 -emitting soil (Chapter-3). Based on the results of this study, the alkali trap method was selected as the most appropriate for collecting and measuring CO_2 fluxes from cultivated or No-tilled agricultural soils. Flux measurements made by EGM-1 were highly variable; moreover, this method requires numerous replicated spot measurements. The flux gradient method was sensitive to large increases in the concentration of emitted CO_2 and is expensive compared to the alkali trap.

To determine whether the impact of pressure fluctuation caused by wind velocity was a significant factor with different effects on soil CO_2 emissions from tilled or no-tilled soils,

a two week laboratory study was conducted using *in-situ* soil cores. Two tillage treatments, simulated-tillage (ST) and Cross Slot[®] No-tillage (NT), and the third treatment was a nondisturbed (ND) control. Negative pressure of 20 millibars was maintained in each replicate chamber by using an individual bubble tower. Results of the laboratory study corroborate the findings in the literature that higher CO₂ fluxes are caused by the onset of lower (negative) air pressure. The effect of pressure change on soil CO₂ fluxes differed with the amount of soil disturbance. For simulated tilled soil (ST) the effect was short-lived (4 hours) and only occurred in the first week. However, for NT and ND soils, the difference between the rate of CO₂ loss between ambient pressure and the onset of lower pressure remained for the two week period. Resulting from this study it was decided that for further studies CO₂-C measurements should be conducted at ambient pressure conditions.

7.1.2 Reduction in CO₂ emissions with No-tillage

To quantify the potential savings of soil C with Cross Slot[®] No-tillage (NT) cultivation, CO₂ fluxes were measured in the laboratory for up to 3-months from *in-situ* soil cores collected from four different sites varying in physico-chemical properties collected across Manawatu region during the summer season (December 2009 and January 2010) (Chapter-4). The total amount of CO₂ emitted under laboratory conditions from the four soils ranged between 1086 and 3264 kg CO₂-C ha⁻¹ for simulated tillage (ST), 973 and 2871 kg CO₂-C ha⁻¹ for No-tillage (NT) and 953 and 3023 kg CO₂-C ha⁻¹ for non-disturbed (ND) treatment. In general, three out of the four soils lost more CO₂ from ST (between 113 and 393 kg CO₂-C ha⁻¹) than they did for the NT treatment, although the differences were not significant at a 5% level of significance.

In order to verify that differences in CO₂ fluxes observed between ST, NT and ND treatments from laboratory situations represented the magnitude of field emissions; CO₂ measurements were also made for one of the soils at a field site near Sanson during autumn (/winter) (April to July 2010) and (spring/) summer (November 2010 to February 2011) season. The three tillage treatments compared in the field trial were: 1) tillage with rotary tiller (RT) to the depth of 10 cm followed by bar harrow, 2) direct seeding with Cross Slot[®] No-tillage. Static chambers were installed immediately over the slots after the Cross Slot[®] No-tillage seeding. In conjunction with field measurements, CO₂ measurements were made from *in-situ* soil cores. Closed-base static chambers enclosing the soil cores were placed in the field during the autumn season and in the laboratory during the summer season.

Measurements continued for 110 days during the autumn season and 99 days for the summer season. Under field conditions, total CO₂ emissions were significantly higher from RT (2580 kg CO₂-C ha⁻¹) than NT (2215 kg CO₂-C ha⁻¹) and ND (2405 kg CO₂-C ha⁻¹) treatments during the autumn season, and from RT (3330 kg CO₂-C ha⁻¹) than NT (2877 kg CO₂-C ha⁻¹) and ND (2807 kg CO₂-C ha⁻¹) treatments during the summer season at a 5% level of significance. Overall, the results of the field study suggest that Cross Slot[®] NT cultivation resulted in a combined annual conservation of ~3.0 Mg CO₂ ha⁻¹ (818 kg CO₂-C ha⁻¹) in comparison to the RT treatment from bare soils. Carbon dioxide measurements made from both the field chambers and *in-situ* soil cores placed in the field following RT/ST, NT and ND treatments were similar in magnitude (~2.5 Mg CO_2 -C ha⁻¹) during the autumn season. However, during the summer season the magnitude of CO₂ emissions from RT/ST, NT and ND treatments both in the field and laboratory were not similar. Under field conditions, carbon dioxide emissions were ~3.0 Mg CO₂-C ha⁻¹ compared with ~ 2.0 Mg CO₂-C ha⁻¹ for laboratory-simulated conditions during the summer season. Lack of variation in soil moisture and temperature in laboratory incubated soil cores resulted in the difference in magnitude of CO₂ emissions from field and soil cores. Therefore, the concept that soil cores from the field being brought into the laboratory and tillage-treatment simulated to get reliable estimates of CO₂ emissions under field conditions appears to be unworkable if field soil climate conditions are not simulated.

7.1.3 Influence of soil type and residue input rates on CO₂ emissions

The soils used in the laboratory and field experiments quantifying CO₂ emissions differed in residue amounts, texture (sandy loam and silt loam) and moisture status. Therefore a laboratory study was conducted to assess the influence of soil type on the decomposition of pasture residues (Chapter-5). Four soils: Carnarvon sandy loam, Ohakea stony silt loam, Ohakea silt loam (used in the laboratory and field study) and an allophanic, Horotiu silt loam received ryegrass pasture residues at 0, 5.11, 6.81, 8.51 mg C g⁻¹ ovendried soil equivalent to 0, 12, 16 and 20 Mg oven-dried residues ha⁻¹ and were incubated in Agee jars at 80% of their field capacity moisture content for 129 days. During the 129 days incubation, the proportion of pasture residue decomposed did not differ significantly between differences in the soil texture and the residue input rates.

In order to develop a potential soil test to predict CO_2 emissions, different labile-C fractions (HWC, MBC, H₂O₂-C, K₂Cr₂O₇-C and K₂SO₄-C) were determined on day zero i.e. before starting the laboratory decomposition study from all the four soils. All the labile-C

fractions were correlated with the total amount of CO_2 respired during the incubation period. However, none of the labile-C fractions showed strong relationships with the CO_2 respired during the incubation period. Therefore none of these laboratory soil tests can be used to predict the CO_2 losses. Hence, a combination of laboratory incubation and a predictive model to predict the CO_2 losses from conventional and No-tillage soils was explored.

7.1.4 Modelling laboratory CO₂ fluxes to predict field CO₂ emissions

In an effort to develop a CO_2 predictive model, relationships between CO_2 respired in the short term and the total amount lost during the entire measurement period following cultivation were explored (Chapter-6). Based on the CO_2 measurements done on *in-situ* soil cores under laboratory conditions and collected from different sites (Chapter-4), the amount of CO_2 respired during 12 days incubation showed a highly significant relationship with the total amount respired during the entire measurement period.

Hence, 12-day soil incubation was selected to develop a predictive model, as the commercial test cannot be longer as it has to provide timely advice to the farmer. Based on 12 day laboratory incubation data, a simple static regression model was developed. However, a weak relationship between laboratory and field data suggested that a more complex carbon decomposition compartment model was required. Therefore, two and three compartment models were developed and tested to predict full-term laboratory incubations. When tested to predict the amount of CO₂ evolved during the full-term laboratory incubation, the three compartment model gave a higher coefficient of determination in comparison with the two compartment model due to the inclusion of the third intermediate-C pool. To predict the field CO₂ emissions, temperature and moisture factors were included in the three compartment model because laboratory incubations were carried out at constant temperature and moisture but in the field both these factors are variable. The temperature and moisture sensitive model was used to predict the CO₂ emissions measured during the summer season. The model accurately predicted the amount of C lost from No-tillage soils (observed= 2.87 Mg C ha⁻¹; predicted (modelled) = $2.80 \text{ Mg C ha}^{-1}$) but the predicted amount for rotary tilled soils $(predicted (modelled) = 2.30 \text{ Mg C ha}^{-1})$ was 30 per cent less than the amount that was lost in the field (observed = 3.33 Mg C ha⁻¹). The model predicted C loss was higher for No-tillage soils than the rotary tilled soils which was contradictory to what was observed in the field. Therefore further work is required as the data obtained during this Ph.D. study was insufficient to provide or develop a model that could be used to predict CO₂ loss from

conventional and No-tillage cultivation in New Zealand soils based on 12-day laboratory incubations.

7.2 Contribution to New Zealand agriculture

New Zealand's total greenhouse gas emissions are minor from a global perspective (0.5% of global emissions) (http://www.landcareresearch.co.nz/science/greenhouse-gases). New Zealand's economy is largely agriculture-based therefore New Zealand's greenhouse gas emissions profile is unique in comparison to other developed countries as agriculture is the main contributor (35.3 Mt-CO₂-eq) of the total greenhouse gas emissions (77.0 Mt-CO₂-eq) (projections by year for 2012) (http://www.mfe.govt.nz/publications/climate/greenhouse-gas-inventory-2012). Results of the field trial conducted during this Ph.D. study suggest that Cross Slot[®] NT cultivation resulted in a combined annual conservation of ~3.0 Mg CO₂-eq ha⁻¹ (818 kg CO₂-C ha⁻¹) in comparison to rotary tillage cultivation. The carbon dioxide released from approximately 800,000 hectares which continue to be tilled annually with conventional tillage practices in New Zealand (Personal Communication C J Baker 2009, see Appendix 1) could be about 2.4 Mt-CO₂-eq per year (800,000 X 3 Mg CO₂-eq ha⁻¹). Conversion of all 800,000 hectares which continue to be tilled annually in New Zealand to Cross Slot[®] NT seeding could reduce the contribution of agriculture to the total New Zealand greenhouse gas emissions by 7.0% per year.

7.3 Future research

The research work described in this thesis suggests some areas that require further investigation.

- The laboratory and field CO₂ measurements in this study were made on the sites that were established under Cross Slot[®] No-tillage cultivation during the last 2 to 15 years owing to unavailability of the plots paired with other forms of tillage. Long-term field sites are needed to examine the effect of No-tillage seeding on soil carbon dynamics and CO₂ conservation in comparison to conventional tillage practices. Paired plots comparing the two tillage systems should be established at different sites varying in soil and climatic factors in New Zealand to provide precise scientific information regarding the benefits of No-tillage to farmers.
- The decomposition study carried out in this Ph.D. study used four soils with a narrow range of textures (sandy loam & silt loam) and used only ryegrass pasture residues. Further studies using crop residues (barley and maize) and a wider range of soil
types/texture (sand & clay) could be helpful in understanding the decomposition pattern of different crop residues in different soils types in New Zealand.

• The model developed on the basis of 12 days' laboratory incubation data could not predict the CO₂ emissions from rotary tilled soils in the field accurately. Further studies are required to examine the effect of drying-rewetting events on soil CO₂ fluxes which could be helpful in predicting the CO₂ fluxes accurately in the event of rainfall, and these studies need to be done over a range of temperatures and soil moistures to account for the microbial biomass C pool.

Appendix 1

QUANTIFYING THE HECTARES OF LAND UNDER NO-TILLAGE IN NEW ZEALAND

(a) Forestry

There were 1.7 million hectares of planted forests in NZ in 1999 (MAF statistics, 1999).

(b) Arable crops

There were 212,000 hectares of arable crops grown in New Zealand in 1999 (MAF statistics 1999). The areas of annual arable cropping in NZ vary widely year-by-year and are largely driven by price fluctuations. The 1999 figure was in fact lower than the 10-year average because it reflected relatively low crop returns at the time.

With arable crops, the soil requires tillage both before and after the crop (the latter in order to establish the next crop or return the land to pasture). The total area tilled annually in relation to arable crops is therefore $2 \times 212,000 = 424,000$ hectares.

(c) Pasture

There were 10.3 million hectares under pasture in NZ in 1999 (MAF statistics 1999). Approximately 45% (4.6 m ha) is believed to be accessible to tractors. Tractor-accessible pastures are renewed at least every 20 years. There is a growing trend towards renewing pastures more frequently but a conservative 20-year cycle is used in these comparisons.

Therefore approximately 230,000 ha (4,600,000 X 0.05) of new long-term pastures are sown annually in New Zealand.

To avoid counting the area sown to pasture twice (since much of this area is common with the postcrop area that is tilled after arable or forage crops) we have assumed that all of the arable crops will be returned to pasture. In practice, such is not the case. Some is returned to forage crops and other arable crops and some of these crops, in turn, are returned to pasture. But the simplified assumption above allows the calculations to be more transparent and has no effect on the final figure or conclusions.

Assuming that all of the second crop after an arable crop is sown to long-term pasture, the combined annual area tilled for arable crops and new long-term pastures per year in NZ is approximately 406,000 ha {424,000 –(230,000-212,000)}.

(d) Forage crops

It is estimated that approximately 240,000 ha are sown to brassica forage crops annually in NZ (seed industry estimate, 2002). It is believed that about 35% of all forage crops in NZ are non-brassica species such as annual ryegrasses, chicory, specially-sown silage crops (rather than silage harvested from surplus long-term pastures) and forage oats (total, 84,000 ha).

The total area sown annually to forage crops in NZ is therefore approximately 324,000 ha (240,000 + 84,000).

As with arable crops, two tillage events take place for every one forage crop grown (one before and one after the crop).

The total area tilled for forage crops annually in NZ is therefore approximately 648,000 ha.

THE TOTAL AREA TILLED ANNUALLY FOR AGRICULTURAL CROPS IN NEW ZEALAND (THAT HAS THE POTENTIAL TO BE CONVERTED TO N0-TILLAGE) IS APPROXIMATELY 954,000 (648,000+406,000; SAY 1 MILLION) HECTARES

HOW MUCH NO-TILLAGE IS CURRENTLY PRACTISED IN NZ?

It is believed that approximately 200,000 ha of crop and pasture establishment in New Zealand is undertaken using no-tillage methods (industry estimate). This represents 20% of annual crop and pasture establishment.

Approximately 800,000 hectares therefore continue to be tilled annually.

CONCLUSIONS

- By converting all remaining agricultural tillage in NZ to no-tillage there is potential to turn approximately 800,000 hectares that are currently emitting carbon dioxide cumulatively, into 800,000 hectares that are instead, sequestering carbon cumulatively.
- The carbon dioxide released from approximately 800,000 hectares of tillage in NZ could be about 2.4 million tonnes per year (800,000 X 3 tonnes/ha).
- Not only would carbon emissions from agricultural crops be greatly reduced under a notillage regime, the carbon balance for agricultural crop and pasture establishment (carbon dioxide emissions balanced against sequestration of soil carbon by decaying crop residues) would become positive.
- By contrast, if all remaining crop and pasture establishment is undertaken by tillage the carbon balance will remain strongly negative.

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