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CHANGES IN SOIL C DYNAMICS AND N_2O EMISSIONS UNDER MINIMUM TILLAGE CEREAL CROP PRODUCTION IN BOREAL AGROECOSYSTEMS

– IMPLICATIONS TO CLIMATE CHANGE MITIGATION

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ACADEMIC DISSERTATION IN ENVIRONMENTAL ECOLOGY

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ABSTRACT

Soils comprise more carbon (C) than any other terrestrial source and hence even a small change in the C content can be significant in regards to atmospheric CO_2 concentration. Cultivated soils have lost soil organic carbon (SOC) during the latest decades in Finland. New cereal crop management practices, like no-till (NT) and reduced tillage (RT), can affect not only SOC stocks and stabilization, but also nitrous oxide (N_2O) emissions. The aim of this study was to gain better understanding about the changes in soil C dynamics and N_2O emissions as a result of management practice changes in the boreal region, and the implications of these changes to climate change mitigation.

Changes in SOC stocks and stabilization rates under different tillage (NT, RT, CT (conventional tillage with a moldboard plow)) and straw management (straw retention, straw removal, straw burning) practices, were studied at different sites with clayey and coarse textured soil across southern Finland. This was done by soil fractionation method (wet sieving and microaggregate isolation) to elucidate the composition of different soil fractions, namely large and small macroaggregates, microaggregates, silt and clay and macroaggregate-occluded soil fractions, and where the C is stored within them. The effects of *Lumbricus terrestris* on SOC were studied using the same method. Nitrous oxide fluxes were monitored biweekly for 2 years under CT, NT and RT practices using closed chambers. Measurements of several environmental and soil parameters were taken to study the underlying factors controlling the observed changes in soil C stocks and N_2O emissions under the different management practices.

Climate change mitigation potential through the studied cereal crop management practices seems small in the humid boreal region based on the results of this study. The minimum tillage treatments did not sequester SOC at any of the study sites which had been under NT or RT for a decade and the total C stocks were lower in the 0–15 cm topsoil layer at one clayey site under RT compared to CT after implementing RT for 30 years. Higher decomposition rate in NT compared to CT and a fairly high original SOC content at the clayey sites possibly hindered C sequestration. However, the aggregate stability was enhanced in NT cropping systems compared to CT, and NT increased the amount of SOC in large macroaggregates at several sites and in microaggregates within macroaggregates in the coarse textured site. *L. terrestris* mediated the formation of soil aggregates and the increase of SOC in the topsoil but possibly enhanced the decomposition rate in the soils. Cumulative N_2O emissions were higher under NT compared to both CT and RT at the clayey sites and lower at the coarse textured site. However, the coarse textured site under NT received slightly less N fertilizer compared to CT. Increased N_2O emissions under NT on clayey soils were likely due to denser soil structure with consistently higher soil moisture content and poor aeration. Therefore, mitigating N_2O emissions requires special attention to soil structure and drainage. This study suggests that RT is a notable option to control N_2O emissions. In the future, climate change could increase precipitation and the frequency of freeze-thaw cycles in boreal agroecosystems possibly enhancing N_2O fluxes and C losses of cultivated soils which puts pressure on finding new mitigation measures.

LIST OF ORIGINAL PAPERS

This thesis is based on the following papers, which are referred to in the text by their Roman numerals:

I. Sheehy, J., Six, J., Alakukku, L., Regina, K., 2013. Fluxes of nitrous oxide in tilled and no-tilled boreal arable soils. Agriculture, Ecosystems and Environment 164, 190–199.

II. Sheehy, J., Regina, K., Alakukku, L., Six, J., 2015. Impact of no-till and reduced tillage on aggregation and aggregate-associated carbon in Northern European agroecosystems. Soil & Tillage Research 150, 107–113.

III. Singh, P., Heikkinen, J., Ketoja, E., Nuutinen, V., Palojärvi, A., Sheehy, J., Esala, M., Mitra, S., Alakukku, L., Regina, K., 2015. Tillage and crop residue management methods had minor effects on the stock and stabilization of topsoil carbon in a 30-year field experiment. Science of the Total Environment 518–519, 337–344.

IV. Sheehy, J., Kaseva, J., Nieminen, M., Six, J., Regina, K. *Lumbricus terrestris* mediated redistribution of C and N into large macroaggregate-occluded soil fractions in fine-textured no-till soils. Submitted to Biology and Fertility of Soils.

AUTHOR'S CONTRIBUTION

I. Corresponding author. JS planned and established the field trial together with KR. JS performed the field measurements, laboratory and data analysis and wrote the paper under the supervision of KR and Johan Six with a contribution from LA.

II. Corresponding author. JS planned the field study with KR and Johan Six. JS performed the laboratory and data analysis under the supervision of Johan Six and wrote the paper under the supervision of KR and Johan Six with a contribution from LA.

III. Co-author. JS supervised and helped with the laboratory and data analysis in an effort to transfer and set up the fractionation method from UC Davis to Natural Resources Institute in Jokioinen. JS wrote a significant part of the paper together with the corresponding author PS with a contribution from JH, EK, VN, AP, ME, MS and LA under the supervision of KR.

IV. Corresponding author. JS planned the field study under the supervision of KR with a contribution from MN. JS performed the field measurements with a contribution from MN and conducted the laboratory and data analysis under the supervision of Johan Six. JS performed the statistical analyses with a contribution from JK. JS wrote the paper under the supervision of KR and Johan Six.

ABBREVIATIONS

C	Carbon
CH_4	Methane
CO_2	Carbon dioxide
cPOM	Coarse particulate organic matter
CT	Conventional tillage
GHG	Greenhouse gas
IPCC	Intergovernmental Panel on Climate Change
LM	Large macroaggregate
m	Microaggregate
mM	Microaggregates within macroaggregates
MWD	Mean weight diameter
N	Nitrogen
NH_4^+	Ammonium
N_2O	Nitrous oxide
NO	Nitric oxide
NO_2^-	Nitrite
NO_3^-	Nitrate
NT	No-till
OM	Organic matter
RT	Reduced tillage
sM	Small macroaggregate
SOC	Soil organic carbon
SOM	Soil organic matter
s+c	Silt and clay
s+cM	Silt and clay within macroaggregates

1. INTRODUCTION

1.1 AGRICULTURE AND CLIMATE CHANGE

Globally averaged combined land and ocean surface temperature has risen 0.85°C since 1880 (IPCC, 2014) while the Arctic has warmed twice as fast as the rest of the world during the 20th century (ACIA, 2005). Due to its northern location, Finland is exposed to this polar amplification of climate change-induced warming and has experienced a mean temperature increase of over 2°C within the last 166 years (1847–2013) (Mikkonen *et al.*, 2014). Agriculture contributes to global anthropogenic emissions of three major greenhouse gases (GHGs); carbon dioxide (CO_2), nitrous oxide (N_2O) and methane (CH_4), accounting for about 10–12% of the total emissions of GHGs. The atmospheric concentration of all three gases has risen sharply during the past decades (Fig 1, Data retrieved from: http://www.eea.europa.eu/). In Finland, without taking into account the energy use or land use and land use changes, agriculture contributes 9% of the GHGs, of which CO_2 plays a minor role, emitted into the atmosphere (Statistics Finland, 2014).

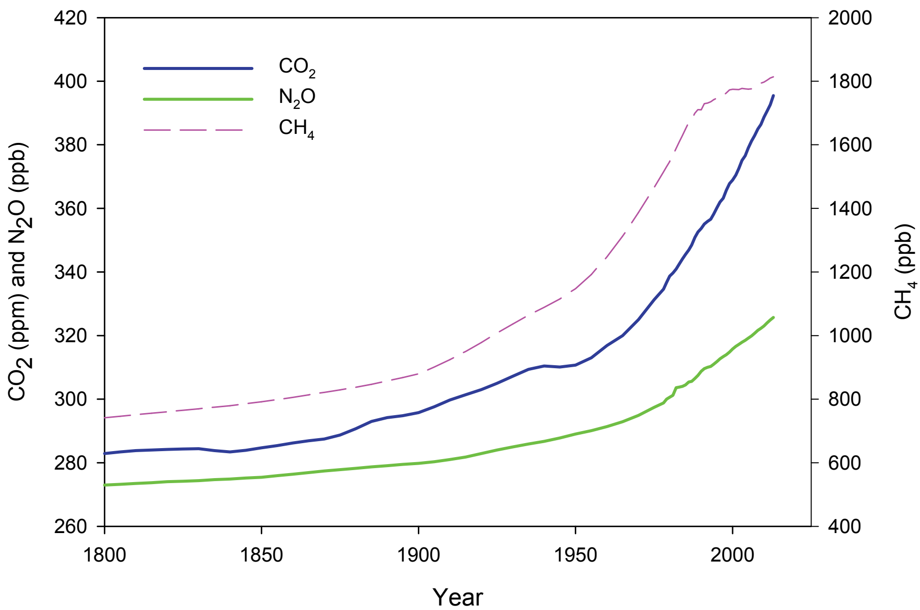

Figure 1. Atmospheric concentrations of CO_2 (ppm), N_2O (ppb) (left axis) and CH_4 (ppb) (right axis) between 1800 and 2013 (Data retrieved from: http://www.eea.europa.eu/).

The European Council confirmed in early 2011 the new climate objective of an 80–95% reduction in GHG emissions compared to the level of 1990 by midcentury (Watson *et al.*, 2013). To achieve the climate goal of the European Union, all aspects of the economy must be taken into account. This includes the non-emission trading sectors that were assigned a reduction target of 16% in 2005–2020 as a part of the Kyoto Protocol (EU, 2009). In Finland this has translated to a 13% reduction for agriculture (Ministry of Employment and Economy, 2008). These decisions cover agricultural emissions only partly; e.g. CO_2 emissions from soils are not included. Agricultural land use related CO_2 emissions will gain more and more attention in the EU in 2015-2020 as the member states are committed to report actions in cropland and grazing land management (EU, 2013).

Agricultural land use inevitably leads to emissions of CO_2 into the atmosphere and a concurrent decrease in soil organic carbon (SOC) (Lal, 1997). This is mainly due to the initial land use change which escalates soil respiration while decreasing carbon (C) inputs into the soil (Carter *et al.*, 1998; Gregorich *et al.*, 2005; Elder and Lal, 2008). The loss of SOC as a result of agricultural use compared to natural state is typically 30–40% in northern regions (Ellert and Gregorich, 1996; Carter *et al.*, 1998; Grünzweig *et al.*, 2004). According to IPCC guidelines, the system is considered not to experience conversion effects after the initial land-use change and instead enters a “steady-state” (IPCC, 2006). In this state, cropland management is the key modifier of soil C stocks depending on how these practices affect C input versus output from the system (Paustian *et al.*, 1997; Ogle *et al.*, 2005). In addition, the changes in the climate system will strongly influence the whole terrestrial C cycling and land-atmosphere CO_2 fluxes (Fischlin *et al.*, 2007; Frank *et al.*, 2015).

Agricultural N_2O emissions have increased by 17% between 1990 and 2005 (Smith *et al.,* 2007). Between 40–50% of global N_2O emissions originate from human activities with most of the increase being attributed to the expansion of agricultural land area and enhanced microbial N_2O production. This effect is associated with human perturbations to the nitrogen (N) cycle like over application of N fertilizers and manure to grow more food (Nevison, 2000, Reay *et al.,* 2012). In Finland, this problem is enhanced by the large amount of cultivated organic soils which add up to 12% of the cultivated land area but contribute to approximately 30% of the N_2O emissions of Finnish agricultural lands (Lapveteläinen *et al.*, 2007). Nitrous oxide emissions are reported as part of national inventories of GHGs in many countries around the world (Lokupitiya and Paustian, 2006), and their mitigation has a growing role in global and local climate policies.

In 2012, agricultural area occupied about 5000 Mha of land which is 38% of the land on Earth (FAOSTAT: http://faostat3.fao.org/download/R/RL/E, data retrieved 15.03.2015). Of this land 70% was under pasture and 30% was under arable production. Driven by a growing population agricultural land area increased by over 10% in just four decades, even though technological improvements have increased land productivity and per capita food availability (Smith *et al.*, 2007). The need to find ways of reducing GHG emissions from arable soils, while maintaining sustainable agricultural production and meeting the needs of a growing population, is evident.

1.1.1 CARBON CYCLE AND SOIL ORGANIC CARBON

Soil organic carbon has implications for global C cycle since soils are the second largest reservoir of C after oceans (Stockmann *et al.*, 2013). Just the first meter of soils store globally about 1500 Gt of C whereas vegetation stores about 600 Gt (Adams et al., 1990, Eswaran *et al.*, 1993, Batjes, 1996; Schlesinger, 1997). Hence, even a small change in the SOC dynamics can greatly affect atmospheric CO_2 concentration (Jenkinson *et al.*, 1991; Cox *et al.*, 2000).

The net amount of CO_2 released to the atmosphere from agricultural soils is dependent on the rate of SOC formation versus decomposition (Van Breemen and Feijtel, 1990). Due to large fluxes via plant uptake, the net flux of CO_2 from agricultural lands is estimated to be fairly small (~0.04 Gt CO_2 yr^{-1}). However, C depletion after the initial push is sustained by erosion, crop removal and biomass burning (Gregorich *et al.*, 2005; Lal, 2007). A 35-year study conducted in Finland found that cultivated soils have in general been losing SOC during the past decades (Heikkinen *et al.*, 2013). Thus, soil C sequestration is one of the options for mitigating GHGs from agricultural soils. Enhanced crop and grazing land management which include, for example, improved tillage practices, straw management and nutrient use, together with land restoration are the most prominent techniques to increase C sequestration into agricultural soils (Armentano and Menges, 1986; Smith *et al.*, 2007). In Finland, crop cultivation on organic soils is also a significant source of CO_2 among the emissions reported in the land use, land use change and forestry sector (LULUCF) (Statistics Finland, 2014).

Atmospheric CO_2 is converted to carbohydrates via plant photosynthesis that produces plant biomass, in other words, organic matter (OM). Some of this C is immediately released back into the atmosphere by plant and soil respiration and some is lost from the ecosystem through crop harvesting. Remaining organic C is retained on the soils as residues like straw, roots and casts (Janzen *et al.*, 1998). Carbon enters into the soil through decomposition of residues and processing of soil organic matter (SOM) by soil fauna and micro-organisms (Stevenson 1994). The quantity and quality of residues affect the potential C sequestration rate depending on the proportion of OM being humified or stabilized in the soil (Kätterer, 2011). Rasse *et al.* (2005) argued that most of the soil C is composed of root C compared to shoot derived C and, that the effect of roots increases with depth. This is especially true in cropping systems where little aboveground residues are left on the soil.

Residues with a lignin content of less than 15% and N content of more than 2.5% are considered high quality residues, and they enhance residue decomposition and the release of nutrients compared to low quality residues (Palm *et al.*, 2001).

Environmental factors, like soil temperature, moisture content and pH, along with soil organisms also play a critical role in determining the decomposition rate of OM. Soil respiration is the main source of CO_2 released into the atmosphere (Raich and Schlesinger, 1992), and it consists of plant derived respiration by plant roots, rhizo-microbial respiration and microbial respiration of recently died plant residues, as well as SOM-derived respiration (Kuzyakov, 2006). Soil respiration normally increases exponentially with increasing temperature (Xu and Qi, 2001).

Soil organic matter can be stabilized in the soil through three main routes as described by Six *et al.* (2002) (Fig 2). First of the stabilizing mechanisms is chemical stabilization, where SOM and soil minerals such as clay and silt particles bind together. Secondly, SOM is stabilized through physical protection by soil aggregates and thirdly by biochemical stabilization that is driven by the chemical composition of SOM itself and chemical complexing processes (Six *et al.*, 2002). Soil aggregates are formed by fresh organic residues binding with soil mineral particles. These soil aggregates bind more strongly to each other compared to other surrounding soil particles and they increase the residence time of C in soils by both offering physical protection from microbial decomposition and by creating more anaerobic conditions in the soil that slow down the decay of SOM (Six *et al.*, 2002). Especially microaggregates within macroaggregates (mM) are a crucial part of long-term sequestration and storing of SOC (Six *et al.*, 2000; Denef *et al.*, 2007). While the potential for soils to store C depends on the soil texture and the quality of residue input, the soil C concentration generally increases with added C inputs with C being stored in different parts of the soil until the soil reaches its saturation level. The changes in the amount of C in soil are so slow that it is very difficult to observe only by measuring the total amount of C in soil. This is why it is important to know in which soil fractions SOC is stored.

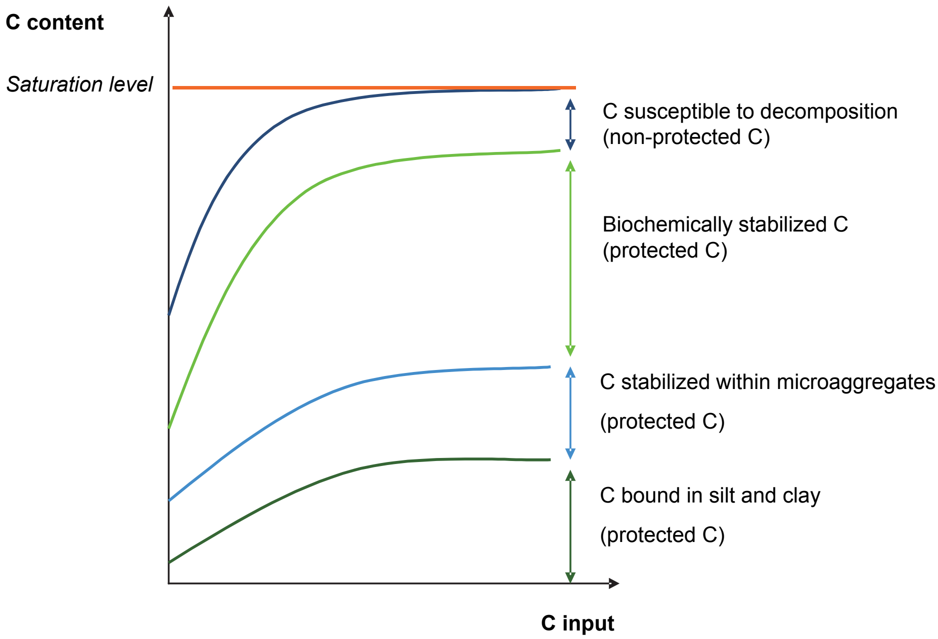

Figure 2. Soil saturation level is defined by a combination of different C pools: 1) Silt and clay protected C, 2) microaggregate protected C, 3) biochemically protected C and the, 4) unprotected C pool. Adapted after Six *et al.*, (2002).

Earthworms are known to influence soil processes including soil aggregation, decomposition of residues and soil structure modification, which has created them a reputation as ecosystem engineers (Jones *et al.*, 1994; Lavelle *et al.*, 1997; Fonte *et al.*, 2009; Giannopoulos *et al.*, 2010). Earthworms represent the largest group of animals in the soil by biomass and they can consume up to 2 t of litter ha^{-1} yr^{-1} (Whalen and Parmelee, 2000). They are categorized in three different groups, epigeic, endogeic and anecic, based on their feeding habits and the soil environment they occupy (Bouché, 1977; Lavelle and Spain, 2001) (Fig 3). The earthworm studied in this study, *Lumbricus terrestris* L., is an anecic earthworm that makes permanent, typically less than 1 m deep, vertical burrows, which open at the soil surface. These species feed on the fresh surface litter, which they pull down into their burrows and leave litter and cast-made middens within sight on the soil surface (Lee, 1985).

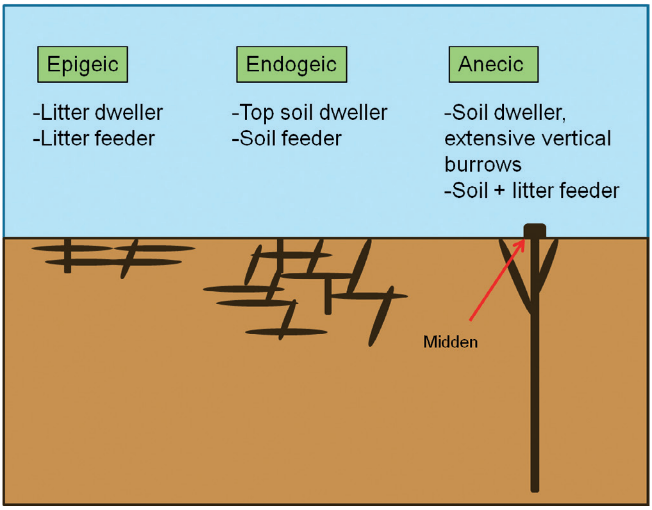

Figure 3. Earthworm categories based on their feeding habits and the soil environment they occupy.

Soil C dynamics are affected by earthworms, including *L. terrestris,* due to the efficient residue burial and consumption (Subler and Kirsch, 1998; Eisenhauer *et al.*, 2008), and defecation of casts which result from intimate mixing of ingested chopped organic residues, that are often high in C due to preferential selection, and soil particles in earthworm's digestive tract (Barois *et al.*, 1993; Shipitalo and Le Bayon, 2004). The casts are significant for C stabilization because of the relative stability of dried and aged casts and the formation of new macroaggregate-occluded microaggregates within the earthworm gut (Bossuyt *et al.*, 2004; Six *et al.*, 2004; Pulleman *et al.*, 2005; Fonte *et al.*, 2007; Fonte *et al.*, 2009; Six and Paustian, 2014). Casts can be deposited, for example, in *L. terrestris* middens on the soil surface and in burrow walls where they can slightly increase SOC content of the burrow lining (Don *et al.*, 2008). Fonte *et al.* (2007) observed an increase in newly added residue derived C into macroaggregate-occluded microaggregates even in a low input agroecosystem while Bossuyt et al. (2004) showed a link between C depletion in microaggregates within macroaggregates in a secondary forest due to a loss in earthworm abundance. These support the theory of the biological origin of this soil fraction and highlight the importance of soil management via soil biota (Six and Paustian, 2014).

1.1.2 NITROUS OXIDE FLUXES AND SOIL NITROGEN

In natural conditions N enters the soil though biological N fixation. This is carried out by micro-organisms which live both in symbiotic relationships with plants and free in the soil. Nitrogen is often the limiting nutrient in soils which is why in agroecosystems plants are amended with N fertilizers either in mineral or organic form. Organic N, that is bound to OM i.e. crop residues, re-enters the cycle through mineralization by soil micro-organisms (Canfield *et al.*, 2010). In the mineralization process organic N is transformed into ammonium (NH_4^+). Immobilization is an opposite process where inorganic N is being used and bound to plant or microbial biomass (Jansson and Persson, 1982).

The main microbial processes (65%) contributing to N_2O emissions are anaerobic denitrification, and aerobic nitrification that produces N_2O as a side product when oxygen (O_2) is limited in the soil (Doran, 1980; Groffman, 1984; Six *et al.*, 2002, Smith *et al.*, 2003). Denitrification is thought to be the most significant one in agroecosystems contributing to N_2O emissions (Ball *et al.*, 1999; Smith and Conen, 2004; Monteny *et al.*, 2006). It is a reduction process where nitrate (NO_3^-) is reduced to nitrite (NO_2^-) and further to N_2O and finally N_2 (equation 1, Smith *et al.*, 2003). This process can be extremely quick since after soil O_2 is consumed by roots and microbes, NO_3^- and NO_2^- are the best electron receivers in the soil. However, denitrification can also occur when O_2 is present (Smith *et al.*, 2003).

$$NO_3^- \rightarrow NO_2^- \rightarrow NO + N_2O \rightarrow N_2 \qquad (1)$$

In nitrification, NH_4^+ or nitrogen from organic nitrogen compounds, is transformed in an aerobic process first into NO_2^- and further oxidized into NO_3^- as shown in equation 2 (Canfield *et al.*, 2010). If the soil O_2 levels drop the ammonia oxidizing soil bacteria can use NO_2^- as an electron receiver and reduce it to nitric oxide (NO) and N_2O. This is called denitrification of nitrifying bacteria (Smith *et al.*, 2003).

$$NH_4^+ \rightarrow NH_2OH \rightarrow NO_2^- \ (N_2O) \rightarrow NO_3^- \qquad (2)$$

Denitrifiers consist of more than 60 known genera of Bacteria and Archaea as well as some fungi, protozoa and benthic organisms (Demanèche *et al.*, 2009). In addition to soil nitrate and O_2 level, soil pH, temperature, moisture content and the quantity and quality of SOM, all control the denitrification process (Gregorich *et al.*, 2005, Johnson *et al.*, 2007). Due to this high spatial and temporal variability of these emissions, it is hard to calculate accurate estimates in a national, or even on a field, scale. In addition, the boreal climatic conditions add complexity to the calculations with frequent freeze-thaw cycles and a short growing season (Regina *et al.*, 2013).

Currently, on average, only about 50% of added N gets utilized by the growing plants in agroecosystems while the other half is vulnerable to be leached out of the root zone or emitted to the atmosphere as N2O and N2 (Erisman *et al.*, 2007). Nutrient use efficiency is even lower, typically below 40%, for wheat, rice and maize which currently use about 50% of the globally added fertilizer (Ladha *et al.*, 2005; Canfield *et al.*, 2010). In Finland a meta-analysis found an average N uptake of 36% for wheat and barley and 43% for oat (Valkama *et al.*, 2013). Therefore, N-efficient agriculture that produces proper yields with minimum nutrient losses is considered one of the best ways to mitigate high N2O fluxes (Snyder *et al.*, 2009).

Earthworm feeding enhances litter decomposition through increasing the surface area of SOM (Ingham *et al.,* 1985). Smaller organic compounds and mineral nutrients are then available for other soil microorganisms or plants to use. For example, James (1991) showed that 10–12% of the yearly plant N uptake in a tallgrass prairie was present in earthworm casts produced in one year. Similarly, Cortez *et al.* (2000) reported a significant increase in the quantity of inorganic N in the soil in the presence of earthworms. This indicates that, in addition to other soil fauna, earthworms have an essential role in supplying available N to the plants, and thus facilitate crop production in agroecosystems (Lubbers *et al.*, 2011). Earthworms can modify N cycle also in other ways, for example, offering ideal conditions for microbial nitrification and denitrification to occur within their guts, casts and burrow walls (Drake and Horn, 2007; Costello and Lamberti, 2008) potentially leading to increased N_2O emissions from soils as a negative side effect of enhanced N mineralization (Lubbers *et al.*, 2013).

1.2 CLIMATIC EFFECTS OF MANAGEMENT PRACTICES IN CEREAL CROP PRODUCTION

1.2.1 MINIMUM TILLAGE PRACTICES

Minimum tillage practices, which include both reduced tillage (RT) and no-till (NT), are being increasingly adopted in order to reduce labor costs, energy use and to preserve water and soil C as compared to conventional tillage (CT), the practice with annual topsoil turnaround (Cole *et al.*, 1997). In NT agriculture the crop is sown into the ground without any prior tillage and RT (shallow stubble cultivation) is a management practice between CT and NT disturbing the soil to a shallower depth compared to CT. At a global scale, the adoption of NT has increased more than 230% in the latest decade, reaching 111 million ha in 2009 (Derpsch *et al.*, 2010). This represents approximately 8% of total arable land in the world. In Europe, the adoption of NT has been clearly slower than, for instance, in North and South America (Soane *et al.*, 2012). In Finland, the area of agricultural land converted to NT, has, however, increased rapidly and is estimated to have reached over 150,000 ha in only a little over ten years after it was introduced to farmers (Tike, 2011). About 13% of the annually sown area is now under NT and 25% under RT.

Minimum tillage practices increase C sequestration in most parts of the world by increasing soil aggregation and decreasing mineralization (Abdalla *et al.*, 2013). When the disturbance of the soil decreases and the soil fauna changes after implementing NT practice, the amount of C-rich macroaggregates begins to increase (Six *et al.*, 2004). Especially microaggregates within macroaggregates (mM), a soil fraction between 53-250 µm, can store a substantial amount of SOC, which is well protected against physical disturbance, and can represent the majority of the C sequestration potential observed in minimum tillage (Denef *et al.*, 2004; Six and Paustian, 2014). Denef *et al.* (2004) found that over 90% of the difference in SOC content between CT and NT management practices was associated with the mM soil fraction. Increased aggregate stability as a result of NT management practice has been found especially in the tropics (Lal *et al.*, 1999; Bayer *et al.*, 2006; Maia *et al.*, 2010) and temperate regions of the world (Lal *et al.*, 1997). According to Freibauer *et al.* (2004) the potential for management practices to sequester SOC in European (EU15) agricultural soils, that on average are currently depleted of soil C, is 0.3–0.4 ± 0.1 Mg C ha^{-1} yr^{-1} and < 0.4 Mg C ha^{-1} yr^{-1}, for NT and RT respectively, which are in the same range as the estimate of 0.2–0.4 Mg C ha^{-1} yr^{-1} for conservation tillage practices reported by Watson *et al.* (2000) for Australia, USA and Canada.

Conventional tillage, in which the soil is usually plowed to a depth of 20 cm, is considered a practice that enhances losses of SOC (Martins *et al.*, 1991; Puget and Lal, 2005; Wright and Hons, 2005). The turnover rate of SOM increases under intensive tillage (Six *et al.*, 2000), soil aggregation slows down and decomposition rate of OM increases (Dolan *et al.*, 2006). Tillage induces several factors affecting these processes including direct physical disturbance due to plowing and indirect effects through soil exposure to wet-dry and freeze-thaw cycles at soil surface (Paustian *et al.*, 1997a), changes in soil microclimate and microbial community as well as litter placement (Holland and Coleman, 1987). However, recent studies have challenged the view of minimum tillage practices increasing soil C sequestration for cold, humid regions of the world and shown that this may be true only in relatively arid climates (Vandenbygaart *et al.*, 2003). Also, CT practice may not decrease the SOC stocks as quickly in cool and humid climatic regions (Gregorich *et al.*, 2005; Hermle *et al.*, 2008).

N_2O is a greenhouse gas approximately 300 times more potent than CO_2, and it has been estimated that its increase under NT could offset 75–310% of the advantage gained from C sequestration under NT (Li *et al.*, 2005). It was estimated that if all the European agricultural soils that could be converted to NT adopted this management practice, N_2O emissions would increase by the equivalent of 20.5 Tg of C emitted per year in comparison to CT (Smith *et al.*, 2001). This is due to NT soils often exhibiting a more dense soil structure (Tebrügge and Düring, 1999; Schjønning and Rasmussen, 2000) and higher moisture content (Sharratt, 1996; Gregorich *et al.*, 2008), which favor the activity of anaerobic denitrifying bacteria.

Field measurements have shown great variability in N_2O emissions under NT or RT compared to CT depending on different soil types and climatic conditions (Aulakh *et al.*, 1984; MacKenzie *et al.*, 1998; Ball *et al.*, 1999; Elmi *et al.*, 2003; Kaharabata *et al.*, 2003; Chatskikh and Olesen, 2007; Beheydt *et al.*, 2008; Chatskikh *et al.*, 2008; Ussiri *et al.*, 2009; Abdalla *et al.*, 2010). The different duration of the experiments may partly explain the contradicting observations; Six *et al.*, (2004) reported results of a meta-analysis indicating increased N_2O emissions during the first years after converting to NT, with emissions reducing back to normal levels or less in humid climate after 20 years of NT. Rochette (2008), on the other hand, argued that N_2O fluxes only increase from poorly aerated soils under NT, especially in cool, humid climates. Grandy *et al.* (2006) found that N_2O fluxes did not increase under NT in a 10-year study and only offset 56–61% of the C sequestered on loamy soils in south-western Michigan. However, data on the long-term effects of different management practices on emissions of N_2O in boreal climate are scarce.

Effects of earthworms are especially interesting in long-term minimum tillage settings where more organic residues are available on the soil surface or within the top soil layer. Conversion from CT to NT or RT have been shown to increase earthworm diversity and abundance (Clapperton *et al.*, 1997; Chan, 2001), including *L. terrestris*, that benefits when its vertical home burrows are not disturbed and residues remain available (Kladivko, 2001). This is also true in Finland where NT increased both the earthworm biomass and number of individuals under NT compared to CT (Visa Nuutinen, unpublished results).

1.2.2 STRAW MANAGEMENT

All of the observed differences in SOC content and C sequestration potential in agroecosystems cannot be directly earmarked to tillage effects. Residue stratification within the soil profile, soil compaction, yield and erosion levels, for example, can add to the divergence between SOC levels (Yang *et al.*, 2013). As SOC content largely depends on the C inputs and decomposition rates of residues in the soil, straw management practices can play a key role in determining the C sequestration rates of a particular field. Straw retention in crop fields has been shown to increase soil aggregation and build-up of SOC depending on the quality and quantity of the residue application (Chivenge *et al.*, 2007) while straw removal can decrease SOC compared to straw retention (Rasmussen and Parton, 1994; Kätterer and Andrén, 1999; Mann *et al.*, 2002). However, the magnitude of the potential C sequestration following straw retention depends on climatic and soil conditions, like temperature and soil moisture content (Curtin and Fraser, 2003; Powlson *et al.*, 2011).

In Europe, burning of harvest residues used to be common practice but is now banned in most countries. Combustion for energy or partial combustion to biochar may become more common in the future, possibly affecting straw retention rates on crop fields (Funke *et al.*, 2013). In the tropics, residue burning is still a common practice to enhance nutrient mineralization. However this practice is known to eventually lead to nutrient loss and reduced soil fertility (Hemwong *et al.*, 2008) in addition to causing a significant amount of air pollution. It can also be harmful to beneficial soil fauna and microorganisms (Wuest *et al.*, 2005; Jain *et al.*, 2014).

1.3 OBJECTIVES OF THE STUDY

In Finland, cultivated soils have been evaluated to lose SOC during the last decades (Heikkinen *et al.*, 2013) while new cereal crop management practices, like NT and RT, have started to gain in popularity (Tike, 2011) and climate change has already increased the average temperature and precipitation in the country (Tietäväinen *et al.*, 2010). These are huge landscape scale changes affecting Finnish food production in a fundamental way. Monitoring of SOC levels and N_2O fluxes across landscape units and over time is crucial for the accurate assessment of spatial and temporal variations in SOC pools and N_2O fluxes (Hartemink *et al.*, 2014). The aim of this thesis was to gain better understanding about the changes in soil C dynamics and N_2O emissions as affected by tillage practice in the boreal region and the climatic implications of the different management practices. The changes under NT and RT compared to CT are partly mediated through soil fauna and residue placement effects. Hence this project studied the effects of an anecic earthworm, *L. terrestris*, and straw management on C sequestration potential and SOC stock. Measurements of several environmental and soil parameters were taken to elucidate the underlying factors controlling the observed changes under the different management practices.

The objectives of the study were:

1) To measure the SOC stocks under NT, RT and CT and more specifically to study the composition of soil aggregates and the distribution and amount of SOC within the studied soil fractions in boreal agroecosystems (II, III).
2) To quantify the annual cumulative N_2O flux under NT, RT and CT in different soil types and study the short-term effect of soil plowing on the temporal N_2O fluxes (I).

3) To link changes in the soil environment (i.e. soil moisture and temperature) to the observed differences in N_2O emissions under the different management practices (I).
4) To determine the effects of straw management practices and *L. terrestris* to C sequestration potential (III, IV).
5) To assess whether NT and RT are notable options compared to CT to mitigate climatic effects of cereal crop production in boreal, humid climatic conditions (I, II, III, IV).

The main hypotheses of the study were:

1) Due to the humid, cold climate NT and RT management practices would not sequester C in the topsoil (II, III, IV).
2) NT and RT would increase soil aggregation level (II, III).
3) N_2O emissions would be higher under NT and RT compared to CT (I).
4) *L. terrestris* density would be increased in RT and areas of *L. terrestris* middens would have a higher soil aggregation level, more macroaggregate-occluded microaggregates and associated SOC; thus adding to C sequestration potential (III, IV).
5) Straw retention would increase total SOC and soil aggregation level (III).

2. MATERIAL AND METHODS

2.1 STUDY SITE FEATURES AND ENVIRONMENT

This study took place at five different locations around the most heavily used agricultural area in Finland. Sites 1, 2 and 5 were located in Jokioinen (60°49'N and 23°30'E) in southern Finland and maintained by Natural Resources Institute Finland. Site 5 was a long-term experimental field established in 1983 (III). Site 3 was located in Vihti (60°21'N and 24°22'E) and one field pair (site 4) in Säkylä (60°58'N and 22°31'E) in southwestern Finland (I, II and IV) (Fig 4). Sites 1–2 were field experiments with a randomized complete-block design (plot size 250 m^2 and 240 m^2 respectively) with four replicates of each soil tillage treatment (CT, NT and RT) (I, II, IV), site 3 was a field experiment with a randomized complete-block design (plot size 75 m^2) with four replicates of CT and NT treatment (I, II), and site 5 a field experiment with a randomized complete split-plot block design (plot size 60 m^2) where the main-plot treatments (four replicates) consisted of three straw management practices; straw retained, straw removed and straw burnt, and the split-plots had two soil tillage treatments (CT and RT) (III). The fields of site 4 belonged to neighboring farmers (four replicated measurement sites/ field, plot size 100–250 m^2) and consisted of CT and NT treatments (I, II, IV). Different sites had been under the different treatments for 8 (sites 1 and 3), 9 (site 2), 10 (site 4) or 30 (site 5) years. Soils at sites 1–2 and 5 were classified as Vertic Luvic Stagnosol (FAO, 2014), at site 3 as Vertic Cambisol and at site 4 as Eutric Regosol (FAO, 2006). Mean annual precipitation ranged from 614 (site 4) to 647 mm (site 3) and mean annual temperature from 4.6 (sites 1, 2, 3 and 5) to 4.8°C (site 4) (Reference period 1981–2010, Pirinen *et al.*, 2012).

Every autumn (September/October), the soil was mouldboard ploughed to a depth of 20–25 cm on all CT plots and soil was stubble cultivated with tined cultivator to 10–15 cm on RT plots. In spring, CT and RT plots were first leveled by a harrow and then rotary or S-tine harrowed to 4–5 cm depth to prepare the seedbed. Spring barley (*Hordeum vulgare*) was cultivated at sites 1 and 2 during all study years. At site 3, spring oilseed rape (*Brassica rapa* subsp. *oleifera*) was cultivated during the 2008 growing season, spring wheat (*Triticum aestivum*) in 2009 and spring oat (*Avena sativa*) in 2010. Spring barley and spring oilseed rape were cultivated at site 4 during the experiment years. Different spring or winter cereals were cultivated at site 5 since 1983. All treatments at all sites were combisown (drilling and fertilization in the same time) in May. The whole annual fertilizer application (between 70–110 kg of N ha^{-1} yr^{-1}) was placed during sowing. All treatments at sites 1 and 2 received 110 kg of N ha^{-1} yr^{-1} in 2008 and 100 kg of

N ha^{-1} yr^{-1} in 2009 and 2010. Treatments of site 3 received 105 kg of N ha^{-1} yr^{-1} in 2008 and 2009 and 120 kg of N ha^{-1} yr^{-1} in 2010. Site 5 received 90–110 kg ha^{-1} N annually during the 30 year experiment. Granular ammonium nitrate NPK fertilizer was used, except at site 4 where liquid fertilizer (Urea 32, 70-80 kg of N ha^{-1} yr^{-1}) was used in the NT plots and the fertilizing level was lower compared to CT (83-96 kg of N ha^{-1} yr^{-1}) according to farmers experience on the need of N fertilizer on this plot. All sites were harvested in August. At site 5, depending on the straw treatment, chopped harvest residues were left on the field, removed or burned before tillage. Yield information was gathered from all study sites.

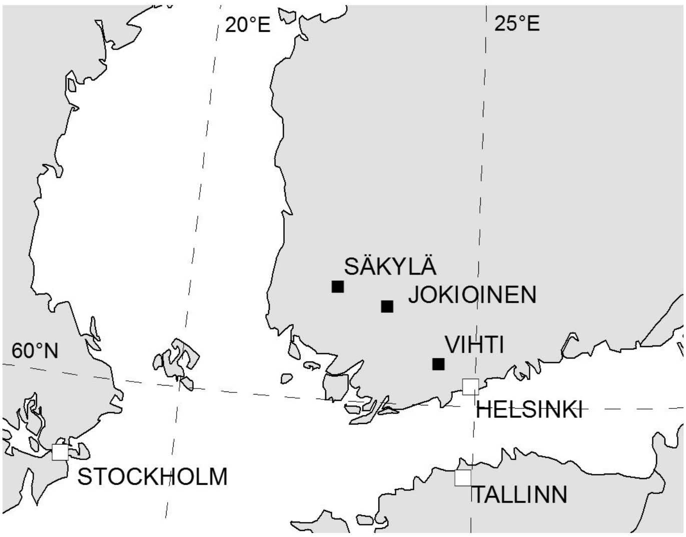

Figure 4. Locations of the different study sites across southern Finland. Study sites 1-2 and 5 were in Jokioinen, site 3 in Vihti and site 4 in Säkylä.

2.2 SOIL PARAMETERS AND ANALYSIS

2.2.1 BACKGROUND VARIABLES AND SOIL SAMPLE COLLECTION

A wide range of key background variables that were measured regarding the different studies are shown in table 1. Daily air temperatures and precipitation were measured at study sites 1–4 (I: Fig 1). In addition soil chemical composition was measured at site 5 (III: Table 1).

Table 1. Measured additional soil and environmental parameters by study site are referred with roman numerals indicating the original paper they are published in.

	Site 1	Site 2	Site 3	Site 4	Site 5
Soil temperature	I	I	I	I	-
Water-filled pore space (WFPS)	I	I	-	I	-
Mineral N content	I	I	I	I	III
Potential denitrification	I	I	I	I	-
Depth of frost	I	I	-	I	-
Soil bulk density	I, II, IV	I, II, IV	I, II	I, II, IV	III
Bulk SOC	I, II, IV	I, II, IV	I, II	I, II, IV	III
Total N	I	I	I	I	-
Earthworm burrows	I	I	I	I	-
L. terrestris midden density	-	-	-	-	III
Microbial C	-	-	-	-	III

Samples for physical soil fractionation from CT, NT and RT treatments were taken in October 2009 after harvest but before tillage operations at study sites 1–4 from a 0–20 cm soil layer (II). The soil samples were taken with a 3 cm auger. Ten randomly taken samples from each plot were pooled together. Samples were passed through an 8 mm sieve before they were air dried and stored. Soil samples from site 5 to study the effect of different long-term straw and tillage management practices were collected in early June 2013 from five soil layers: 0–5, 5–10, 10–15, 15–20 and 20–40 cm as ~20 soil cores pooled to one sample per layer per plot. Samples were stored air dried except the samples for microbial biomass and mineral N determination which were stored at -18°C. Soil physical fractionation was done for the soil layers 0–5 cm and 15–20 cm. In addition SOC was determined in the 20–40 cm layer. Soil bulk density in the layers 0–5, 5–10, 10–15 and 15–17.5 cm was determined using the Kopec corer with a diameter of 5 cm.

Visibly clear *L. terrestris* middens were sampled from the top of the study fields at sites 1, 2 and 4 in September 2010 about a month after harvest to study its effects on soil aggregation and SOC content (IV). Out of the 16 earthworm species that exist in Finland, *L. terrestris* is the only anecic earthworm species (Terhivuo, 1988). Thus, its middens are easily identifiable. Also, at the time of the field work the conditions for observations were favorable because in all study plots the soil surface had remained intact and the straw residue had been left in all plots. At the sites of high *L. terrestris* density majority of the straw had been gathered by the worms which made the middens distinct and easily observable. The soil samples were taken from earthworm middens and surrounding bulk soil to a depth of 5 cm with a 5 cm diameter soil corer. The midden soil samples included the burrow entrance of *L. terrestris* and were comprised mostly of soil, casts and straw. The bulk soil samples represent the soil without *L. terrestris* midden-burrow-complexes and were taken at least 15 cm away from any visible middens. Since the earthworm-created structures can last for decades and can therefore be considered fairly stable (Hagedorn and Bundt, 2002), we assumed bulk soil to show less earthworm related effects on SOC in comparison to midden soil with high present earthworm activity. In total, 16 midden soil –bulk soil pairs were sampled per treatment (midden vs. bulk) and per site. Four midden soil samples were combined in each location and the same was done for the bulk soil samples. The final amount of repetitions was four samples per treatment per study site. Soil samples for the determination of soil bulk density were taken from the 0–5 soil layer once in 2010.

2.2.2 SOIL FRACTIONATION METHOD

The aggregate size distribution was analyzed by separating different soil fractions in two stages, wet sieving and microaggregate isolation, following a protocol developed by Six *et al.* (2000) based on a wet sieving method by Elliott *et al.* (1986) (Fig 5) . The field moist soil samples were sieved carefully without breaking the soil structure through an 8 mm sieve and then air-dried. A subsample of 80 g was taken for the wet sieving process which was done through a series of three sieves that separated the samples into four different soil fractions; large macroaggregates (LM; >2000 μm), small macroaggregates (sM; 250–2000 μm), microaggregates (m; 53–250 μm) and silt and clay (s+c; <53 μm). Prior to wet sieving the samples were submerged into deionized water on top of the 2000 μm sieve for a period of 5 min to enable slaking which breaks down the unstable aggregates with pressure buildup. The sieving was done by manually moving the sieve up and down 50 times during a 2 min period. The sieve was backwashed and the fraction remaining on top of the sieve was collected in an aluminum pan and oven dried in 60°C. Organic material floating on the water after sieving with the 2000 μm sieve was discarded as it is not considered SOM by definition. The sieving was repeated with the remaining sieves by pouring the water and soil from the earlier sieving through the next one.

Soil fractions within LM and sM fraction (or combined M) were further isolated with a method described in Six *et al.* (2000) (II, III, IV). The goal of this method was to break down the macroaggregates while avoiding the breakdown of the released microaggregates. The subsample was put into deionized water on top of a 250 μm mesh and shaken in a reciprocal shaker with a continuous flow of running water with 50 stainless steel beads (4 mm diameter) until all the macroaggregates were broken down (2–3 min of shaking depending on soil type). The microaggregates and other released material went through the mesh screen with the running water ending up on a 53 µm sieve that was sieved as in the wet-sieving method. As a result three different fractions were separated: coarse particulate organic matter (cPOM; >250 µm), microaggregates within macroaggregates (mM; 53–250 µm) and silt and clay (s+cM; <53 µm).

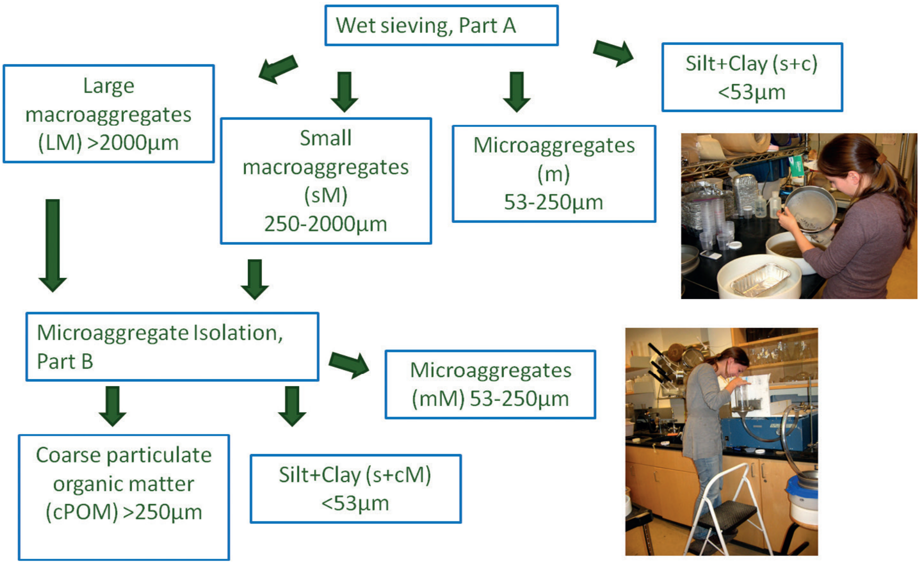

Figure 5. Soil fractionation protocol.

The mean weight diameter (MWD) of the aggregates was calculated according to Van Bavel (1949). Carbon content of all fractions from wet sieving and microaggregate isolation were analyzed with a CN-analyzer (CN-2000 LECO Corp., St Joseph, MI, USA (II, III) and PDZ Europa ANCA-GSL/20-20 isotope ratio mass spectrometer Sercon Ltd., Cheshire, UK (II) or Costech Instruments, ECS 4010 (IV). The SOC content of the different aggregates and total SOC stock calculations for all study sites were based on the equivalent soil mass method which takes soil bulk density into account (Ellert and Bettany, 1995; Lee *et al.*, 2009) (I, II, III, IV). Organic matter content was derived from C content using the Van Bemmelen factor of 1.724 (III).

2.3 N_2O FLUX MEASUREMENTS

A closed chamber technique was used to measure N_2O fluxes at sites 1–4 from June 2008 to June 2010 (Fig 6) (I). The method was adapted from Regina and Alakukku

(2010). One base frame made of steel (60 cm x 60 cm) for the gas collection chambers was installed in each of the four replicate plots. The measurements were done with aluminum chambers, each closed with a water seal that was formed when a groove on the upper end of the frame was filled with water to ensure the gas-tightness of the chamber. In winter, NaCl was added to the water to avoid ice formation. The base frames were removed only during field operations, like tillage or harvesting, and installed back at the same locations afterwards. Venting tubes were installed on the chambers to minimize pressure changes (Hutchinson and Livingston, 2001). The emissions were measured biweekly, one field during one day between 10 a.m. and 2 p.m. and all fields during the same week. In October 2009, more frequent measurements were done to study the short-term effects of tillage operations on N_2O fluxes at sites 1 and 2. The first measurements were done between 6 and 10 hours after tillage operations, followed by measurements 24 h, 48 h, 4 days and 7 days after soil tillage.

Figure 6. Gas sampling at site 1 in Jokioinen in October 2009.

To measure N_2O flux, three gas samples were taken during a 30 minute chamber enclosure at 0, 15 and 30 minutes. At each time point, a 20 ml sample was taken with a plastic syringe (Becton, Dickinson and Company, Franklin Lakes, NJ, USA) and transferred immediately into a pre-evacuated 12 ml glass vial (Exetainer, Labco Ltd., High Wycombe, UK). Gas samples were analyzed within 72 hours of sampling using a HP 6890 Series gas chromatograph. Standard gas mixture (AGA Gas AB, Lidingö, Sweden) of known concentrations of N_2O was used for diluting a calibration curve with seven points. The gas production rate was calculated using equation 3:

$$F=\Delta C/\Delta t*V/A \qquad (3)$$

where C is the concentration of N_2O, t is time, V is chamber volume, and A is the chamber area. Calculation of the amount of N in the sample was based on the ideal gas equation. The chamber temperature was measured during each gas sampling. The cumulative annual fluxes for each management practice were calculated by linearly interpolating the emissions between consecutive sampling days. The fluxes were averaged by calculating the cumulative flux value for each chamber separately and then averaging the treatments in each field. The emission rates of N_2O were calculated as the mass of N in N_2O (N_2O -N).

2.4 DECOMPOSITION RATE OF CROP RESIDUES

Barley straw (*Hordeum vulgare*) and pea residue (*Pisum sativum*) bags were installed at study sites 1, 2 and 4 from November 2009 to September 2010 under two different tillage practices, CT and NT, to three different depths: on the top of the soil (0 cm), buried 10 cm deep, and buried 20 cm deep, (IV). Residue bags (10 x 15 cm) were made out of polyester mesh and 5 g of air dried, untreated barley straw or pea residue (peas were removed), was put into each bag. The bags with the crop residues in them were oven dried overnight at 40°C for a final weight. The bags were installed in two rows four meters from the end of the study plots and one meter from each side of the plots. The two rows were 50 cm apart from each other. We had four barley straw bags at each depth and two pea residue bags at each depth for a total of 18 residue bags at each study site. Half of these bags were pulled out at the end of April before the start of the growing season and half were left in the study plots until the end of the growing season (September). Afterwards the residue bags were air dried for a week, the residue samples then moved to paper bags and oven dried at 40°C before grinding the samples for analysis.

We calculated the loss on ignition (LOI) from the original barley straw and pea residue, field residue bag samples as well as soil samples by igniting the samples at 550°C for 5 days in a high temperature muffle furnace. This leaves the mineral part of the soil as ash while organic matter is burnt. The coefficient calculated from the original crop residues was used to correct the calculations of the amount of barley straw and pea residue that was decomposed while the bags were installed in the field. The results were also corrected for the OM in the residue bags originating from soil.

3. RESULTS

3.1 SOIL AGGREGATION AND SOIL ORGANIC CARBON

3.1.1 TOTAL SOC STOCKS

The total SOC stocks were significantly different between management practices only at site 5 where the mean SOC stock in the 0–15 cm soil layer was lower in RT compared to CT with averages across the three straw treatments of 46.8 Mg C ha^{-2} and 47.6 Mg C ha^{-2} in RT and CT respectively (Table 2) (III). Straw treatment had no effect on total SOC stocks and the difference between the tillage practices did not vary with straw treatment (III). *L. terrestris* midden soils had a significantly increased concentration of total SOC in the 0–5 cm soil layer when analyzed across all sites compared to surrounding bulk soil (Table 2) (IV).

3.1.2 TOPSOIL AGGREGATION

Mean weight diameter (MWD), which is an indicator of aggregate stability at the time of sampling, of the whole soil had a decreasing trend in the order of NT > RT > CT at all study sites, including within different straw management practices (Table 2) (II, III). Statistically significant differences were found at sites 1 and 4 between CT and NT (II). RT treatment differed significantly compared to CT only at site 5 where RT increased the MWD (III). The largest relative difference between treatments was found at site 4 where MWD in NT was twice as high compared to CT. Soil type also affected the general aggregate stability as MWD was higher at sites 2 and 3, with the MWD reaching up to 2.1 mm in NT at site 2, while the lowest values were found in the coarse soil at site 4 where the MWD of CT and NT was as low as 0.28 mm and 0.58 mm, respectively. MWD was significantly higher in the *L. terrestris* middens versus surrounding bulk soil at the clayey sites 1 and 2, but not at the coarse textured site 4 (IV).

No-till increased the amount of macroaggregates (LM and sM combined) at site 4 (II). Reduced tillage significantly increased the percentage of macroaggregates and decreased the percentage of m and s+c fractions at site 5 (III). More LM fractions were found in the *L. terrestris* midden soil than in the surrounding bulk soil at study sites 1 and 2 but less sM fractions at site 2 (IV). The greatest portion of LM fraction in middens was found at site 2 where they represented as much as 35% of the soil mass (II).

Table 2. Division of dry bulk density, mean weight diameter (MWD) and the different SOC components by soil layer and tillage treatment and *L. terrestris* midden versus bulk soil (arithmetic mean ± SE) (Data from Sheehy *et al.*, 2013 (I), Sheehy *et al.*, 2015 (II), Singh *et al.*, 2015 (III) and Sheehy *et al.*, manuscript (IV)).

Site	Treatment	Depth	Bulk SOC* Mg ha^{-2}	MWD mm	C in LM (M) g m^{-2}	C in sM g m^{-2}	C in m g m^{-2}	C in s+c g m^{-2}	C in mM** g m^{-2}
Site 1	CT	0-20 cm	57.3 (±3.5)a	0.55 (±0.07)a	88 (±16)a	2029 (±291)a	2817 (±13)a	792 (±49)a	1040 (±225)a
	RT	0-20 cm	61.2 (±2.8)a	0.59 (±0.06)ab	158 (±28)ab	2623 (±194)a	2510 (±178)a	803 (±43)a	1480 (±120)a
	NT	0-20 cm	61.2 (±2.5)a	0.84 (±0.10)b	478 (±146)b	2844 (±208)a	2172 (±192)a	625 (±35)a	1834 (±250)a
	Midden	0-5 cm	20.2 (±0.5)a	0.85 (±0.05)a	440 (±45)a	859 (±31)a	555 (±44)a	177 (±14)a	207 (±16)a
	Bulk	0-5 cm	19.0 (±0.5)b	0.63 (±0.04)b	196 (±17)b	761 (±72)a	758 (±85)a	226 (±22)a	90 (±11)b
Site 2	CT	0-20 cm	59.7 (±1.3)a	1.36 (±0.27)a	650 (±290)a	3322 (±26)a	1488 (±89)a	507 (±46)a	2449 (±256)a
	RT	0-20 cm	56.9 (±1.2)a	1.36 (±0.15)a	841 (±186)b	3151 (±200)a	1266 (±73)ab	430 (±38)ab	2451 (±83)a
	NT	0-20 cm	60.8 (±0.6)a	2.10 (±0.20)a	1773 (±54)b	3036 (±56)a	954 (±58)b	320 (±17)b	3014 (±27)a
	Midden	0-5 cm	18.0 (±0.5)a	0.68 (±0.08)a	737 (±59)a	657 (±48)a	304 (±24)a	98 (±8)a	365 (±26)a
	Bulk	0-5 cm	16.6 (±0.3)b	0.66 (±0.03)b	305 (±30)b	895 (±34)b	329 (±32)a	112 (±11)a	170 (±24)b
Site 3	CT	0-20 cm	83.7 (±3.9)a	1.20 (±0.17)a	888 (±272)a	4855 (±268)a	1896 (±308)a	730 (±86)a	3802 (±170)a
	NT	0-20 cm	69.4 (±4.1)a	1.58 (±0.18)a	1354 (±221)a	3579 (±244)b	1442 (±64)a	563 (±34)a	3358 (±376)a
Site 4	CT	0-20 cm	53.0 (±0.3)a	0.28 (±0.01)a	1067 (±91)a	n.d.	3198 (±108)a	1038 (±22)a	508 (±37)a
	NT	0-20 cm	49.5 (±1.5)a	0.58 (±0.03)b	1998 (±130)b	n.d	2208 (±101)b	747 (±25)b	1158 (±84)b
	Midden	0-5 cm	17.3 (±0.9)a	1.16 (±0.04)a	229 (±21)a	663 (±53)a	519 (±42)a	112 (±11)a	111 (±9)a
	Bulk	0-5 cm	16.9 (±0.8)b	0.94 (±0.05)a	207 (±20)a	834 (±56)a	492 (±48)a	220 (±22)a	117 (±12)a
Site 5	CT	0-5 cm	15.4 (±0.6)	0.55 (±0.01)a	39 (±6.9)a	576 (±41)a	637 (±61)a	188 (±12)a	298 (±30)a
	CT	5-10 cm	16.0 (±0.4)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	CT	10-15 cm	16.2 (±0.7)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	CT	15-20 cm	7.6 (±0.3)	0.68 (±0.03)b	37 (±5)a	345 (±26)a	262(±27)a	73 (±6)a	217 (±25)a
	CT	20-40 cm	33.8 (±4.8)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	RT	0-5 cm	15.8 (±0.6)	0.67 (±0.03)a	93 (±19)a	675 (±45)a	528 (±47)a	161 (±7)a	401 (±37)a
	RT	5-10 cm	16.1 (±0.5)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	RT	10-15 cm	14.9 (±0.7)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	RT	15-20 cm	6.4 (±0.4)	0.79 (±0.02)b	53 (±11)a	343 (±23)a	159 (±12)a	50 (±4)a	228 (±25)a
	RT	20-40 cm	30.3 (±4.6)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.

n.d. = not determined
*Calculated for the mass of the soil in the corresponding soil layer.
**Values for midden and bulk soil are from within LM fraction only
Letters a, b indicate differences between treatments within each site at $p<0.05$

Small macroaggregates were the dominant soil fraction in CT and NT at sites 2 and 3 representing more than half of the sample size (53% and 55%, respectively) and under NT at site 4, whereas free microaggregates dominated at site 1 and under CT at site 4 occupying 47% and 65% of the soils, respectively (II). At site 5 the deeper soil layer (15–20 cm) had proportionally more LM and sM fractions than the topsoil layer (0–5 cm). However, the difference in the aggregate composition between the two soil layers tended to be smaller when straw was retained in the soil compared to when straw was removed or burnt (III). The amount of free microaggregates in the soil was significantly lower in midden soil compared to bulk soil at site 1 (IV).

The composition of macroaggregate-occluded soil fractions was less variable with mM fraction being the dominant fraction at all study sites under the different tillage practices (II, III). Site 3 had relatively the largest amount of mM fraction within macroaggregates (g mM fraction g^{-1} M) with 63% in CT and 64% in NT. However, NT or RT did not significantly increase the amount of mM fraction present in the 0–20 cm soil layer compared to CT at sites 1–4 (II). At site 5 there was evidence that the two measured soil layers deviate from each other in terms of aggregate composition with significantly more cPOM in the 15–20 cm soil layer compared to the top layer (0–5 cm) (III). Within the *L. terrestris* midden soil the proportional weight of cPOM from LM fractions was significantly higher compared to bulk soil at site 2 (IV). On the other hand, the proportional weight of mM formed within LM fractions was significantly lower in midden soil than surrounding bulk soil at site 2. Within sM fraction, the proportional weight of cPOM was higher in the middens versus surrounding bulk soil at site 2 and lower at site 1 (IV). The proportion of mM fraction within sM fraction was significantly higher in the middens at site 1, lower in the middens at site 2 and without a difference at site 4 (IV).

3.1.3 AGGREGATE-ASSOCIATED C IN TOPSOIL

Enrichment of SOC was observed in the LM fraction at sites 1, 2 and 4 (Table 2) (II). This effect was diluted at some sites by a loss of SOC in sM with a statistically significant depletion of SOC content at site 3. SOC content in both m and s+c fraction decreased significantly in NT compared to CT at sites 2 and 4 (II). RT had no effect on the SOC content compared to CT (Table 2) (II, III). Enrichment of SOC in *L. terrestris* middens was found only in LM fraction after wet sieving with significant differences at sites 1 and 2 (Table 2) (IV).

The amount of SOC in mM fraction increased significantly at site 4 in NT compared to CT (Table 2) (II). No significant differences were found between RT and the other two treatments. Increases in SOC content under NT compared to CT were also found in macroaggregate-occluded cPOM fraction at site 4 and s+cM fraction at sites 1, 2 and 4 in (II). We observed an increase in SOC content in *L. terrestris* middens compared to bulk soil in cPOM, mM and s+cM within LM fractions at sites 1 and 2 and cPOM at site 4 (Table 2) (IV). Within sM fractions, a decrease of SOC in middens was found in mM fraction at site 2 (IV). However, a redistribution of SOC into bigger aggregates was detected in *L. terrestris* middens at all sites.

3.2 N_2O FLUXES AND RELATED ENVIRONMENTAL PARAMETERS

Largest N_2O fluxes were observed between April and July (I: Fig 4). High peaks in April are due to snow melt and thawing of frozen soil. Larger peaks in May and June occurred after combined sowing and fertilization as well as rain events. Smaller peaks in N_2O fluxes were observed also after individual high rain events. Tillage in the autumn of 2009 did not have any statistically significant effects on the observed N_2O fluxes. The lowest hourly flux was observed at site 2 where the flux was typically below 0.05 mg N_2O-N m^{-2} h^{-1}. In general, average fluxes ranged from 0.05 to 0.14 mg N_2O-N m^{-2} h^{-1} for the clayey soils and from 0.06 to 0.09 mg N_2O-N m^{-2} h^{-1} for the coarse soil.

Annual cumulative emissions of N_2O for years 2008 to 2009 and 2009 to 2010 ranged from 2.4 kg N_2O-N ha^{-1} yr^{-1} to 10.2 kg N_2O-N ha^{-1} yr^{-1} (Fig 7) (I). The lowest annual N_2O flux was found in the first year in CT at site 2 and the greatest in the second year in NT at site 3. The largest difference in annual N_2O fluxes between management practices was observed in the first year at site 1 where N_2O emissions were 150% and 90% greater in NT soils compared to their CT and RT counterparts, respectively. Emissions of N_2O were significantly higher in NT compared to CT at site 3 and lower at site 4 (I). Cumulative N_2O emissions in RT were significantly lower compared to NT at site 1 (I).

There were no differences in the potential denitrification between the tillage treatments but statistically significant differences were found between different study sites (I), the values being greatest at site 2 and lowest at site 4. Significant differences in NO_3^- between treatments were only found in October 2009 at site 4. In 2009, all fields had more NH_4^+ left after harvest in NT compared to CT treatments. Significant differences in the NH_4^+ levels were found at sites 1, 3 and 4. Highest values of NO_3^- after harvest were found at site 2 in 2008 and site 1 in 2009 and the lowest values at site 3 in both studied years.

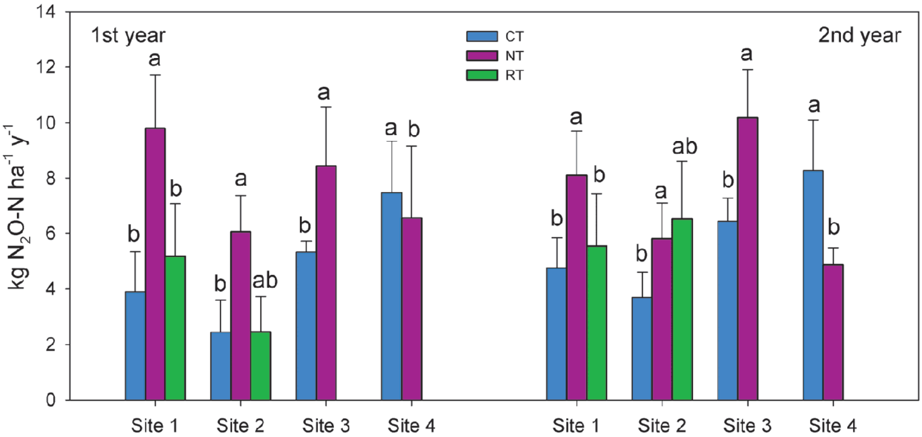

Figure 7. Cumulative N_2O-N fluxes (kg ha^{-1} yr^{-1}±SE) for CT, RT and NT. Letters indicate differences between treatments within each site at p<0.05 (Data from Sheehy *et al.*, 2013 (I)).

No-till increased WFPS compared to CT during the growing seasons of 2008 and 2009 and was significantly different between management practices even though temporal variation was relatively high (I). The highest peak of 87% WFPS was found at site 2 after a heavy rainfall event. The average WFPS was between 45–59% in CT while the average values in NT were between 53–71% (I). The frost went deeper and developed faster in the fall in CT compared to NT. Depth of frost in RT was between that of CT and NT (I). Soil temperature varied less in NT by being higher during the colder periods of the year and slightly cooler during hot summer days (I).

The strongest correlations between N_2O fluxes and environmental parameters were found between WFPS, soil bulk density and earthworm burrows which all had a significant positive correlation with the emissions (Table 3).

3.3 DECOMPOSITION RATE OF CROP RESIDUES

Barley straw decomposed significantly faster under NT compared to CT at sites 1 and 4 and pea residue at sites 1 and 2 (Fig 8; unpublished results). The average decomposition rate was 57%, 56% and 59% at sites 1, 2 and 4, respectively, under CT, and 65%, 62% and 60% at sites 1, 2 and 4, respectively, under NT.

Table 3. Pearson correlations between different environmental/soil parameters and continuous or cumulative N_2O fluxes.

	r	R^2	p	N
	N_2O flux (continuous)			
Air Temperature	-0.029	0.008	0.205	1974
Soil Temperature, 5 cm	-0.048	0.009	0.068	1446
Precipitation	-0.034	0.021	0.127	1974
Water-Filled Pore Space, 15 cm	0.554	0.307	<0.001	403
	N_2O flux (cumulative)			
Soil Bulk Density, 0–20 cm	0.406	0.165	<0.001	64
Earthworm Burrows,	0.346	0.120	0.005	64
Soil Organic Carbon, 0–20 cm	0.156	0.017	0.190	72
Total Nitrogen, 0–20 cm	0.130	0.024	0.277	72
Nitrate (NO_3^-), 0–20 cm	-0.315	0.099	0.048	40
Ammonium (NH_4^+), 0–20 cm	0.325	0.106	0.041	40
Potential denitrification, 0–20 cm	-0.302	0.091	0.015	64

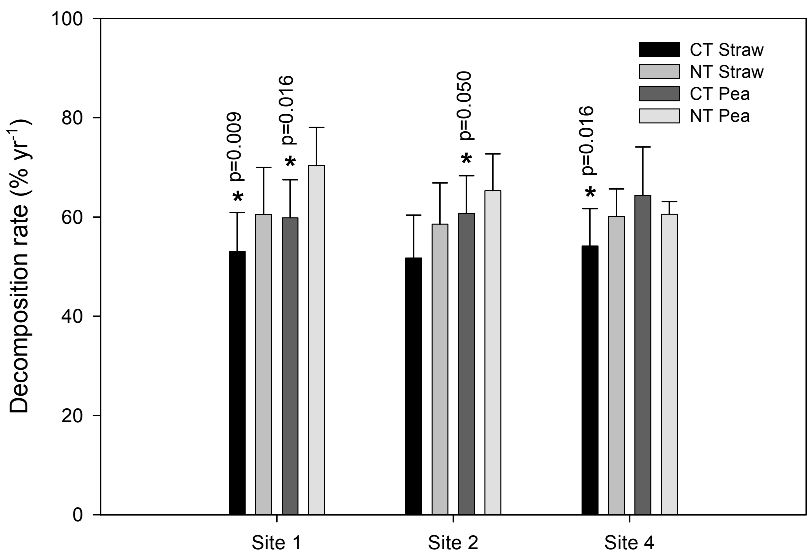

Figure 8. Residue decomposition as % of the original mass of barley straw and pea under CT and NT. Statistically significant differences between tillage treatments within different litter are denoted by a star (and p-value) (Part of the data from Sheehy *et al.*, 2015 manuscript (IV)).

4. DISCUSSION

4.1 CARBON SEQUESTRATION POTENTIAL IN CEREAL CROP PRODUCTION

The common understanding is that reducing tillage intensity increases soil C stocks. The relative C stock change factors used by IPCC (2006) for agricultural soils that have been under NT or RT for over 20 years, are 1.15 and 1.08 for humid boreal climate, for NT and RT, respectively. This means, for example, an increase of 8% in soil C stock under RT in humid boreal climatic conditions during 20 years. Contrary to this we found a decrease of total SOC in the topsoil layer of 0–15 cm in 30 years under RT compared to CT (III) and no significant changes in the SOC content in the topsoil layer of 0–20 cm in 10 years under NT or RT (II). This is in contrast to most studies finding minimum tillage practices to sequester SOC from temperate regions to the tropics and in various soil types, including a Spanish Aridisol (Alvaro-Fuentes *et al.*, 2009), two Brazilian Oxisols (Denef *et al.*, 2007) and Alfisols in Kentucky (Chung *et al.*, 2008). However, in a study conducted in eastern Canada, NT was found to decrease the SOC stock close to the depth of the ploughed soil layer (Gregorich *et al.,* 2009), and a study in France found no significant changes in SOC content between NT, RT and CT in 41 years (Dimassi *et al.,* 2014).

Relatively high soil moisture content may be a limiting factor for C sequestration in humid and boreal climatic conditions favoring decomposition in Finnish croplands. There is increasing evidence that a fairly low precipitation is a prerequisite for high C gains in minimum tillage practices. This difference between arid and humid regions was clearly shown in a data compilation in Canada where NT practice increased soil C stock only in the more arid west (Vandenbygaart *et al.*, 2003). In addition, Dimassi *et al.* (2014) found a negative linear relationship between the mean annual precipitation and C stock change in NT. The importance of aggregate breakdown in the topsoil layer may be high in the boreal climate with frequent winter freezing and thawing of the soil which will break down soil macroaggregates created during the summer months (Le Guillou *et al.*, 2012). Since the formation of LM fraction and macroaggregate-occluded microaggregates is mainly driven by soil biota (Six *et al.*, 2006) not only the disruption but also the formation of aggregates has a seasonal trend. One reason for the small differences in C sequestration rates between the management practices found in this study could be that the effect of the freeze-thaw cycles is significant enough to negate the positive effects of minimum tillage or other management practices (VandenBygaart *et al.*, 2003; Le Guillou *et al.*, 2012; Edwards, 2013).

The results showed increased decomposition rate under NT compared to CT at sites 1, 2 and 4. At site 5, the reduced amount of SOC, accompanied by increased *L. terrestris* density, higher microbial C and mixing of residues within the topsoil layer under RT, also point to an increased decomposition rate (III). The straw mixed into the soil by the cultivator may be even more efficiently decomposed than the plowed straw that enters deeper in the soil. Also, the high densities of *L. terrestris* may have directly and indirectly accelerated the decomposition in RT, particularly that of the surface residues, through the efficient consumption and incorporation of straw (Bohlen *et al.*, 1997). Angers *et al.* (1997) suggested that the decomposition rate of crop residues incorporated deep into the soil slows down under CT due to limited aeration under these climatic conditions. It is also possible that high amounts of crop residues stratified in the topsoil in NT practice, accelerates microbial activity (Hungria *et al.*, 2009). In Finland the topsoil is usually also warmer and has higher soil moisture content under NT compared to CT especially during fall and winter months favoring higher decomposition (I). Some studies have shown that rapid breakdown of newly formed soil aggregates may occur if increased amount of earthworms also enhances the mineralization of polysaccharides and other organic gluing compounds (Guggenberger *et al.*, 1996; Ge *et al.*, 2001). This highlights that a faster decomposition rate under NT, and likely RT, possibly hinders C sequestration rate under these management practices.

Traditionally clay soils are thought to encompass a lower decomposition rate due to physical and chemical protection, thus enabling SOC accumulation (von Lützow *et al.*, 2014; Six and Paustian, 2014), but there are some studies contradicting this by showing higher levels of decomposition in clay soils (Müller and Höper 2004; Dilustro *et al.*, 2005; Wei *et al.*, 2014). The results showed a significant increase in the decomposition rate of pea residue in clayey textured site 2 versus coarse textured site 4, thus highlighting the complicated nature of factors affecting decomposition rate and C dynamics in soils (IV).

Agroecosystems under NT may also experience reduced yields which in turn may negatively affect C sequestration rates due to reduced amounts of residue (West and Post, 2002; Venterea *et al.*, 2006; Blanco-Canqui and Lal, 2008). Reduced yields were found at sites 1 and 2 where the average yield since transition has been 4% and 12% lower during the years 2001–2010, respectively, under NT compared to CT (II). This was also the case at site 3 where the average yield of the last decade in CT was as much as 22% higher compared to NT. This might be one of the key factors affecting the SOC depletion that was discovered in the sM fraction at site 3 in NT compared to CT. Similar results have been reported by Follett *et al.* (2001) and Lal *et al.* (1999) on Bojac soil with lower yields and reduced amounts of residue returned to the field each season resulting in lower soil C content in NT compared to CT. On the other hand, we found a significant loss of SOC under RT compared to CT at site 5 regardless of straw management practice while yields did not differ significantly between RT and CT. It is possible that the root C inputs are more dominant than shoot C inputs into the soil under minimum tillage practices compared to CT (Rasse *et al.*, 2005). It may also be that the C inputs matter less than the decomposition rate of the soil.

There is a possibility that the clayey soils of sites 1–3 and 5, which had significant C redistribution into bigger aggregates but no added SOC under NT or RT versus CT, also had a low saturation deficit and therefore less potential to accumulate SOC. High SOC content of clayey soils is generally acknowledged (Nichols, 1984; Wiseman and Puttmann, 2006; Heikkinen *et al.*, 2013) due to physical protection through a network of smaller sized pores and chemical protection through stronger adsorption of SOC on to the soil minerals (Chan *et al.*, 2008; West and Six, 2008). Every agroecosystem has its own equilibrium SOC stock and therefore a limit to saturate added C (Stewart *et al.*, 2007). A Canadian compilation of studies found that a C content of >45 Mg ha^{-1} was limiting C gain in management changes (VandenBygaart *et al.*, 2003). Our study sites 1–3 had a range from 57 to 84 Mg C ha^{-1} (0–20 cm) and site 5 an average value of 47 Mg C ha^{-1} (0–15 cm). Also, long-term studies have shown that soil C stock does not necessarily increase with higher C inputs (Paustian *et al.*, 1997b; Reicosky *et al.*, 2002), and the efficiency to stabilize SOM decreases in soils with high C levels compared to soils with lower C levels under the same management practices (Campbell *et al.*, 1991).

Several studies support the view of Six and Paustian (2014) that the mM fraction serves as a diagnostic fraction for management induced changes in SOC levels (Denef *et al.*, 2004; Kong *et al.*, 2005; Chung *et al.*, 2008). Findings show that, of the increase in SOC levels under NT compared to CT, 49–112% can be attributed to the change in the microaggregate-within-macroaggregate C (mM-C) (Six and Paustian, 2014). In our study this was only supported by results from coarse textured site 4 where mM fraction gained SOC under NT. However, with no significant increase in total SOC levels we cannot calculate the contribution of the change in mM-C. Even though we saw a depletion or no change in the overall C stock at the clayey sites (sites 1, 2, 3 and 5) under NT or RT, there was a higher level of larger aggregates (increased aggregation level), and more SOC stored in the LM fractions (per unit of LM) under NT at sites 1 and 2 compared to CT as well as more SOC stored in the mM fraction in the top soil layer (0-5cm) under RT at site 5.

The contribution of earthworms to C sequestration is an intricate issue which has been described as the “earthworm dilemma” arising from the simultaneous effect of earthworms on C mineralization and stabilization (Lubbers *et al.*, 2013). Based on a meta-analysis of mainly relatively short term laboratory studies Lubbers *et al.* (2013) concluded that earthworms do not affect soil SOC stocks but increase CO_2 emissions by 33%. On the contrary, Zhang *et al.* (2013) challenged this view and claimed that C stabilization by earthworms overrides their enhancing effect on mineralization. The present study cannot clarify this controversy showing both positive and negative effects of earthworms in respect to climate. *L. terrestris* presence increases the potential to store C within LM and mM fraction and, in clay soils, enhances the aggregation level. However, the observed preferential C stabilization reached its full potential only at site 1 where 99% of the difference in total SOC in the 0–5 cm soil layer between midden and bulk soil was associated with the C found in macroaggregate-occluded microaggregates. At site 2 the corresponding number was 34%. At this site the increase in C found in mM within LM fraction was counteracted by a decrease of C in the mM within the sM fraction. This highlights the importance of this fraction as a microsite for C sequestration (Six and Paustian, 2014). On the other hand, higher earthworm abundance at sites 2 and 4 under NT versus CT (Visa Nuutinen, unpublished results) and higher *L. terrestris* density under RT versus CT at site 5 (IV) possibly contributed to higher decomposition rates. However, Fonte and Six (2010) argued that since most of the C in earthworm casts is associated with macroaggregate-occluded microaggregates the rate of decomposition in these casts would, with time, possibly decrease to a level below the level of non-ingested soil.

Some justified criticism has been given regarding the usefulness of aggregate fractionation to understanding soil dynamics (Young and Ritz, 2000; Young *et al.*, 2001), but it has been argued that the isolation of aggregates has its purpose as a complimentary tool in addition to other known techniques, like tomography (De Gryze *et al.*, 2006) or thin sectioning (Pulleman *et al.*, 2005), by revealing soil properties and dynamics that could not be seen by utilizing just one technique (Six and Paustian, 2014). The comparison of the different techniques is important for learning about the functioning of different organic binding agents in aggregates of different stabilities. For example, a comparison between less destructive sieving methods and slacking method (Kemper *et al.*, 1985) has revealed the role of SOM as a key binding agent to hold together microaggregates within macroaggregates (Six and Paustian, 2014). Also, some of the sites in this study were fairly young giving that the management practices were adopted only about a decade ago (sites 1–4). Six *et al.* (2004) concluded that the positive effects of NT may not show until at least a decade after the transition from CT, especially in humid climates. In addition, majority of the SOC analyses were done from the soil layer of 0–20 cm (sites 1–4; equivalent soil mass was taken into account) which may not be deep enough to accurately measure the changes in total SOC content or to see the C stratification effect on the topsoil under NT. At site 5 we changed our approach and sampled both deeper into the soil and in increments to better capture the effect of depth. However, most of the changes in SOC can be expected to happen in the affected soil layer of 0–20 cm and the results will therefore give a good estimate of changes happening in the studied agroecosystems. In addition, for the management practice effects to be meaningful from a climatic perspective the changes in SOC levels should be seen in the whole 0–20 cm soil layer even if stratification under NT is predominantly happening in the top 5 cm soil layer.

In current climatic conditions, tillage and straw management practices only have a minor effect on C dynamics in Finnish cereal cropping systems (Table 4). Increasing winter temperatures (Pirinen *et al.*, 2012; Jylhä *et al.*, 2014) may increase the freeze-thaw cycle frequency breaking down soil aggregates, and higher temperature and precipitation (Pirinen *et al.*, 2012; Jylhä *et al.*, 2014) could potentially lead to higher decomposition rates. These changes may increase SOC losses from cereal cropping systems in the future. However, NT management practice has been reported to be advantageous with respect to erosion control in boreal areas (Børresen and Ulén, 1991; Puustinen *et al.*, 2007). As one of the measures to decrease erosion and therefore C and other soil fixed nutrient transport from land to watercourses, Rural Development Programme for Mainland Finland 2014–2020 encourages increasing the crop or crop residue covered area outside the growing season when erosion and particulate phosphorus leaching peak (Ministry of Agriculture and Forestry, 2014; Puustinen *et al.*, 2007). However, this phenomenon was not measured and therefore its effect cannot be estimated in the scope of this study.

Table 4. Potential climatic effects of NT and RT compared to CT expressed as + increasing emissions, - decreasing emissions and 0 no significant difference.

	Site 1		Site 2		Site 3	Site 4	Site 5
	NT vs CT	RT vs CT	NT vs CT	RT vs CT	NT vs CT	NT vs CT	RT vs CT
N_2O	+	0	+	0	+	-	n.d.
Total SOC	0	0	0	0	0	0	+
SOC in mM	0	0	0	0	0	-	-/+
Decomposition rate	+	n.d.	+	n.d.	n.d.	+	n.d

n.d. = not determined

4.2 FACTORS CONTROLLING N_2O FLUXES

The mean annual N_2O-N emissions, from all sites and treatments combined, were 5.8 kg ha^{-1} yr^{-1} in the first year and 6.4 kg ha^{-1} yr^{-1} in the second year (I), which are a little higher than found in a compilation of N_2O measurements done between 2000–2009 in Finland with average fluxes of 3.5 kg N_2O-N ha^{-1} yr^{-1} under annual crop production (Regina *et al.*, 2013). However, our data includes NT management practice contrary to the above mentioned results, with values that fall within the reported ranges for mineral agricultural soils (0.8 kg N_2O-N ha^{-1} yr^{-1} to 24 kg N_2O-N ha^{-1} yr^{-1} in NT and 1.8 kg N_2O-N ha^{-1} yr^{-1} to 13.2 kg N_2O-N ha^{-1} yr^{-1} in CT) (Aulakh *et* *al.*, 1984; MacKenzie *et al.*, 1998; Ball *et al.*, 1999; Kaharabata *et al.*, 2003; Syväsalo *et al.*, 2004; Syväsalo *et al.*, 2006; Chatskikh and Olesen, 2007).

In alignment with our hypothesis, NT increased N_2O emission in the clayey textured sites by 86% on average negatively affecting the net GHG balance (Table 4). Thus, for the clayey soils, the results are consistent with the results from similar climatic conditions from Canada compiled by Gregorich *et al.* (2005) indicating that NT increases N_2O emissions from clayey soils in a humid climate. Six *et al.* (2004) came to the same conclusion and added that the increase is most distinct during the first decade after converting to NT, but the emissions from NT systems might decrease after that. The study sites have been under NT for approximately 10 years and may have still been experiencing the changes associated with conversion. A long, continuous period of NT practices may gradually improve the soil structure as more soil aggregates form and the biological activity enhances and adapts to the new conditions improving, for example, the aeration of the soil. However, in an experiment in France the N_2O emissions did not decrease and were consistently higher in NT compared to CT 32 years after the conversion (Oorts *et al.*, 2007). N_2O flux data from site 4 with coarse soil texture, however, gave contrasting results, i.e., the emissions in NT were lower both years compared to CT. This has also been found in an experiment established in Denmark on loamy sand soil in 2002 where N_2O fluxes measured from 2003 to 2005 were lower from NT compared to CT (Chatskikh *et al.*, 2008). Sand or clay content has been shown to often correlate with measured N_2O emissions (Chadwick *et al.*, 1999; Bouwman *et al.*, 2002; Freibauer, 2003).

Poor aeration accompanied with favorable soil moisture content and temperature are the most likely driving factors of greater N_2O emissions of the clayey soils under NT. The higher soil bulk density at sites 1 and 3 with lower micro- and macroporosity (Regina and Alakukku, 2010) indicates that wet soil was less aerated in NT compared to CT. Water-filled pore space (WFPS) was rarely above 60% in CT but in NT it was more often above this threshold, which is thought to induce denitrification in the soil (Linn and Doran, 1984). While WFPS was occasionally as low as 30% in CT during the growing season, it stayed above 50% in the NT systems at sites 1 and 2, while the NT system on the coarse textured soil at site 4 had lower levels of WFPS highlighting the better water holding capacity of clayey soils. Soil WFPS explained the variability of N_2O emissions better than precipitation. These results are in accordance with the results of Rochette (2008) who concluded that N_2O emissions are generally increased in NT in poorly aerated soils but not necessarily in medium or well aerated soils with better drainage.

The average difference in annual N_2O emissions between CT and NT was 2.9 and 1.4 kg N_2O-N ha^{-1} yr^{-1} in the first and second year, respectively, and reflected differences in WFPS. This was very close to the average difference of 2 kg N_2O-N ha^{-1} yr^{-1} reported by Rochette (2008) on poorly aerated soils. However, it should be noted that the differences between CT and NT at the different study sites ranged from

-2.2 to 4.6 kg N_2O-N ha^{-1} yr^{-1} on the coarse loamy soil and clayey soils, respectively. Since the global warming potential of N_2O is about 300 times greater than that of CO_2 and there was no increase in the total SOC under NT, the higher emissions of N_2O in the clayey soils under NT are effectively having a negative effect on the net GHG balance. The situation is reversed at site 4 where the positive net impact of NT is twice as high as the negative impact of NT on the clayey soils. The farmer at site 4 reduced the N fertilizer rate (about 15%) in NT, which might partly explain the lower emissions for this field. However, our data is too small to accurately calculate the N_2O losses related to the difference in N application. Also, at this site there was different fertilizer types used in CT versus NT which reduces the comparability of the treatments.

We observed a clear increase in N_2O emissions after fertilization every year. Sometimes there was a delayed effect probably due to lack of moisture in the soil but the first rain events after fertilization triggered denitrification activity. Elevated levels of N_2O have been reported several weeks after fertilization events (Gregorich *et al.*, 2008; Zebarth *et al.*, 2008). In support of findings in other studies (Maljanen *et al.*, 2003; Regina *et al.*, 2004; Regina and Alakukku, 2010), our data also showed large peaks in N_2O fluxes after soil moisture content rose as a result of snow and frost melt in the spring creating optimum conditions for denitrification. It has been shown that thawing also accelerates microbial activity (Regina *et al.*, 2004) and increases amounts of NH_4^+, NO_3^- and dissolved organic C in the soil (Herrmann and Witter, 2002; Jacinthe *et al.*, 2002). Jungkunst *et al.* (2006) reported that agricultural lands that are characterized by regular freeze-thaw events are more likely to have higher N_2O–to–N-input ratios compared to similar soils characterized by less regular frosting. Boreal soils may be even more susceptible due to the long period of no plant uptake of N (Syväsalo *et al.*, 2004). In this study, the winters were cold with very few freeze-thaw events and winter-time emissions were relatively low, about 40% of the annual emissions.

The presence of earthworms in the soil has been reported to accelerate the denitrification by soil-derived bacteria (Drake and Horn, 2006). This was supported by the results of this study with a significant positive correlation between the amount of earthworm burrows and N_2O emissions. Higher amounts of earthworm burrows and N_2O emissions were found in NT compared to CT at site 1, which might partly explain the higher emissions in NT at this site.

In the face of global climatic change, Finland is expected to have warmer winters and more annual rainfall (Pirinen *et al.*, 2012; Jylhä *et al.*, 2014), possibly introducing more frequent freeze-thaw cycles and rain events, which could potentially lead to increased occurrence of high N_2O emission peaks from agricultural soils. In addition, NT cereal crop management practice is gaining popularity in Finland, and as shown by our data, can potentially increase N_2O emissions. Since the increase of N_2O emissions in NT appears to be closely related to soil aeration, mitigating these emissions requires special attention to soil structure and drainage. Also, a long continuous period of NT practices may gradually improve the soil structure as the biological activity enhances and adapts to the new conditions. On the other hand RT did not show any significant differences compared to CT in cumulative N_2O emissions in the studied years making RT a notable option for clayey soils.

5. CONCLUSIONS

Climatic benefits of either minimum tillage or straw retention seem small in the humid boreal climatic region since soil C levels did not increase under NT, RT or straw retention and N_2O emissions increased in clayey soils under NT. While no C sequestration was observed, as expected, some improvements were seen in aggregate stability in NT cropping systems compared to CT in addition to increased amount of earthworms. No-till also increased the amount of SOC in microaggregates within macroaggregates in the coarse soil underlining a potential for future C accumulation in this soil type. *L. terrestris* mediated the formation of soil aggregates and the increase of SOC in soil particles within LM fractions without showing an effect on overall C sequestration. In addition *L. terrestris* may enhance the decomposition rate in the soils under NT and RT. The low C sequestration potential of minimum tillage practices is most likely the result of a humid and cool boreal climate which causes increased soil moisture content and decomposition rate compared to arid regions where most of the research on this issue originates. In addition, frequent freeze-thaw cycles as well as high original SOC content are likely to hinder C sequestration. On the other hand a long, continuous period of NT practices may gradually improve the soil structure as the aggregation level increases and the biological activity enhances and adapts to the new conditions. Increased N_2O emissions on clayey soils were most likely due to more dense soil structure with increased soil moisture content and poor aeration in NT compared to CT. On the contrary to our hypothesis we found lower N_2O emissions under NT in the coarse soil but it may have been due to decreased level of applied N fertilizer at that site compared to CT. Reduced tillage, with N_2O emissions comparable to CT, is a notable option for clayey soils to mitigate potentially increasing N_2O emissions. In general, mitigating N_2O emissions requires special attention to soil structure and drainage since increased soil moisture content and soil density under NT correlate with higher emissions.

6. FUTURE PERSPECTIVES

Since the role of SOC in relation to climate change was realized in the 1980's a mounting number of research projects globally have focused on SOC. However, because of the complicated nature of soil C dynamics, the wide array of soil types, climatic conditions, human perturbations, and other environmental factors that play a role in determining the fate and speed at which SOC is processed through the soil, it has been difficult to determine how SOC content changes in soils in relation to climate change and climatic conditions in general. There is a need for more detailed soil information in climate models. Development of climate and soil C models would highly benefit from technological improvements enabling accurate, less time consuming sampling of SOC. This would allow for higher amounts of soil samples over larger areas of land while modeling can also offer a complimentary approach to measurement based assessments.

In Finland, some of the main goals of future SOC research could be: 1) maintaining or establishing new long-term field experiments across different landscapes and management practices in addition to model development to monitor changes in SOC over time and space, 2) focusing on identifying the key components affecting C sequestration potential in mineral soils (clay soils) especially in relation to root versus shoot inputs and fungal contributions and 3) measuring and monitoring SOC changes in peat soils which comprise of large amounts of soil C. Climate change will continue to affect agroecosystems at large and even a small change in the soil C dynamics is likely to have a significant climatic impact. The above-mentioned research accompanied by model development would enable a more timely and accurate response to climate change induced changes in soil C dynamics.

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