THE ROLE OF ZERO-TILLAGE IN CLIMATE CHANGE MITIGATION

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Abstract

An estimated 11% of the world's land surface is under cultivation for crop production, and an additional 6.6% of the land will need to come into cultivation to feed a population of 9.1 billion people in 2050. Agricultural soils are a significant source of greenhouse gas emissions, contributing one-fifth of CO_2 emissions globally, one-third of CH_4 emissions and two-thirds of N_2O emissions. Changes in the management of agricultural soils can affect their role as a source or sink in the global carbon cycle, and the scale and composition of their greenhouse gas emissions. Therefore, understanding the role of different crop cultivation regimes in regulating these processes is a key global challenge

Conventional tillage is a traditional technique for crop production which has been practiced for centuries. This approach is beneficial in destroying large soil clods, burying weeds, preparing the seedbed and incorporating crop residues into the soil profile. However, through the process of loosening the upper 15-20 cm of soil, this management can result in large scale topsoil erosion. The annual soil loss from a plough-based farming averages 1.5 mm of erosion, which is almost 90 times faster than which new soil is produced. Zero-tillage, whereby the soil surface is not mechanically loosened, and the crop residue remains on the surface, is an effective management strategy to reduce soil erosion and limit water runoff. As a result, there is extensive evidence that zero-tillage is beneficial for the functioning and quality of the soil in many agricultural settings. There has, however, been contrasting evidence surrounding the climate change mitigation capabilities of zero-tillage in comparison to conventional tillage.

The current literature suggests that zero-tillage practices may reduce greenhouse gas emissions in terms of decreased CO_2 emissions, but an increase in the more damaging N_2O emissions may offset any potential benefits. Uncertainty also remains regarding the extent to which zero-tillage can sequester more carbon throughout the soil profile, with some studies highlighting incorrect sampling and calculation methodologies which have overestimated the potential of zero-tillage.

This project assesses the extent to which zero and conventional tillage can play a role in climate change mitigation across commercial and experimental farms, as well as evaluating the magnitude of change over time, and across contrasting climates. The results indicate an increase in soil organic carbon stocks in zero-tilled compared to conventionally tilled soils which was both microbially and physically mediated. X-ray Computed Tomography revealed that zero-tillage significantly increased soil porosity (with a mean increase of 13%) and pore connectivity (by a mean increase of 4%) in the long-term but decreased inter aggregate porosity (by 4%), providing a potential physical mechanism for the protection of soil organic carbon. Carbon analysis indicated that zero-tilled soils feature a large labile pool comprising the increased presence of lignin and polysaccharides.

Moreover, soils under zero-tillage for 31 years in Brazil also showed a shift in the chemical composition of organic carbon to a more recalcitrant fraction following long-term zero-tillage, suggesting soils were accumulating rather than mineralising soil organic carbon. Furthermore, across both the U.K and

Brazilian soil samples, global warming potential was at least 30% lower from zero-tilled soils compared to conventionally tilled soils, primarily driven by lower CO_2 emissions under zero-tillage. Moreover, the net global warming potential of zero-tillage systems decreased further with time in management.

However, further factors, including the effect of management on crop yield and nutrient efficiency need to be explored to understand the broader implications of widespread adoption. This study demonstrates that soil management practices strongly influence many biophysical and chemical properties, mitigating CO_2 emissions, alongside delivering further soil health benefits. These improvements, which will be leading drivers in the adoption of zero-tillage systems for farmers, include improved soil quality, increased soil pore connectivity, and reduced soil erosion. These effects have the potential to benefit soils globally, provided economic mechanisms are available to promote the implementation of zero-tillage as a mitigation strategy of global warming.

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List of abbreviations

µ	Micro
µm	Micron
∅	Porosity
ANOVA	Analysis of variance
BOXCOX	Transformation of data to normal distribution
CH_4	Methane
CO_2	Carbon dioxide
CT	Computed Tomography
ECAF	European Conservation Agriculture Federation
ESM	Equivalent soil mass
FAO	Food and Agriculture Organisation
FTIR	Fourier Transform Infrared spectroscopy
GC	Gas chromatography
GHG	Greenhous gas
GWP	Global warming potential
ha	Hectares
IPCC	Intergovernmental Panel on Climate Change
I Index	Immature soil organic matter
kg	Kilogram
kV	Kilovolt
LOI	Loss on ignition
M	Million
mg	Milligram
mM	Microaggregates formed within macroaggregate
N_2O	Nitrous oxide
R Index	Refractory soil organic matter
ROI	Region of interest
SC	Total soil carbon
SOC	Soil organic carbon
SOM	Soil organic matter
TOC	Total organic carbon
VG	Volume graphics

1.0 Global adoption of zero-tillage and its impacts on carbon storage and the regulation of greenhouse gas fluxes

1.1 Introduction

At present, an estimated 11% of the world's land surface is under cultivation for crop production (FAO, 2016). Furthermore, projections suggest that to feed a population of 9.1 billion people in 2050, another 6.6% of the land will need to come under cultivation, alongside increasing cropping intensity and improving yields (FAO, 2016). Conventional tillage is a traditional method for crop production, in which the soil is prepared for planting by complete inversion, typically with a mouldboard plough, to a depth of 20–30 cm (OECD, 2001). Subsequent passes are made across the soil with secondary tillage implements such as power harrows to smooth the soil surface (Fig. 1.1). This approach is beneficial in mechanically destroying large soil clods, burying weeds and incorporating crop residues into the soil profile. In the 1930s environmental concerns with regards to soil erosion, pollution of water bodies and significant fuel and labour costs associated with conventional tillage resulted in new concepts to reduce soil disturbance and to keep the soil covered, and the term conservation tillage was introduced (Kassam et al., 2009).

Conservation tillage, an umbrella term that encompasses all practices that minimise soil disturbance (including zero and minimum tillage, Fig. 1.1), has been proposed as an effective method to reduce soil erosion and limit water

runoff (Busari et al., 2015). According to the FAO (2014) and Eurostat (2013a), 'Conservation Agriculture' is characterised by three principles:

i) no-till or minimal mechanical soil disturbance (less than 25% of the soil surface),

ii) maintenance of permanent plant residue (at least 30%) on the soil surface for erosion control and moisture conservation,

iii) diversification by intercropping or rotation.

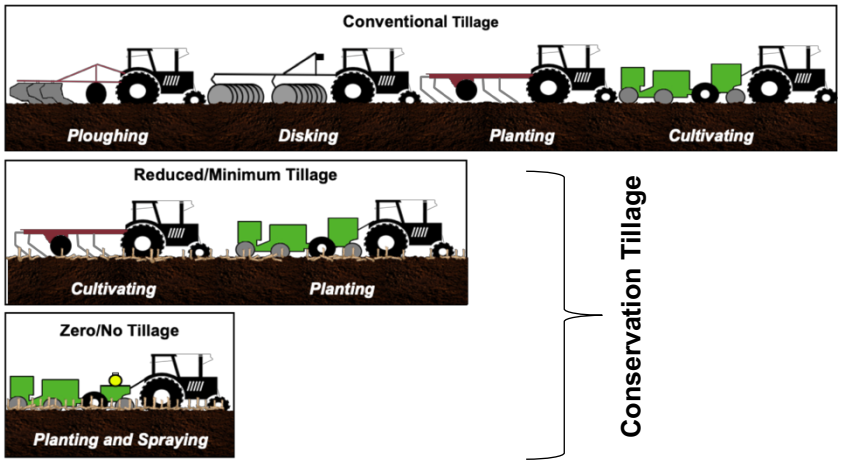


Figure 1.1 The different machinery operations required for conventional, reduced and zero-tillage (figure adapted from FAO, 2003).

Zero-tillage or direct drilling, where seeds are sown directly into soil (Fig. 1.1) that has not been disturbed or inverted since the harvest of the previous crop (Eurostat, 2013b) is the main focus of this review. This practice is one of the most promising strategies for minimising soil erosion (Montgomery, 2007), increasing biological activity (Green et al., 2007) and promoting improved aggregate stability (Song et al., 2016). However, the extent of conversion to zero-tillage varies geographically, and there is conflicting evidence about the

effect of such a conversion on carbon storage and greenhouse gas (GHG) emissions.

1.2 Benefits and limitations of zero-tillage

There is extensive published evidence that zero-tillage is beneficial for the functioning and quality of the soil, in many, though not all, agricultural settings (Table 1.1).

Table 1.1 A summary of the benefits and potential limitations associated with zero-tillage management.

Benefits	Potential limitations
Soil erosion reduced as crop residue is maintained on the soil surface and no mechanical disaggregation (Miura et al., 2008).	No crop residue is mixed throughout the soil profile (Feng and Balkcom, 2017).
Soil moisture conservation through increased crop residue (Derpsch et al., 2010).	Lack of disturbance may cause the bulk density to increase (Salem et al., 2015).
Minimum fuel and labour costs (average fuel savings range from 50 – 80% compared to conventional tillage) (Triplett and Dick, 2008).	In heavy clay soils, increased moisture may lead to waterlogging and slow soil warming (Gardner et al., 1992).
Additional organic carbon in the surface layer (Baker et al., 2007).	Problems with residual plough pans (Peigné et al., 2007).
Stones are not brought to the surface (Soane et al., 2012).	Increased variability of crop yields (Pittelkow et al., 2015).
Increased soil biological activity, especially of earthworms (Staley, 1999).	Increased dependence on herbicides and pesticides (Keil et al., 2015).
	A high initial investment cost for zero-till machinery (Laukkanen and Nauges, 2011).

Zero-tillage is theoretically applicable to most farmland globally, but uptake is still relatively small, ca. 7.9% of global cropland area (Derpsch et al., 2010). The main barriers to widespread adoption include steep learning curves and the availability and investment costs of specialised zero-till machinery (Derpsch et al., 2010). There are conflicting reports about the use of herbicides under zero-tillage, with some farmers reporting an increase. Romaneckas et al., (2009) observed a significantly higher number of weeds present in zero compared to conventionally tilled soils, with the number of weed species increasing from 22 to 26 in sugar beet. Ambani and Kapoor (2016) showed that, when growing wheat, conventional tillage reduced the number of weeds by 12 species, from 38. Others suggest that the application of zero-tillage does not increase the use of herbicides compared to conventional farming, and the emergence of black-grass becomes less prevalent with time under zero-tillage (Godwin, 2014).

1.3 Global adoption of zero-tillage

In general statistics on the area of arable land under zero-tillage are not officially reported, but the adoption of zero-tillage is gathering momentum, with zero-till cropland expanding at an estimated 10 M ha per year, from 111 M ha in 2009 to 157 M ha in 2014 (Derpsch et al., 2010; Kassam et al., 2015). However, the extent of conversion to zero-tillage varies geographically (Fig. 1.2).

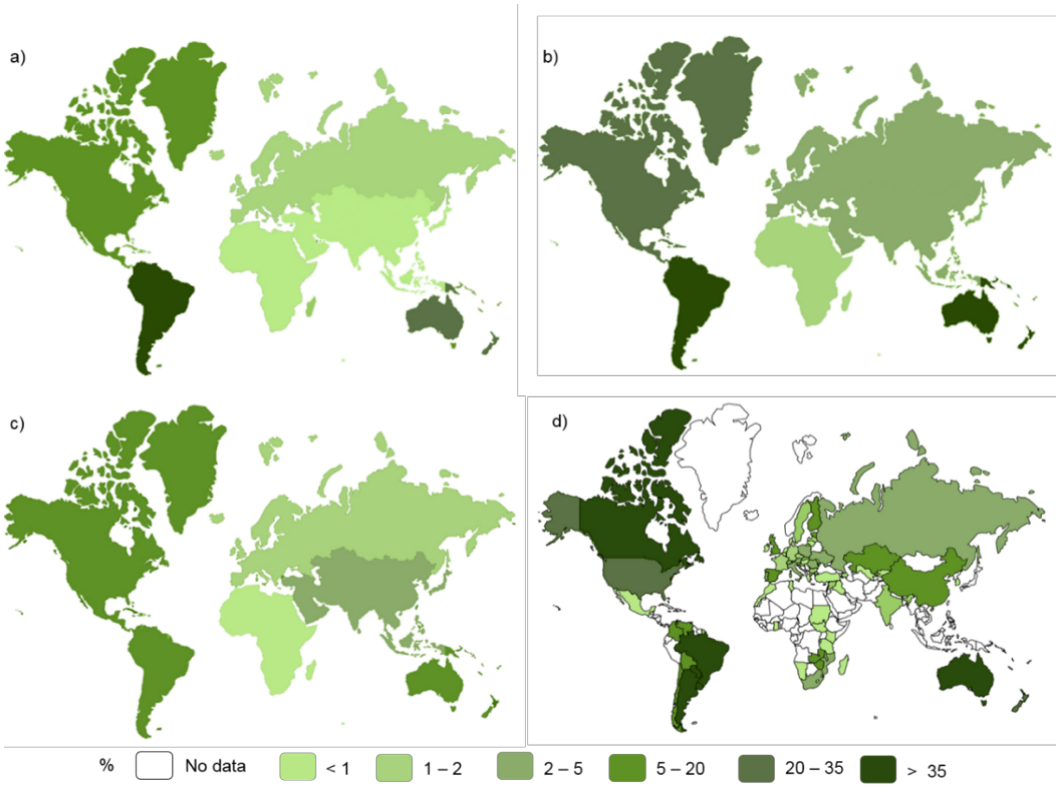


Figure 1.2 Percentage of arable land under zero-tillage (Europe) and zero & minimum tillage (Asia, Africa, Oceania, North and South America). (a) 2008, (b) 2014, (c) change between 2008 and 2014 and (d) national percentages of arable land under zero and minimum tillage in 2014. Dataset and sources are shown in Table A1 in Appendix A.

1.3.1 Adoption in Europe and Asia

Arable land under zero-tillage in Europe increased by 1.3% from 2008 to 2014 (Fig. 1.2a-c), with the latest data suggesting 3.4% of arable land is currently under zero-tillage (Fig. 1.2b). From 1999, the European Conservation Agriculture Federation (ECAF) has been encouraging conversion to zero-tillage, with increased adoption evident throughout the continent (ECAF, 2005) (Fig. 1.2). In England, results of a postal questionnaire indicated in 2016 that less than 1% of farmers adopted zero-tillage on all their available arable land, but 17% of farmers applied zero-tillage to at least one field in their holdings (Alskaf et al.,

2019). In 2008, zero-tillage was reportedly practised in two countries in Asia, increasing to 11 in 2014 (Kassam et al., 2015). In Asia, the largest increase was in Kazakhstan, where the adoption of zero-tillage increased by 7% (Fig. 1.2d). The World Bank (2013) estimated that conversion to zero-tillage in Kazakhstan had increased yields by 30–40% as a result of the crop stubble trapping snow and increasing soil moisture when the weather warmed.

1.3.2 Adoption in Africa and Oceania

In Africa, the adoption of zero-tillage was reported in 14 countries, with the most significant proportional increases (2008 – 2014) in Zimbabwe, Zambia and South Africa (8.3, 5.3 and 2.9% respectively). Reported levels are still relatively small as the capacity for large-scale adoption is still weak (FAO, 2008) and farmers often use crop residues for fuel and animal feed (Giller et al., 2009). Other concerns from African farmers include an initial loss of crop yield, changes in labour requirements and difficulties caused by weed control, given the costs of herbicides. In Oceania, New Zealand was amid the original countries in the world to implement and advance zero-tillage systems, with 28% of its cropland under zero-tillage as of 2014 (Fig. 1.2b and 1.2d). Due to water, time and fuel savings associated with zero-tillage, adoption is projected to increase throughout Oceania, particularly in those Australian states with current smaller rates of adoption such as New South Wales and Queensland (Derpsch et al., 2010).

1.3.3 Adoption in North and South America

In 1972, 1.3 million ha of cropland was under zero-tillage in the United States of America, compared to 35 million ha in 2014 (Fig. 1.2b and 1.2d), an average

annual growth of 8.0%. The initial adoption of zero-tillage in the 1950s was slow as farmers required better planting equipment, cheaper herbicides, and more considerable experience and knowledge, but uptake began to increase in the 1980s (Islam and Reeder, 2014). Globally, the most substantial adoption has been in South America, with 49% of cropland under zero-tillage (Fig. 1.2b), with the largest uptake in Argentina, with 74% of arable land in zero-tillage. (Fig. 1.2d). The Argentine Association of Direct Sowing Procedures (AAPRESID), formed in 1986, contributed to the development and spread of zero-tillage by hosting annual conferences for knowledge dissemination which is visited by over 2,000 farmers each year. In Brazil, the adoption of zero-tillage increased from one million ha in 1990 to 23.6 million in 2005; during this period, estimated soil erosion declined from 3.4-8.0 t/ha in conventional tillage compared to 0.4 t/ha in zero-tillage (Derpsch, 2008).

1.4 Crop yield

The uncertainty surrounding the impact of zero-tillage on crop yield is an important reason why farmers are cautious about adopting this management. There is no consensus as to whether yields are maintained, increase or decrease under zero-till. This is of critical importance given concerns about global food security, particularly as global food demand, is expected to rise by 35% by 2030 (FAO, 2017). Any change in agricultural management that could negatively impact crop yield would have severe global consequences.

Yield responses to zero-tillage vary widely depending on climate (Fig. 1.3). Some studies suggest that yields do increase when in zero-tillage in water-limited conditions (Farooq et al., 2011). However, some studies have

demonstrated that zero-tillage can reduce crop yield due to the potential for soil waterlogging (as a result from crop residue increasing water retention) in colder climates which can inhibit crop establishment and increase compaction, in turn limiting root growth (Ogle et al., 2012). Increasing waterlogging can also alter soil fertility and may lead to soils with nutrient deficiencies (Ahmed et al., 2013).

A global meta-analysis by Pittelkow et al., (2015) of 520 observations in the tropics (latitude of 0–20 degrees) showed a decline of crop yield by 15.1% under zero-till. This is particularly concerning as zero-till is a common agricultural management, on 40, 16, 12 and 8% of cropland in Brazil, Bolivia, Venezuela, and Colombia, respectively. Similarly, in the sub-tropics (latitude of 20–30 degrees) crop yield decreased by 6% under zero-tillage compared to conventional tillage from 559 observations. The smallest decrease was found at temperate latitudes (33–66 degrees), where yield declined by an average of 3.4% across 4842 observations.

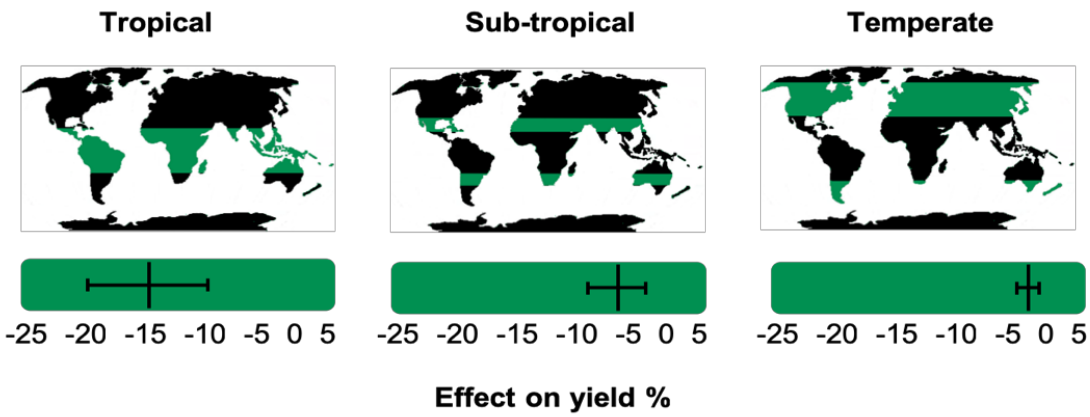


Figure 1.3 Yield impacts of zero-tillage relative to conventional tillage in tropical, sub-tropical and temperate latitudes (figure created from Pittelkow et al., 2015 database). Error bars represent 95% confidence intervals.

Yield responses also vary widely depending on crop type. The meta-analysis by Pittelkow et al., (2015) demonstrated, across all observations (6005), crop yield decreased by 5.1% (Fig. 1.4). The largest decrease was found for root crops, as surface consolidation can be detrimental to their establishment and prevent adequate drainage and soil aeration. It is generally perceived that deep conventional tillage is necessary before seedbed preparation to ensure a loose structured soil which is optimal for tuber formation and quality (Drakopoulos et al., 2016). However, studies carried out by Carter et al., (2011) and Hou and Li (2018) show that zero-tillage does not negatively affect root crop yield, due to lower costs of production and improved soil protection, and in semiarid climates may even increase yield compared to conventionally tilled soils.

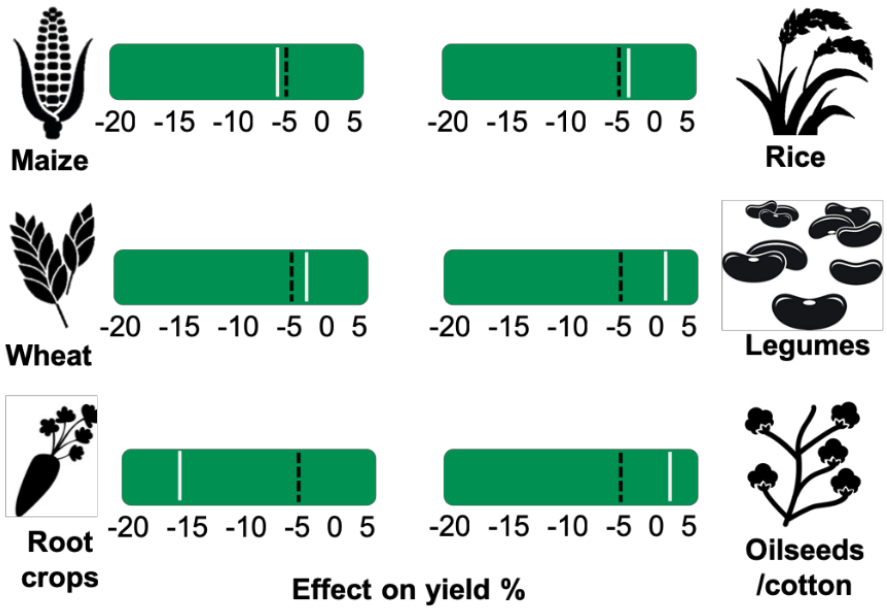


Figure 1.4 Global yield impacts of zero-tillage relative to conventional tillage in different crop categories (figure created from Pittelkow et al., 2015 database). The black dotted line indicates the global average across all crops and the white line represents the average for each crop category.

Crucially, the most extensive zero-tillage adoption (Fig. 1.2) has occurred in countries focusing on grain crop production, with North America and China being the largest producers of wheat (FAO, 2019), and North America, Brazil, and Argentina of maize (FAO, 2018). Several meta-analyses show yields for grain crops are only moderately affected (Knapp and van der Heijden, 2018; Pittelkow et al., 2015; Wang et al., 2018) by the adoption of zero-tillage. In long-term experiments (>19 years), on rain-fed production in semiarid climates, it has been observed that zero-tillage enhanced grain yields over time with an increase in soil water content. The study also highlights the importance of the additional storage of water in deep soil layers in maintaining or increasing crop yield in zero-tillage systems (Lampurlanés et al., 2016).

A global meta-analysis by Knapp and van der Heijden (2018) highlighted that most studies centre around plant yield and neglect temporal yield stability or the variability and reliability of production across years under zero-tillage management. They found, when including the temporal stability in the analysis (and excluding any studies where zero-tillage had been implemented for four or fewer years), that zero-tillage crop yield did not differ significantly from those of conventional tillage, demonstrating that a transition to zero-tillage does not affect yield stability. Similarly, Mangalassery et al., (2015) collated yield responses from 61 datasets which made paired comparisons between zero and conventional tillage systems. They concluded that the difference in crop yield between the two approaches was negligible, where 53% of publications reported an increase in crop yield with zero-tillage, and 47% reported greater yields under conventional management. However, they also note, in soils that had been in zero-tillage for ten years or more, soybean yield increased by 7.7%. In summary,

several meta-analyses have suggested the adverse effects of zero-tillage on crop yield can be counterbalanced in the long-term (>10 years), following the development of an enhanced and regenerated soil structure, which will support higher crop yields in the future.

1.5 Soil structure

Soil structure is "the arrangement of the solid parts of the soil and the pore spaces located between them" (Dexter, 1988). The structure of soil controls several of its properties: physical (air porosity, water availability, hydraulic conductivity, and gas diffusivity), biological (activity and location of micro-organisms) and chemical (ion exchange capacity, redox, and pH). These, in turn, determine the soil quality and crop yield (Bertrand et al., 2015; Czarnes et al., 2000; Kutílek, 2004). Soils with a "good structure" have numerous pores between aggregates with efficient root growth and much biological activity evidenced, for example, by earthworm burrows and casts. Soils with degraded structure have few pores and fissures and are commonly compacted. This limits biological activity and root growth, reducing nutrient uptake by crops and making them more susceptible to pests and diseases (Finch et al., 2014). A change in soil management can degrade or improve soil structure (Bronick and Lal, 2005) and tillage systems modify soil properties depending on factors such as soil type and texture, climate and previous land use (Ferreras et al., 2000).

1.5.1 Quantification of soil structure

Soil structure can be studied at scales from microns — particles and microaggregates (micromorphology) to metres and kilometres (macromorphology) (Peng et al., 2015). Soil macromorphology is studied in the

field through observation, description, and interpretation of a two-dimensional vertical cut through the soil profile. Conventionally, soil micromorphology has been studied by the examination of thin sections under the microscope. Soil structure has often been evaluated quantitatively by the measurement of functional properties which are strongly influenced by the structure and so serves as proxies for it. These include hydraulic conductivity, water retention curves, infiltration and gas diffusion rates (Zhou et al., 2009).

Structural differences between zero- and conventionally-tilled soils have been observed in thin sections. Pagliai et al., (1984) demonstrated that conventionally tilled soils had a greater porosity which was represented by a few large and very irregular elongated pores (Fig. 1.5a). However, thin sections of soils under zero-tillage for 18 years showed more abundant root remains, a greater number of pores, and a large proportion of regular elongated pores (Fig. 1.5b). This led the conclusion that soil conditions are more favourable for root growth under zero-tillage.

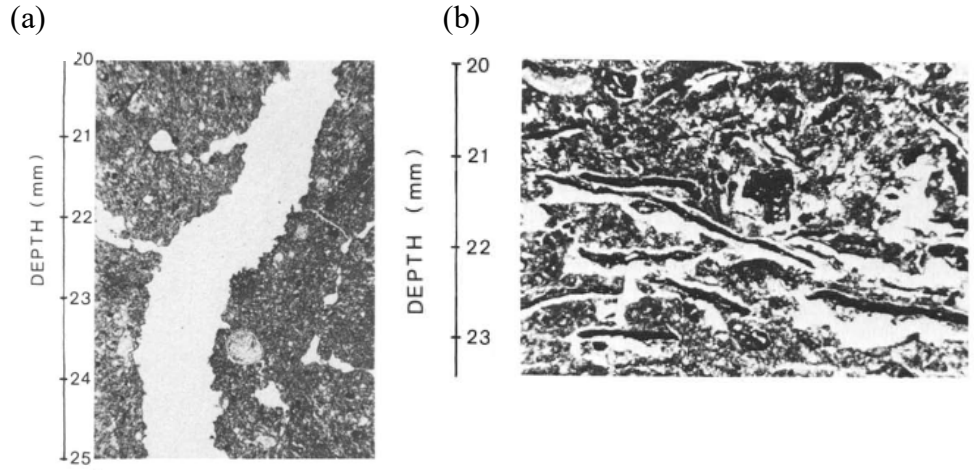


Figure 1.5 Vertical thin sections of soil samples from (a) conventionally tilled soils where a greater proportion of total porosity was represented by few large and irregular elongated pores, and (b) zero-tilled soils where there was evidence

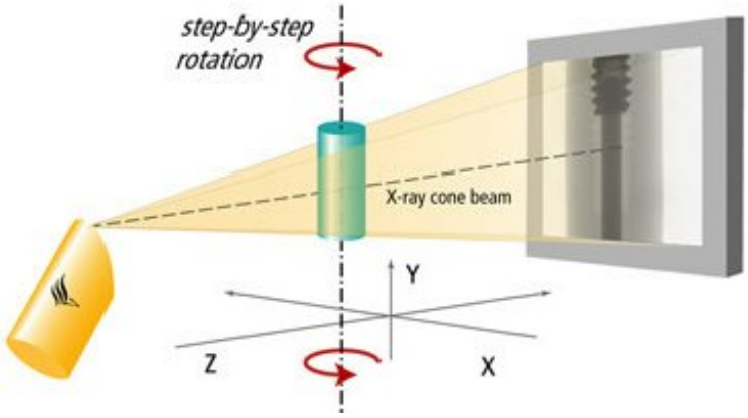
of abundant root remains in the smaller planar pores. Figures from Pagliai et al., (1984).

Optical microscopy, although inexpensive, has several disadvantages. The preparation of sections, which includes polishing the surfaces and removing water from the sample by methods of air oven drying and freeze-drying, disturbs the in situ structure of the soil (ASTM, 1970; Gee and Or, 2002).

Cassel and Whitbeck (1994) posed the question, "would there ever be a method to allow us… to look inside a soil system to observe individual pores, how they vary in size, how regular or irregular they are, how they are arranged and interconnected, which ones are filled with water, and which ones are actively involved in the water and solute transport processes?". In recent years, non-invasive methods that allow soil properties to be investigated without disturbance of their natural architecture and allow 3-dimensional visualisations of internal soil structure have become increasingly powerful. These methods include but are not limited to; X-ray Computed Tomography (CT), nuclear magnetic resonance, and gamma-ray tomography.

The development of X-ray CT has been credited to Godfrey Hounsfield and was originally applied to medical imaging in 1973. The first application in soil science was in 1982, when Petrovic et al., reported a linear relationship between soil bulk density and X-ray attenuation. Subsequently, Hainsworth and Aylmore (1983) utilised X-ray CT to evaluate root-related water absorption processes. X-ray CT scanners consist of three components, (i) an X-ray source, (ii) a sample manipulation stage and (iii) a detector (Fig. 1.6). Materials attenuate an

electromagnetic beam by absorption or scattering, and the amount of attenuation, measured by the attenuation coefficient, is a property of the material. X-ray investigations, including CT, examine the structure of an object by visualising the spatial variation of its attenuation coefficient. (Crestana et al., 1985). X-ray CT images, which are obtained through tomographic reconstruction, consist of linear integration of the attenuation coefficients. Important information has been acquired through the application of CT for the detailed and quantitative measurements of soil structure, especially concerning the spatial distribution of soil pores, sizes and pore networks (Taina et al., 2008).



(i) X-ray source (ii) Sample (iii) Detector

Figure 1.6 A schematic representation of the different components of X-ray computed tomography (Welkenhuyzen et al., 2010).

1.5.2 Studies of tillage systems by X-ray Computed Tomography

Zero-tillage significantly changes the physical properties of soils previously tilled conventionally, especially within the upper few centimetres (Palm et al., 2014). Using X-ray CT, studies have quantified pore structure and networks at fine resolutions (from mm to µm scales), which is necessary to predict fluid flow

properties and evaluate interactions between roots and soil matrices (Gantzer and Anderson, 2002; Taina et al., 2008).

Soil porosity (Ø) is the fraction of the total soil volume that is accounted for by the pore space and is a crucial property which controls the movement of air and water throughout a soil profile (Nimmo, 2005). Mangalassery et al., (2014) reported extensive differences in porosity between zero- and conventionally-tilled soil, with zero-tilled soil porosity measured at 14% compared to 21% in conventionally tilled soils, in the upper 10 cm (assessed by X-ray micro CT- at a resolution of 64 μm) (Fig. 1.7). Most studies on the subject report that conventional tillage increases porosity in the short-term, but that mechanical forces breaking apart large soil clods reduce overall soil aggregation in the longer term (Bronick and Lal, 2005). The increase in surface (0–10 cm) porosity of conventionally tilled soils results in easily erodible soil particles (Fig. 1.8), and erosion rates from conventionally tilled soils are typically 1–2 orders of magnitude greater than rates of soil production. In contrast, partially due to a reduction in soil surface porosity, zero-tilled systems produces erosion rates similar to soil formation rates and therefore, could be a foundation for sustainable agriculture (Montgomery, 2007).

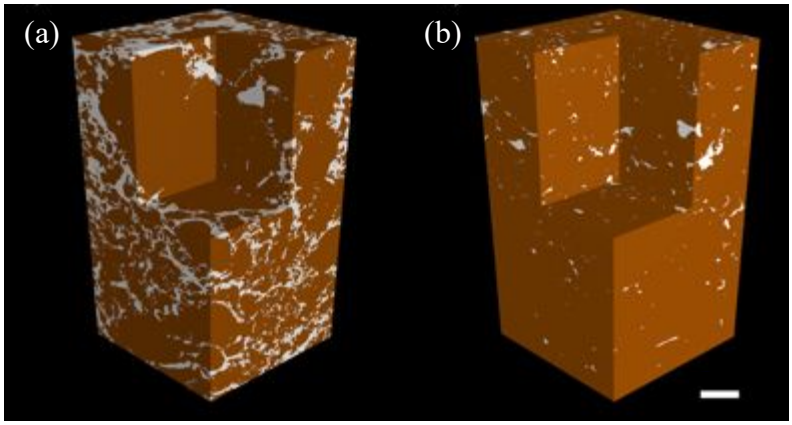


Figure 1.7 Example of (a) conventional and (b) zero-tilled soils. Thresholded 3D image highlighting 'solid' soil in brown and 'pore' space in white. Scale bar = 10 mm (Images from Mangalassery et al., 2014).

Soil pores can be categorised as ultra-micropores (<5 μm), micropores (5–30 μm), mesopores (30–75 μm) and macropores (>75 μm) (Lugato et al., 2009). Soil macropores are naturally occurring continuous apertures, such as worm burrows, root channels or inter-aggregate pores and they create a central pathway for facilitating the flow and transport of water, nutrients and air in soil (Angers and Caron, 1998; Mori et al., 2014). VandenBygaart et al., (2011) observed an initial decline in the number of macropores after the adoption of zero-till adoption, which may contribute to why crop yields tend to decline compared to conventionally tilled soils in the first few years after implementation. However, after four years of zero-till management, the proportion of macropores significantly increased. In agreement, Galdos et al., (2019) observed in a long-term zero-tillage experiment (~30 years), that macroporosity was 5% greater in zero-tilled compared to conventionally tilled systems. Further analysis of pore connectivity showed that the pores under zero-tillage were significantly more connected to the surface than the soil under conventional tillage (Fig. 1.8). These studies indicate long-term adoption of

zero-tillage increases connectivity which has positive implications for water storage, nutrient cycling, soil gas fluxes and root development (Fig. 1.8).

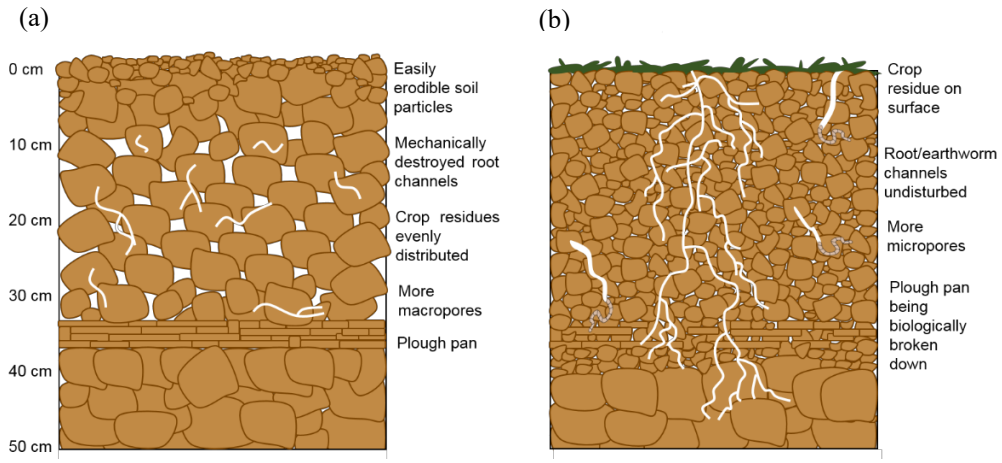


Figure 1.8 A schematic of soil profiles under (a) conventional tillage and (b) long term zero-tillage (right). The entire soil profile is modified under long-term zero-tillage. The surface will be covered in crop residue and the soil is perforated with thousands of micropores created by roots, surface and deep-dwelling earthworms. Old root and earthworm channels leave continuous pores from the surface deep into the subsoil. (Figure adapted from Dorel et al., 2010).

These differences in soil structure between the two managements can be partially attributed to a greater density of earthworms in zero-till systems (Anderson, 1987; Briones and Schmidt, 2017; Peigné et al., 2009). Earthworms are a substantial component of the soil macrofauna and are central for soil fertility. Their burrowing activities improve soil aeration and infiltration of water, and under zero-tillage, the number of biopores (considered macropores) created by earthworms increases throughout the soil profile as their channels are not mechanically destroyed as in conventional tillage systems (Bertrand et al., 2015).

The extent of aggregation and aggregate stability also plays a role in the differences observed in soil structure between the two managements. Soil aggregates are the arrangement of soil particles and are the primary 'cells' of soil structure, playing a significant role in minimising soil erosion and improving soil carbon sequestration and fertility (Bronick and Lal, 2011; Six et al., 2004).

Soil aggregates can be classed as microaggregates (53–250 μm) and macroaggregates (250–2000 μm) (Six et al., 2000). Conventional tillage frequently exposes aggregates to physical disruption, breaking down larger macroaggregates into fine soil particles which can easily be eroded by either rain or wind (Pimentel and Burgess, 2013). Mechanical disturbance also makes the organic matter more accessible to microorganisms, stimulating oxidation and loss of organic matter from the soil profile (Zhou et al., 2009). Fernández et al., (2010) showed in a long-term study (14 years) that macroaggregates increased by 25, 38 and 17% in 0–6, 6–12 and 12–18 cm, respectively under zero-tillage compared to conventional tillage. Sapkota et al., (2012) showed that the aggregate stability index (the ability of soil aggregates to resist disintegration when disruptive forces associated with tillage and water or wind erosion are applied) was three and fourfold larger under zero-tilled system at both 0–5 and 5–10 cm compared to conventionally tilled system.

By implementing zero-tillage, the importance of natural regeneration processes in maintaining a "good soil structure" increases (well-structured soils are usually crumbly and friable and have a wide range of pore space to allow water and air

movement) (Ramesh et al., 2019). Therefore, managing the conversion from conventional to zero-tillage as well as the control of the compaction risk in an established zero-tillage system needs special attention to ensure the sustainability of a zero-tillage system.

1.6 Carbon storage

Carbon stocks in agricultural soils are determined by the balance of inputs, such as crop residues and organic amendments, and losses, through organic matter decomposition (Bhogal et al., 2018). Conventional agricultural practices accelerate the loss of soil organic carbon by increasing the oxygen concentration in the soil profile, destroying soil aggregates and exposing organic carbon for mineralisation (Liu et al., 2006).

Zero-till systems have been proposed as potential management to increase soil organic carbon through several mechanisms. Firstly, to slow macroaggregate turnover allowing fine intra-aggregate particulate organic carbon (iPOM C) to be stabilised (Six et al., 2000). Secondly, through an increase in the proportion of microaggregates formed within macroaggregates, which is crucial for the long-term sequestration of organic carbon. Microaggregates have a better capacity to protect organic carbon against decomposition compared to macroaggregates (Coleman et al., 2007; Kumar et al., 2013). Thirdly, through an increase in earthworm populations associated with minimal disturbance systems. Zero-tillage soils tend to have a larger proportion of anecic (deep borrowing) earthworms, as burrows are destroyed and populations killed by conventional tillage (Jordan et al., 1997). These earthworms create deep vertical burrows that are connected to the soil surface (Peigné et al., 2009) and they

primarily feed on surface organic residues, which they often pull into their burrows (Shipitalo and Le Bayon, 2004). However, not all of the underlying mechanisms in carbon sequestration or reduction are entirely understood. Baker et al., (2007) and Powlson et al., (2016) highlight several limitations of studies (mainly the depth of measurements and calculations of stocks) which may have led to an overestimation of the potential of zero-tillage to sequester more carbon compared to conventionally tilled soils.

It is contentious whether more carbon is sequestered in soil under zero-till than under conventional tillage. Conventional tillage is estimated to have reduced soil organic carbon stocks by two-thirds from pre-deforestation levels (Lal, 2003). A review by Govaerts et al., (2009) synthesised results from 78 studies — in 9% the soil carbon stock was smaller in zero-tilled compared to conventionally tilled soils. However, in 51% of comparisons soil carbon stocks were significantly larger in zero-tilled soils and the remaining 40% there were no significant differences between the two managements. In the studies which showed a smaller carbon stock in zero-tilled compared to conventionally tilled soils, 70% of these only measured the top 30 cm, and the majority of the studies in tropical areas were excluded from the analysis as sampling did not extend deeper than 20 cm.

1.6.1 Depth of soil sampling

There is strong evidence that zero-tillage management can increase organic carbon in the surface 0-20 cm layer (Angers and Eriksen-Hamel, 2008; Baker et al., 2007; Luo et al., 2010) but stocks in this surface layer do not represent all the carbon sequestered in the soil. Baker et al., (2007) noted that very few studies

sampled deeper than 30 cm. For several plants, an estimated 30–50% of the carbon fixed in photosynthesis is originally translocated below-ground. Some are used for essential growth of the rooting structure or for autotrophic respiration, and some are lost to the surrounding soil in organic forms which is known as rhizodeposition. Studies also shows that below-ground plant carbon is a main source for conversion into more stable recalcitrant forms of soil organic carbon (Baker et al., 2007). Shallow sampling (<30 cm) could be adequate for comparative assessments of sequestration of organic carbon between contrasting tillage managements if the growth of roots down through the soil profile was the same in each management. However, there is evidence to suggest that this is not the case (Shen and Tanner, 2010). Zero-tilled systems generally have a lower surface soil temperature due to the insulating effects of surface crop residue and increased moisture retention (Fabrizzi et al., 2005). Soil temperature is one of the key factors controlling root growth, with 5-fold differences in root growth over temperatures– of 45°C (Baker et al., 2007). Root differences between zero and conventional tillage have seldom been studied. Of the studies currently available, Taboada and Alvarez, (2008) identified a reduction in maize root abundance of 40-80% in conventional compared to zero-tilled sites. Another study by Alam et al., (2014) identified in the 0-15 cm interval that the root mass density of wheat was significantly greater in the zero-tilled soils (9.99 mg cm^{-3}) compared to the conventionally tilled soils (8.72 mg cm^{-3}), but the reverse was true deeper in the soil profile (30-45 cm).

A recent metanalysis by Haddaway et al., (2017) showed that zero-tillage significantly increased soil organic carbon stock in the top 30 cm compared to conventionally tilled soil. However, in the studies which took into account soil

layers deeper than 30 cm (66 studies), there was no overall significant difference in soil organic carbon between zero and conventionally tilled soils, highlighting the importance of considering depth when assessing impacts of zero-tillage on soil carbon storage. Indeed, Luo et al., (2010) found that conversion from conventional to zero-tillage resulted in significant topsoil soil organic carbon enrichment but did not increase the total soil organic carbon stock in the whole profile.

Nevertheless, some studies show an increase in soil organic carbon when taking into account soil layers deeper than 30 cm. For example, Boddey et al., (2010) found, in three long-term experiments (15–26 years) in the subtropical region of South Brazil, that soil organic carbon significantly increased by 5–8 Mg ha^{-1} in comparison with conventional tillage when sampling to a depth of 100 cm.

1.6.2 Equivalent soil mass vs fixed depth procedure

Studies tend to quantify soil organic carbon stocks to a fixed depth as the product of soil bulk density, depth, and concentration (Wendt and Hauser, 2013). This method has been designated as "good practice" by the Intergovernmental Panel on Climate Change (IPCC, 2014). However, this method only considers soil organic carbon content at a fixed depth interval, so it is not appropriate for comparisons between management strategies that affect bulk density because corresponding soil material is not compared (Ellert and Bettany, 1995) (Fig. 1.9), therefore potentially overestimating soil carbon sequestration in zero-tillage. Palm et al., (2014) showed that an apparent increase in soil organic carbon stock, when calculated to a fixed depth, can be reduced by 50% if

recalculated using the equivalent soil mass method. It is therefore important to account for soil mass when assessing impacts of zero-tillage on carbon stocks.

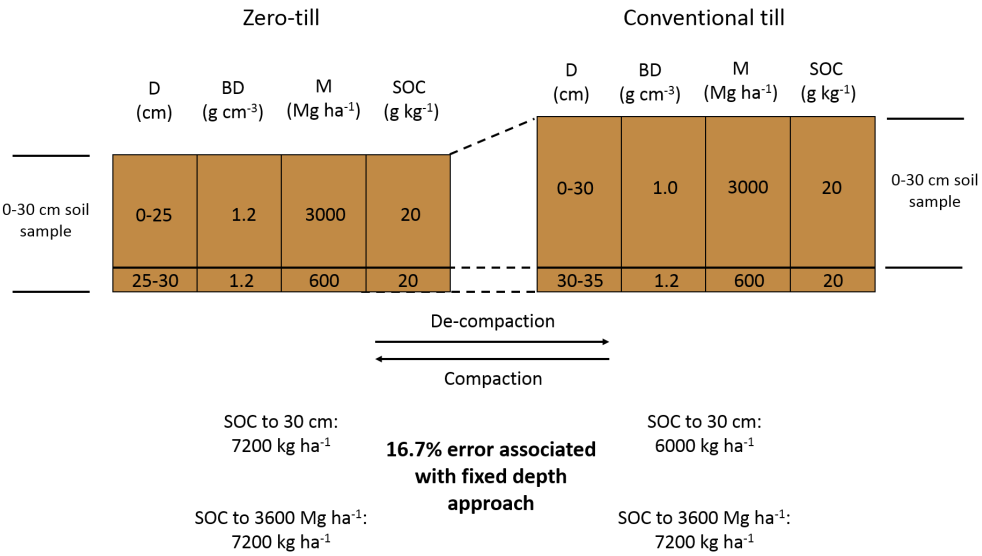


Figure 1.9 The error bias due to quantifying soil organic carbon stocks at fixed depths when different managements result in varying bulk densities. D is depth of the soil profile, BD is the bulk density, M is the soil mass and SOC is the soil organic carbon concentration. Figure adapted from Wendt and Hauser, 2013.

1.7 Greenhouse gas emissions

Agricultural soils are significant sources of GHG emissions, globally contributing one-fifth of CO_2 emissions, one-third of CH_4 emissions and two-thirds of N_2O emissions (EPA, 2014). Zero-tillage is one amongst many different mitigation possibilities proposed to decrease GHG emissions from agriculture. There is a consensus in the literature that zero-tillage reduces fuel consumption due to fewer trips over a field with farm equipment, which results in a reduction in CO_2 emissions. Estimates from Basch et al., (2012) suggest a 44.2 L ha^{-1} yr^{-1} saving in fuel, which would equate to 133.9 kg CO_2 ha^{-1} yr^{-1} reduction of CO_2 emissions. Smith et al., (1998) suggested that a 100% conversion to zero-tillage in Europe could mitigate all fossil fuel-carbon

emissions from agriculture in Europe. However, the precise effects of zero-tillage on soil GHG emissions remain controversial and results significantly vary among studies. Some findings have shown an extensive decrease in soil CO_2, CH_4 and N_2O emissions with adoption of zero-tillage (Alvarez et al., 2001; La Scala et al., 2006), while others stated a substantial increase (Baggs et al., 2000; IPCC, 2007), or no difference compared to conventionally tilled soils (Ahmad et al., 2009; Drury et al., 2006).

1.7.1 Carbon dioxide

Soil CO_2 fluxes are the second-largest component of the global carbon cycle and play an important role in either exacerbating or mitigating climate change (Reth et al., 2005). Differences in CO_2 emissions between conventional and zero-tillage are likely to result from both short and long-term mechanisms. Evidence for greater CO_2 emissions from soils under conventional tillage has been widely reported in the short-term (Abdalla et al., 2013). Through physically disturbing and inverting the soil, conventional tillage breaks down macroaggregates and exposes the stored organic carbon to microbial processes which result in mineralisation. The amount of carbon lost to the atmosphere in the form of CO_2 is strongly correlated with the intensity of disturbance (Reicosky and Archer, 2007) and soil temperature (Singh and Gupta, 1977). Results from Mangalassery et al., (2014) show 21% greater CO_2 emissions in response to conventional tillage compared to neighbouring zero-tilled soils, which was attributed to the total soil porosity and pore size. In a long-term experiment (43 years), Ussiri and Lal, (2009) reported significantly greater CO_2 fluxes (6.6 ± 0.3 Mg CO_2-C ha^{-1} yr^{-1}) from the conventionally tilled soils than from zero-tilled soils (5.5 6.6 ± 0.3 Mg CO_2-C ha^{-1} yr^{-1}). In this study, the CO_2 fluxes were positively correlated

with the proportion of air-filled porosity in the soil and soil temperatures in the top 20 cm. Similarly, in short term experiments (113 days) the same conclusions have been reported, Chatskikh and Olesen, (2007) stated that CO_2 emissions were 23% smaller under zero-tillage (3.30 t CO_2-C/ha) compared to conventional tillage (4.29 t CO_2-C/ha).

In contrast to this, larger CO_2 emissions from zero- compared to conventionally-tilled soils have been reported. Zero-tilled soils tend to have more available soil moisture due to the increased crop residue content on the soil surface. This can lead to an acceleration in soil CO_2 emissions (Cheng-Fang et al., 2012; Oorts et al., 2007). In addition to soil temperature, water content and soil structure impacts on soil CO_2 emissions, the crop type, and crop rotation may also be important controls due to differences in root biomass and respiration. In a zero-till experiment, Rigon et al., (2017) reported that growing sunn hemp reduced soil CO_2 emissions by 12% compared to pearl millet, which they attributed to differences in the nitrogen content of crop residues and soil moisture.

1.7.2 Nitrous oxide

N_2O has a global warming potential (GWP) 265-298 times that of CO_2 for a 100-year timescale (Barker, 2007). The GWP was developed to allow comparisons of the global warming impacts of different gases. “It is a measure of how much energy the emissions of one ton of a gas will absorb over a given period of time, relative to the emissions of one ton of CO_2” (IPCC, 2007). It is widely accepted that agriculture is the primary source of anthropogenic N_2O emissions (IPCC, 2007).

N_2O emissions from agricultural soils are the result of complex interactions between climatic variables and soil biological, chemical, and physical properties. This has led to different and contrasting results being reported in the literature when assessing the impacts of zero and conventional tillage on emissions (Butterbach-Bahl et al., 2013).

Most studies have shown an increase in N_2O emissions from zero-tilled compared to conventionally tilled soils (Abdalla et al., 2013; Six et al., 2002). N_2O in the soil is produced through two microbially-mediated processes: nitrification and denitrification. Under aerobic conditions, nitrification (the oxidation of ammonium to nitrate) occurs, while in anaerobic conditions, the process of denitrification (reduction of nitrate to nitrous oxide and nitrogen) takes place (Bateman and Baggs, 2005). The relative contribution of these two processes to the formation of N_2O depends on changes in soil aeration and water-filled pore space (WFPS- the ration of the volumetric water content of the soils to its volumetric water content when saturated), which can be influenced by tillage practices (Palm et al., 2014). Generally, when WFPS is below 40%, nitrification is the predominant source of N_2O while the contribution from denitrification increases when WFPS increases to 65–75%. Zero-till systems are generally associated with greater soil bulk density compared to conventional practices; therefore, WFPS is greater in anaerobic conditions and denitrification occurs sooner at the same water content with zero-tilled soils (Palm et al., 2014).

A meta-analysis conducted by Huang et al., (2018), provided 299 comparisons of N_2O emissions from zero and conventionally tilled soils from 20 different countries. Across all observations, the average N_2O flux increased by 10.4%

from zero-tilled soils. They reported no significant difference in N_2O emissions between the tillage systems when comparing different climates (humid and dry) and pH (>6.6, 6.6≤ pH≥ 7.3, >7.3). Nitrogen fertilization and crop species gave the largest differences in N_2O emissions between zero and conventional tillage, with zero-tillage exhibiting 16.7 and 15.2% greater N_2O emissions with a high fertilizer rate (>200 kg N ha^{-1} yr^{-1}) and with wheat production.

N_2O production may have antagonistic responses to zero-tillage and considering its large GWP compared to CO_2 and CH_4, this could potentially result in the growth of zero-tillage exacerbating climate change. This is therefore important to assess the impacts of zero-tillage on N_2O emissions for different regions and management systems.

1.7.3 Methane

Most studies have reported either no significant difference in CH_4 emissions following the adoption of zero-tillage or a small decrease compared to conventionally tilled soils (Bavin et al., 2009; Li et al., 2011). Reduced CH_4 emissions might be due to a combination of increased methanotrophic populations (Hütsch, 2001) and increased surface bulk density. An increase in surface bulk density prevents the diffusion of CH_4, thereby enhancing the retention, which leads to oxidation by methanotrophs, resulting in decreased CH_4 emissions. The main biological sink of atmospheric CH_4 is oxidation by methanotrophic bacteria in aerated soils which has been estimated to account for up to 15% of the total atmospheric CH_4 removal (Born et al., 1990).

1.7.4 Global warming potential

True climate change mitigation is only possible if the overall impact of zero-tillage adoption reduces the net GWP when calculated by incorporating the three major biogenic greenhouse gases, CO_2, CH_4 and N_2O (Six et al., 2004). Nevertheless, only a few studies have considered the fluxes of all three major GHGs that are impacted by tillage management (Mangalassery et al., 2015; Piva et al., 2012; Sainju, 2015).

Huang et al., (2018) considered 64 studies, which measured fluxes of all three GHGs, and found that there was no difference in the GWP of emissions from zero-tillage and conventional tillage. However, Six et al., (2004) reported that newly-converted zero-till systems produced emissions with larger GWP than those from conventional tillage, in both humid and dry climates. Under longer-term adoption (>10 years), the net GWP of emissions was significantly reduced in zero-tilled systems in humid climates. Although, whilst this review included 254 measurements for CO_2 emissions, it made observations from only 44 and 5 data points for N_2O and CH_4, respectively.

Mangalassery et al., (2015), in a temperate climate, reported significantly greater N_2O emissions from zero-tilled soils (0.63 ng g^{-1} h^{-1}) compared to conventionally tilled soils (0.36 ng g^{-1} h^{-1}). However, the offset through a substantial reduction in CO_2 emissions from zero-tilled soils resulted in a 31% decrease in GWP from zero-till systems. Similarly, in tropical and subtropical soils, Piva et al., (2012) observed a smaller GWP from zero-tilled soils, principally due to the greater carbon sequestration rate.

In summary, the literature suggests zero-tillage practices reduce GHG emissions in the long-term, but crucially some uncertainty exists as to the length of time after adoption before effects are seen and the different impacts which are shown due to contrasting climates.

1.8 Microbial community and function

Soil micro-organisms play key biochemical functions in the decomposition of organic carbon in the soil and between the soil and the atmosphere. Alterations in soil carbon distribution due to a change in tillage practice are likely to affect both microbial community composition and functions and the resulting GHG fluxes. Several studies have concluded that zero-tillage management increases microbial abundance (biomass) (Balota et al., 2003; Kandeler et al., 1999), microbial diversity, and enzyme activity (Staley, 1999). A study by Wang et al., (2017) indicated that zero-tillage enhanced the stability of the rhizosphere bacterial community and in turn, improved plant production. Similarly, through phospholipid fatty acid (PLFA) analysis Mathew et al., (2012), found that there is a consistently greater abundance of fungi, bacteria, arbuscular mycorrhizal fungi, and actinobacteria in the zero-tilled surface soil, compared to conventionally-tilled soils. However, few studies have examined how contrasting tillage regimes affect the distribution of microbial communities throughout the soil profile, specifically at depths greater than 15 cm. This is a significant knowledge gap, because the extent to which zero-tillage could promote the sequestration of carbon, and reduce the GWP of gaseous emissions, will depend in part on microbial community structure and functions throughout the soil profile.

1.9 Objectives and hypotheses

The review of the literature above has highlighted several gaps in our current understanding. Firstly, does zero-tillage reduce the global warming potential from soils, when taking into account the three major biogenic GHGs (CO_2, CH_4, and N_2O)? At the same time, does zero-tillage have the potential to sequester more carbon than conventionally tilled soils, when taking into account the depth of sampling and changes in soil mass? If both statements hold, how long does it take for zero-till soils to alter biophysical and chemical mechanisms which reduce GHG emissions and improve soil quality? Furthermore, what is the combined effect of the above across a wide range of geographical landscapes, against the variabilities imposed by nature and differences in on-farm management?

We have seen that there is a potential to use X-ray CT as a novel approach to gain insight into some of the key soil processes. In particular, this work will consider the effect of the soil porous architecture created by tillage on net GHG emissions and carbon storage. Also, this body of work will go beyond previous studies by undertaking measurements across a variable landscape and use a novel paired sampling design to encompass a wide range of soil textures, crop managements and years since conversion to zero-tillage across two contrasting climatic systems. **The individual chapters addressing these knowledge gaps are detailed below:**

Chapter 2 examines the climate change mitigation capabilities of zero-tillage in comparison to conventional tillage systems based on GHG emissions and carbon storage [based on U.K. soils].

Questions:

i) How does the conversion from conventional to zero-tillage alter the soil physical properties, greenhouse gas balance and potential carbon storage?

ii) What are the fundamental mechanisms driving these changes?

iii) How do responses to conversion to zero-tillage vary temporally?

iv) How do these processes differ over a regional area?

Hypothesises:

i) Zero-tillage will increase carbon stocks on an equivalent soil mass basis through a change in the functional groups of carbon.

ii) There will be a reduction in greenhouse gas emissions compared to conventionally tilled soils through a modification to the soil porous architecture.

iii) The effects of conversion on soil biophysical properties are enhanced over time as longer-term changes are brought about by the slow processes of soil development and regeneration.

iv) These effects will vary over a regional area due to different soil characteristics and climate variations.

Chapter 3 assesses 80 pairs of zero and conventionally tilled fields to determine how long it takes after implementation of zero-tillage for soil quality to change [based on U.K. soils].

Questions:

i) How do greenhouse gas responses vary with temperature under different tillage systems?

ii) How does aggregate stability vary between tillage managements, years in zero-tillage and seasons?

Hypothesises:

i) Soil greenhouse gas emissions will increase with greater temperature.

ii) Recently converted soils will be more sensitive to an increase in temperature, shown through a greater increase in CO_2 emissions from conventionally tilled soils compared to zero-tilled soils .

Chapter 4 explores the impact of long-term zero-tillage on greenhouse gas emissions, carbon content and biophysical properties in tropical soils [based on Brazilian soils].

Questions:

i) How does the conversion from conventional to zero-tillage alter GHG emissions, soil physical properties and microbial activity across a range of tropical soils?

ii) How do these effects vary over time under zero-tillage?

Hypothesises:

i) Long-term zero-tillage will increase shear strength due to reduced disturbance and increase soil porosity due to the development of biopores.

ii) CO_2 fluxes will be greater from conventionally tilled soils due to mechanical disturbance breaking apart aggregates and exposing organic carbon to mineralisation.

iii) N_2O fluxes will be larger from zero-tilled soils due to increased moisture content and substrate availability.

iv) Conventionally tilled soils will be a net source of CH_4 compared to zero-tilled soils which will be a net sink due to increased methanotrophic activity in undisturbed soils.

v) Microbial activity will be greater in long-term zero-tilled soils due to increased organic matter from surface crop residues and reduced soil disturbance.

Chapter 5 focusses on the micro-scale impacts of zero-tillage by assessing the impact of the adoption of long-term zero-tillage on aggregation and aggregate associated carbon in tropical soils [based on Brazilian soils].

Questions:

i) How does the chemical characterisation of organic carbon stored in aggregates differ under zero and conventional tillage?

ii) Is organic carbon stability greater in microaggregates within macroaggregates under zero-tillage compared to conventional tillage?

iii) Do aggregate size distribution and organic carbon lability change over time since conversion to zero-tillage?

Hypothesises:

i) Zero-tilled aggregates will have a greater proportion of "recalcitrant" organic carbon compared to conventionally tilled, with the proportion increasing with duration under zero-tillage.

ii) There will be a greater fraction of microaggregates with macroaggregates in zero-tilled soils.

iii) The minimised disturbance associated with zero-tillage will result in a greater number of macroaggregates, which will increase with the duration of management.

2.0 Zero-tillage could offer a long-term strategy to mitigate climate change

This chapter examined the climate change mitigation capabilities of zero-tillage in comparison to conventional tillage systems based on greenhouse gas emissions and carbon storage. The chapter also assessed how zero and conventional tillage modifies the soil porous architecture, which impacts on water infiltration, greenhouse gas fluxes and microbial activity. This experiment encompassed a unique large-scale field study, based on 80 closely located pairs of zero and conventionally tilled fields, covering an area of 6,504 km^2 in the East Midlands, U.K., where zero-tillage had been implemented for 1–15 years.

This chapter is currently under review as a research letter in ***Nature Climate Change*** and is presented in the paper format required by this journal. Additional information is supplied after the paper.

2.1 Summary

Changes in the management of agricultural soils can affect their role as a source or sink in the global carbon cycle, and the scale and composition of their greenhouse gas emissions[1]. It has been established that zero-tillage has beneficial effects on soil health and it is now practised on 7.9 % of arable land globally (111 Mha)[2], but there is conflicting evidence on the effect of zero-tillage on carbon storage and greenhouse gas emissions. Here we present a study to measure the effect of zero-tillage on both carbon storage and greenhouse gas release at a regional scale, in this case, the East Midlands of the U.K. We examined sites, where zero-tillage had been implemented for different time periods (1–15 years), compared to nearby paired sites under conventional management. We observed a significantly greater net global warming potential under conventional tillage systems (30 % higher than zero-tillage systems) due to changes in greenhouse gas fluxes and provided evidence that the net global warming potential of zero-tillage systems decreased further with time. Simultaneously, in zero-tillage systems, carbon stocks increased with time and changes in organic carbon chemistry indicated an increase in the proportion of sequestered recalcitrant carbon compared to tilled soils. While further research considering seasonal variations is still needed, our work advances our understanding of the merits of zero-tillage. This study measures both the potential of zero-tillage to sequester carbon, while taking into account the three major greenhouse gases, using the same sampling strategy and analysis throughout 160 fields, something review papers cannot guarantee. We show that zero-tillage could play a crucial role in reducing greenhouse gas emissions and at the same time increase soil carbon sequestration.

2.2 Results and discussion

Globally, soils are a significant store of organic carbon (C), holding an estimated 1,550 Gt C to a depth of 1 m^3. Soils are also a significant source of greenhouse gas (GHG) emissions, globally contributing one-fifth of CO_2 emissions, one-third of CH_4 emissions and two-thirds of N_2O emissions[4]. Zero-tillage (where the seed is sown directly into undisturbed soil) is an increasingly popular strategy to minimise soil erosion, increase biological activity and promote greater soil aggregate stability[5,6]. However, the extent to which zero-tillage reduces GHG emissions and increases soil C storage compared to conventional tillage is extensively debated in the literature[7] and represents a critical knowledge gap in the context of climate change mitigation.

Zero-tillage may increase soil C storage by reducing soil aeration and limiting decomposition[8]. Reduced macroaggregate turnover and the inclusion of organic materials within soil aggregates can both increase and stabilise C stored in zero-tilled soils[9] but, previous studies may have overestimated this effect by ignoring differences in the vertical distribution of soil organic carbon (SOC)[7]. Zero-tillage is known to increase the concentration near the soil surface, but there is commonly a more uniform distribution of C over a greater depth in conventionally tilled soils[10]. The current Intergovernmental Panel on Climate Change (IPCC) method considers only the SOC content in a fixed depth interval, so comparisons between management strategies that affect bulk density are biased because corresponding soil material is not compared[11]. To assess the impact of the conversion on soil C stocks, it is essential to compare equivalent soil masses rather than depth intervals[12].

Uncertainty also remains regarding the composition of the stored organic C, as well as GHG emissions. Six et al., highlight that climate change mitigation is only possible if the overall impact of zero-till adoption reduces the net global warming potential (GWP) of GHG fluxes[9]. Fluxes of CO_2, CH_4 and N_2O may have antagonistic responses to zero-tillage, for example, reduced soil CO_2 and CH_4 emissions but increased N_2O emissions[13].

The key questions which we address here are 1) how does the conversion from conventional to zero-tillage alter the GHG balance and potential C storage? 2) what are the fundamental mechanisms driving these changes? 3) how do responses vary according to the length of time under zero-tillage? and 4) how do these processes vary at a regional scale? To address these questions, we conducted a unique field study, based on 80 closely located pairs of zero and conventionally tilled fields, covering an area of 6,504 km^2 in the East Midlands, U.K, where zero-tillage had been implemented for 1–15 years (Table 2.3). In line with questions one and two we hypothesised that i) zero-tillage would increase C stocks on an equivalent mass basis through a change in the functional groups of C. And ii) there would be a reduction in GHG emissions compared to conventionally tilled soils, through a modification of the soil porous architecture. For question three we hypothesise iii) such effects are enhanced over time as longer-term changes are brought about by the slow processes of soil development and for question four, we hypothesise iv) that these effects would vary over a regional area due to different soil properties. In this study, we measured potential GHG fluxes, following incubation under controlled conditions. This approach was adopted to allow for comparison between

management by avoiding confounding temporal effects on gas production *in-situ*.

Contradicting our first hypothesis, the mean C stock, on an equivalent soil mass basis, across all soils under zero-till did not differ significantly from the mean under conventional tillage (114 and 112 Mg ha^{-1}, respectively). However, consistent with our third hypothesis, time since conversion significantly affected C stocks with soils under zero-tillage storing 4 Mg C ha^{-1} more than conventionally tilled soils after 6–10 years, and 6 Mg C ha^{-1} more after 11–15 years. (Fig. 2.1a). The increase in C stock in long-term zero-tilled soils is attributed to the surface layers, 0–10 and 10 –20 cm, there was no significant difference in C content between the deeper layers. Soil functional organic chemistry, as inferred from infrared spectra, was also influenced by duration of zero tillage; after 6–10 years aliphatic functional groups increased while after 11–15 years aromatic and ether groups increased (Fig. 2.1b, Table 2.4).

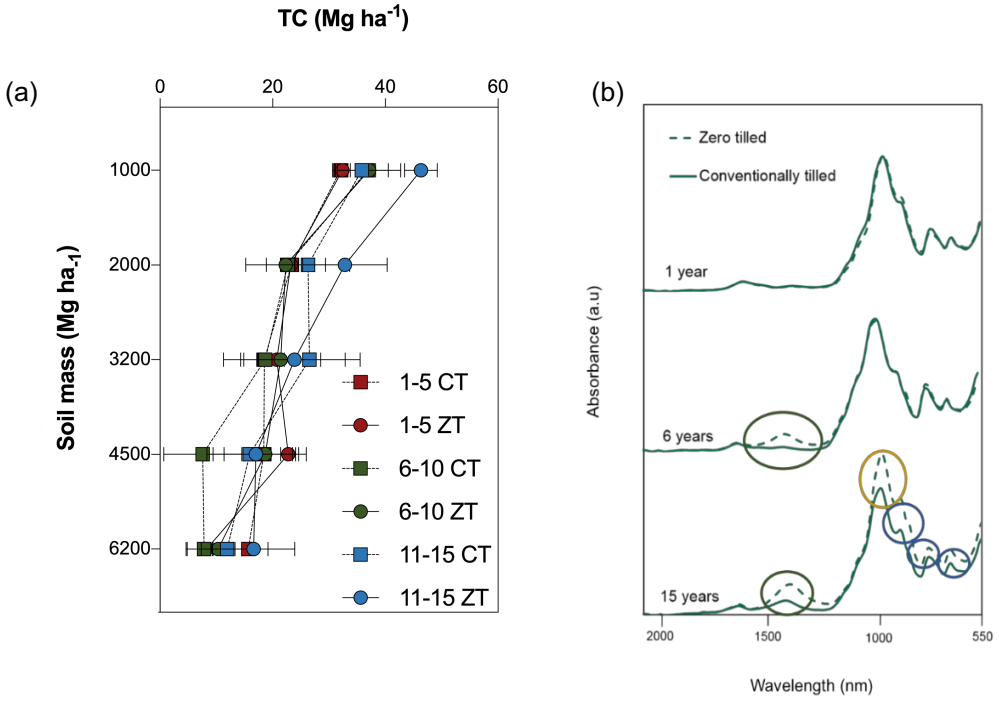


Figure 2.1. (a) Total carbon (TC) stocks shown on an equivalent soil mass basis from paired zero (ZT) and conventionally tilled (CT) soils. Soil mass layers 0–1000, 1000–2000, 2000–3200, 3200–4500, 4500–6200 Mg ha^{-1} correspond roughly to soil depth layers 0–10, 10–20, 20–30, 30–40 and 40–50 cm, respectively. Mean ± one standard error is shown and n=51, 21 and 8 for soils under zero-till management for 1–5, 6–10 and 11–15 years, respectively (b) FTIR spectra to identify organic composition at 0–10 cm for paired observations where soils had been under zero-till management for 1-5, 6-10 and 11-15 years. Green circles illustrate aliphatics, yellow circles for ethers and blue circles for aromatics. For the full FTIR spectra at the different depths, see Fig. 2.8 and Table 2.4.

Conversion to zero tillage altered the distribution of C throughout the soil profile 6–10 years after adoption. The largest increases in soil C occurred at the surface and smaller increases at depth. The greater SOC in zero-tilled surface soils was mainly due to an increase in aliphatics, which are associated with labile organic matter and are depleted following repeated tillage or soil disturbance[14]. Also, a larger proportion of organic C, of intermediate recalcitrance, (peak at 1004 nm, assigned to the ether functional group) was measured in 15-year zero-tilled soils. This C is less susceptible to oxidation on disturbance compared to labile C, and in turn, increases the longevity of stored C. The increase in very recalcitrant aromatics in 15-year zero-tilled soils suggests greater preservation of lignin during decomposition of crop residues, enhanced microbial stabilisation of organic materials[15] or both contributing to further increasing longevity of stored C. Although we have shown substantial shifts on the soil functional organic chemistry with conversion to zero-tillage, it is important to acknowledge that the time for the soil to reach a steady-state

for C storage will vary with climatic conditions, soil type and texture, and the management systems followed[16].

Consistent with our second hypothesis, we found zero-tillage altered the GHG balance which we attribute to modification of the soil porous architecture, an effect that was enhanced over time (in line with our third hypothesis) (Fig. 2.2, Table 2.1). A lack of disturbance under zero-tillage increases the formation of biopores which persist and develop through time by facilitating root growth and undisrupted earthworm activity[17].

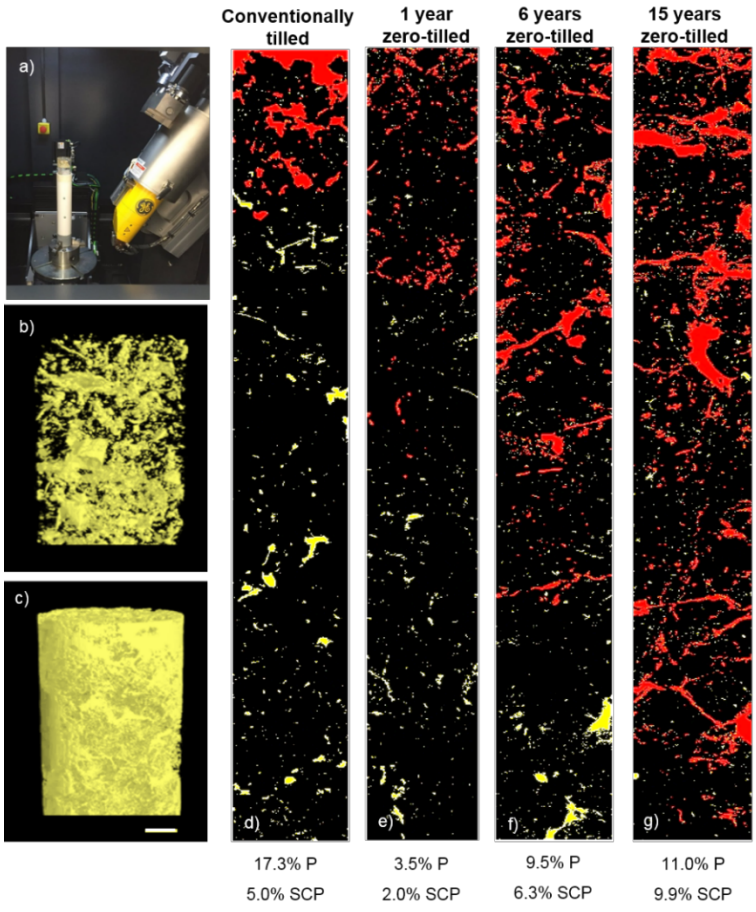


Figure 2.2. (a) Morphological analysis of intact soil cores using X-ray Computed Tomography. 3D images highlighting 'pore space' in yellow in (b) zero-till and (c) conventionally tilled soil cores. (d-g) Images show 'pore space'

in yellow and 'surface connected pores' in red in (d) conventionally tilled, (e) 1 year zero-till, (f) 6 year zero-till and (g) 15-year zero-till soil cores. Where P is the total porosity of the soil core, and SCP is the surface connected porosity of the soil core. Scale bar = 10 mm.

The potential CO_2 flux data (90 to 249 mg m^{-2} h^{-1}) across conventional and zero-tilled soils compare to those reported from previous laboratory incubations from arable soils[16,18,19]. The greater release of CO_2 from conventionally-managed soils (Fig. 2.3a) in our study, highlights the role of mechanical disturbance in the disruption of soil aggregates and exposure of subsurface organic materials for microbial decomposition. We predicted GHG fluxes would be reduced through a modification of the soil porous architecture. We demonstrate that soil porosity initially declines by 13.8 %, after the adoption of zero-tillage (1–5 years) due to consolidation via a lack of disturbance (Fig. 2.2). In turn, soil porosity strongly regulated CO_2 emissions, accounting for 44% of variation with lower emissions from zero-tilled soil (Fig 2.2 & 2.3, Table 2.1, $p<0.001$). Similarly, Mangalassery et al., demonstrated soil porosity decreased by 4.0% following conversion to zero-tillage driving lower CO_2 fluxes[16].

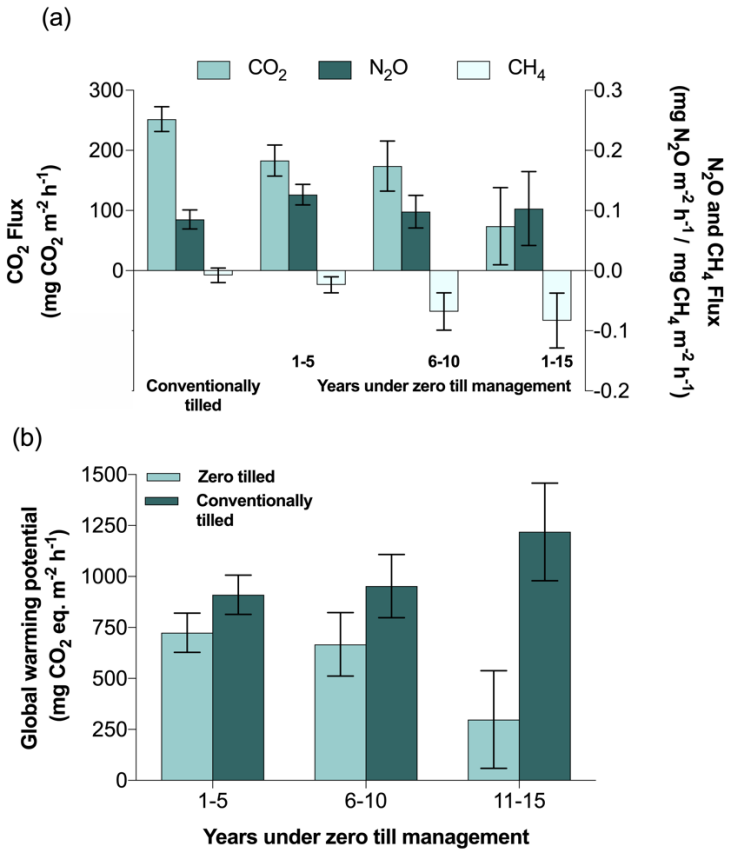


Figure 2.3. (a) CO_2, CH_4 and N_2O fluxes from conventional and zero-tilled soils, where n=80 for conventionally tilled soils and (b) global warming potential from paired zero and conventionally tilled soils. Mean± standard error are shown and n=51, 21 and 8 for soils under zero-till management for 1–5, 6–10 and 11–15 years, respectively.

In contrast to hypotheses ii and iii, potential N_2O fluxes were greater under zero-till soils (0.118 mg m^{-2} h^{-1}) than conventionally tilled soils (0.085 mg m^{-2} h^{-1}), and the duration of zero-tillage did not influence N_2O emissions (Table 2.1). Greater N_2O fluxes from zero-tilled soils have previously been reported[20,21] due to greater water and organic matter content[22]. As a result, there is a larger microbial activity, consuming available O_2, creating anaerobic microsites and enhancing denitrification[23]. In contrast,

conventional tillage disrupts these microsites by increasing oxygenation of the soil, thereby reducing emissions.

Potential CH_4 fluxes were generally positive from conventionally tilled soils (0.007 mg m^{-2} h^{-1}) but negative from zero-tilled soils (-0.04 mg m^{-2} h^{-1}) (Table 2.1) which supported hypothesis ii. In parallel with the N_2O fluxes, CH_4 did not significantly change with time since conversion (Fig. 2.3a, Table 2.1). Zero-tilled soils typically promote CH_4 emissions, but this was contradictory to our findings (Fig. 2.3a, Table 2.1). However, conventional tillage can create an inhospitable environment for methanotrophic organisms. This method destroys hotspots of methanotrophic activity[24] and enhances $NH4_+$ production, therefore inhibiting CH_4 oxidation[25]. Also, increased surface bulk density in zero-tilled soils can reduce CH_4 emissions by enhancing retention time and CH_4 oxidation. Previous studies have reported only gradual responses of CH_4 emissions to soil management, indicating that the recovery of methanotrophic activity in agricultural soil is slow. Soils under zero-till might become a significant CH_4 sink[26,27] only after several decades, suggesting this important ecosystem service is very vulnerable to conventional tillage management.

Critically, our study demonstrated, when combining all three GHG fluxes that the potential GWP from zero-tilled soils was 30 % smaller than that of conventionally tilled soils (Table 2.1) and the duration of zero-tillage significantly reduced GWP (Table 2.1). The reduced GWP was driven by lower CO_2 emissions and increased CH_4 oxidation rates. These findings are in line with a global meta-analysis which reported 66 % lower soil GWP *in-situ*

from recently converted zero-tilled soils compared to conventional tillage[28]. We show that the reduction in potential GWP from zero-tilled soils increased substantially with time, with 75 % lower emissions 11–15 years after conversion compared to paired conventionally tilled soils (Table 2.1). It is plausible that some of the considerable variations in GHG reduction in the literature is linked to this temporal effect and suggests that the full potential of zero-tillage climate mitigation potential might only be realised over time scales of >10 years[33].

Although we show that reduced total porosity in zero-tilled soils is instrumental in reducing GWP, we also observed subsequent development of biopore channels by undisturbed biological activity in longer-term zero-tilled soils increases the surface-connected porosity (Fig. 2.2 and 2.6, $p<0.05$). This, in turn, supported a significant increase in saturated hydraulic conductivity in zero-tilled soils over time from 0.003 to 0.01 cm s^{-1} for 1–5 and 11–15 years, respectively (Table 2.1). This demonstrates that long-term zero-tillage has the dual benefits of mitigating the potential soil CO_2 fluxes and increases CH_4 oxidation and C storage through a reduction in the total soil porosity while reducing the risk of runoff during heavy rainfall through the development of a highly effective, surface-connected porosity.

Table 2.1: Means, standard errors and statistical output from linear mixed modelling (management and duration i.e. time since conversion) for the physio-chemical characteristics of soils under zero (ZT) and conventional tillage (CT).*

Variable	ZT	CT	Management	Duration	Management x duration
Moisture Content (%)	41.8 ± 1.69	35.7 ± 1.67	32.6**	ns	ns
pH	7.14 ± 0.14	6.99 ± 0.14	ns	ns	ns
Microbial C (mg kg^{-1} soil)	497.6 ± 27.06	425.5 ± 27.10	6.1**	5.81**	14.0***
Microbial N (mg kg^{-1} soil)	46.7 ± 4.2	33.0 ± 4.2	15.4***	ns	ns
Bulk Density (g cm^{-3})	1.33 ± 0.02	1.22 ± 0.02	15.7***	ns	ns
Porosity (%)	12.0 ± 1.4	16.4 ± 1.4	13.7***	20.08***	ns
Mean pore size 0-10 cm (mm^2)	0.49 ± 5.5	0.84 ± 5.5	24.6***	ns	ns
Mean pore size 10-20 cm (mm^2)	0.48 ± 8.1	0.76 ± 8.1	19.7***	ns	ns
Saturated hydraulic conductivity (cm s^{-1})	0.0056 ± 0.003	0.0124 ± 0.003	17.4***	3.3**	ns
CO_2 (mg CO_2 m^{-2} h^{-1})	169.8 ± 20.4	252.2 ± 20.4	9.6**	12.6**	7.7**
CH_4 (mg CH_4 m^{-2} h^{-1})	-0.04 ± 0.021	0.007 ± 0.022	1.9**	ns	ns
N_2O (mg N_2O m^{-2} h^{-1})	0.118 ± 0.1	0.085 ± 0.1	4.8**	ns	ns
Global warming potential (mg CO_2 eq. m^{-2} h^{-1})	665.2 ± 76.3	950.8 ± 76.4	10.8***	10.4**	9.3**

ns: not significant *Mean ± standard error of the mean, n=80, log likelihood ratio (*L*) shown for statistical output. ** $p<0.05$ *** $p<0.01$

We assessed how these processes varied over a region, hypothesising that there would be substantial spatial variation. Across the East Midlands, conventionally tilled soils had an average GWP of 950 mg CO_2 eq. m^{-2} h^{-1} (±76.4), compared to 665 mg CO_2 eq. m^{-2} h^{-1} (±76.3) from zero-tilled soils. The variograms for GWP under zero and conventional tillage (Fig. 2.9) show in both cases, there is a substantial apparent intercept to the function showing short-range variation not resolved by sampling. For soils under zero-tillage, there was additional variation, spatially dependent up to about 10 km whereas this spatially correlated variation was minimal under conventional tillage. Consistent with this, spatially interpolated potential GWP from conventionally tilled soils was uniform compared to zero-tilled soils. For conventionally tilled soils there was a GWP hotspot in the south-western part of the study area (850 mg CO_2 eq. m^{-2} h^{-1}), but over most of the study area predicted GWP was in the range 650–700 mg CO_2 eq. m^{-2} h^{-1} (Fig. 2.4a). In contrast, predicted GWP from zero-tilled soils ranged from 200–800 mg CO_2 eq. m^{-2} h^{-1} revealing considerable spatial heterogeneity (Fig. 2.4b). A similar pattern emerged from predicted C stocks, with generally quite uniform C stocks in conventionally tilled soils. In contrast, zero-tilled soils showed a considerable variation in C stocks, ranging from 50–450 Mg ha^{-1} C across the area (Fig. 2.4c & d). The evidence we put forward from across the East Midlands, U.K., demonstrates long-term zero-tillage can both substantially reduce potential GHG emissions whilst simultaneously increasing C stocks across a range of contrasting soil types.

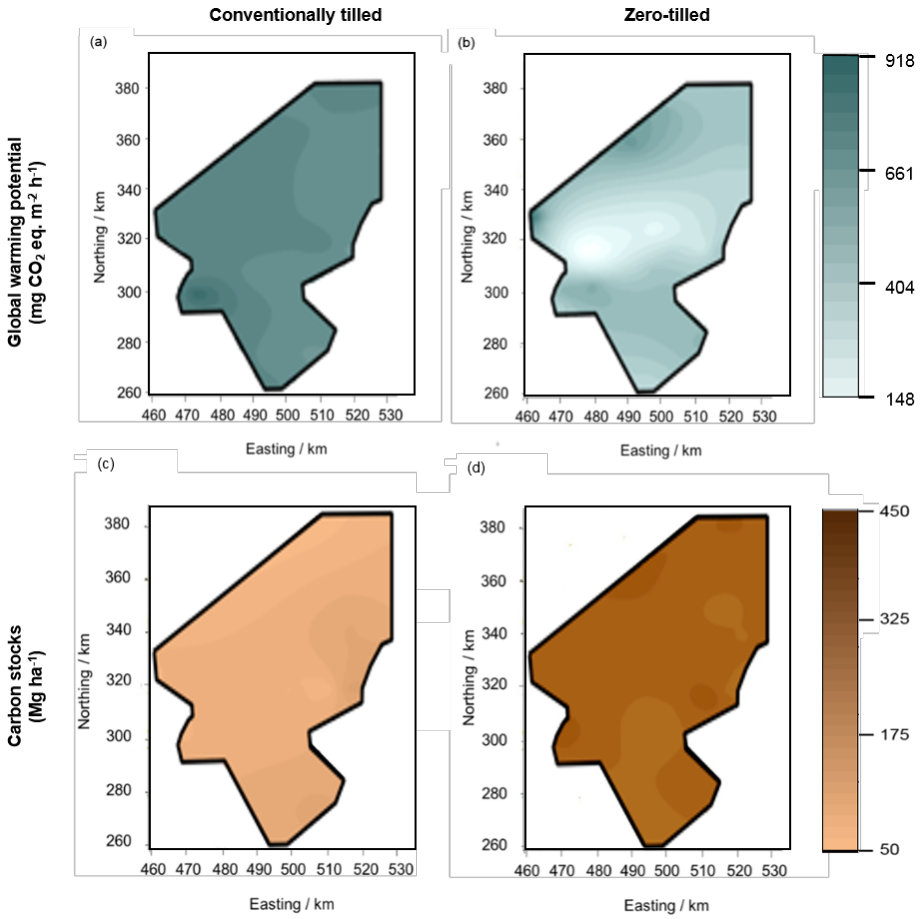


Figure 2.4. Predicted spatial co-variation of (a&b) global warming potential in mg CO_2 eq. m^{-2} h^{-1} and (c&d) total carbon stocks in Mg ha^{-1} in (a&c) conventional and (b&d) zero-tilled soils. See figure 2.5a in methods for geographical location. Note that the scale bars are different within and between managements to illustrate the amplitude of differences. Co-ordinates are relative to the British National Grid-Ordnance Survey.

Our findings contrast with Lugato et al., which suggest the mitigation potential of soil C management has been overestimated as a result of neglecting N_2O emissions in the long-term[13]. However, Lugato et al., used the LUCAS data set in which the soil was sampled to 20 cm depth only; such shallow sampling is unsuitable for addressing these questions as plant roots often extend much deeper[10]. In our study, which considers soil C stock changes

to a depth of 50 cm, as well as the balance between the three major GHGs, zero-tilled soils sequestered more C compared to conventionally tilled soils. While the increased N_2O effluxes from zero-tilled soils were compensated for by reduced CO_2 and CH_4 fluxes.

The climate mitigation potential of zero-tillage also needs to be considered in the context of short- and long-term impacts on yield because mitigation benefits at one site are of no value if reduced production is compensated for by cultivating more land. To date, the majority of studies report little or no difference in yield between the zero and conventional tillage management. A meta-analysis conducted by Pittelkow et al., concluded that yields under zero-till declined in the first 1–2 years following conversion, but corresponded to conventional tillage yields after 3–10 years, bar for both maize and wheat when grown in humid climates[29]. Given the urgent need for climate change mitigation to meet the 1.5 °C warming target[30], and avoid almost certain strong negative impacts on yield from climate change, we suggest in our context zero-tillage is one of many important tools to reduce the carbon footprint of food productions. First via the large fuel saving[31] lowering fossil fuel emissions associated with zero-tillage and second via the reduced GWP and the long-term C sequestration in the soil which we report here.

2.3 Methods

2.3.1 Site selection and sample collection

This study was conducted across the East Midlands of England in the U.K., covering an area of 6,504 km^2 (Fig. 2.5). Sites, 160 in total, were located in 80 pairs of fields, each pair comprising one conventionally tilled field and one zero-tilled field. The fields in each pair were adjacent to each other so that the sample sites were no further than 10 metres apart. This was to reduce climatic and soil variability within pairs. Paired fields could not be selected independently and at random according to a probability sampling design as farmers were recruited based on having a zero-tilled field adjacent to a neighbouring conventionally-tilled field thus sampling was constrained by farmers that volunteered. All zero-tilled soils had been managed this way for between one and 15 years whereas the conventionally tilled soils were subjected to annual mechanical turnover to a depth of at least 10 cm. All sample collection was undertaken between November and December 2015, approximately one to two months after sowing cereals. This time of year, was pre-selected due to early and slow root growth in cold temperatures, ensuring changes to soil structure were minimised, and access to the soil surface (without an established crop) could be readily achieved.

Soil sampling was undertaken approximately 4 m from the field boundary and not on the headland. Intact soil cores (5 cm diameter and 30 cm depth) were collected using a manual core sampler that used transparent sample liner tubes (Van Walt Ltd, Haslemere, U.K.) for X-ray CT and GHG emission analysis. Additional intact soil cores (6 cm diameter and 15 cm depth) were also collected in a PVC cylinder for saturated hydraulic conductivity measurements. Samples of the surface soil were collected using a stainless-steel cutting cylinder (7 cm

diameter and 4 cm height) for the measurement of dry bulk density. To determine depth profiles in soil carbon, microbial biomass carbon and nitrogen, soil water content and determination of organic matter functional chemistry soil samples were taken to a depth of 50 cm, in 10 cm increments, using a Dutch auger. All samples were kept at 4°C until analysis.

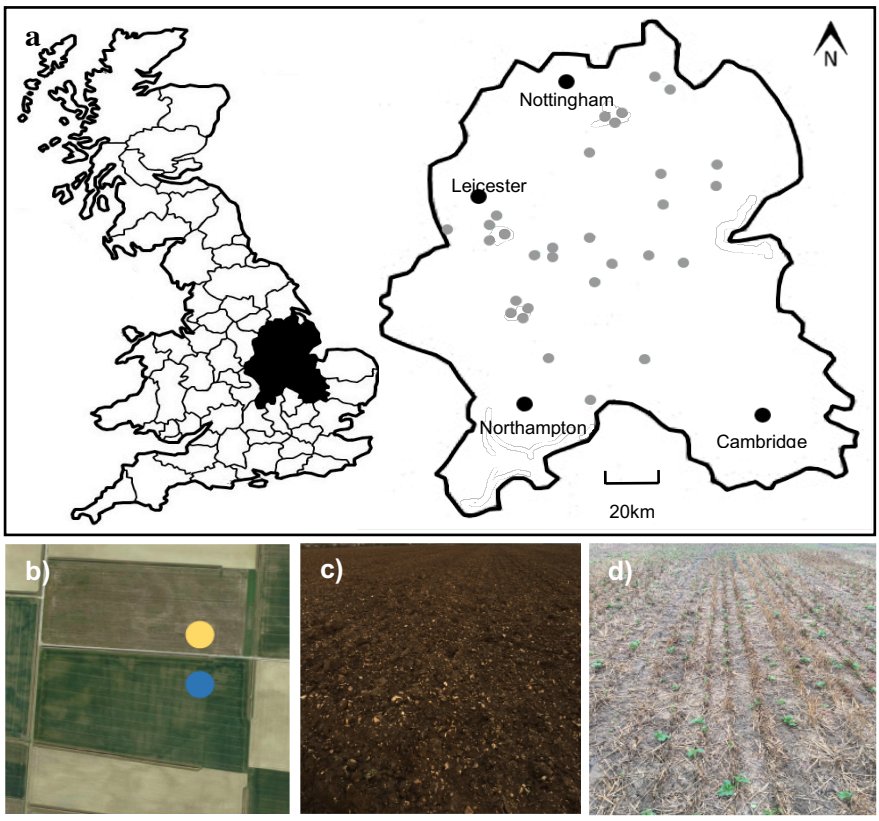


Figure 2.5 (a). The location of the 31 sampled farms (grey circles), where a maximum of three pairs of zero and conventionally tilled fields were taken from each farm until a selection of 80 pairs were collected. An example (b) of a pair in Lincolnshire where the soil samples were not taken more than 10 meters apart, the conventionally tilled field in shown in yellow (c) and the zero-tilled field shown in blue (d).

2.3.2 Laboratory analysis

2.3.2.1 Soil physical properties

The soil was oven-dried at 105 °C and weighed to determine the dry bulk density and gravimetric water content. Particle size analysis was performed using the hydrometer method[32] and textural classification made according to the Soil Survey of England and Wales classification[33]. Saturated hydraulic conductivity was determined using the standard constant head method[34].

2.3.2.2 X-ray Computed Tomography

Before the measurement of GHGs, 3-D X-ray Computed Tomography (CT) was undertaken on all 160 intact soil cores in a Phoenix V|Tome|X m X-ray scanner 240 kV (GE Measurement & Control Solutions, Wunstrof, Germany) at the Hounsfield Facility at the University of Nottingham. This allowed visualisation and quantification of soil porous architecture. Voxel resolution was set to 50 µm, potential energy of 160 kV and a current of 180 uA. Soil cores were scanned in three sections (i.e. depths), with a total scan time of 30 minutes per core. In this chapter (2), soil cores were scanned at a resolution of 50 µm, therefore the porosity observed accounts for pore sizes which are greater than 50 µm, any pore sizes below this resolution are not accounted for. Therefore, throughout this thesis, the 'total porosity' is the 'detectable porosity' at that particular resolution for each chapter. A total of 2998 image projections were captured for each core. Images were reconstructed at 32-bit using Phoenix Datos x 2 reconstruction tool to form a 3D volume which was visualised using VG Studio Max 2.2.5 software. Due to the irregularity of soil surface between samples, the first 50 images slices from the top of each core were excluded from the analysis.

Analysis of soil pore morphology was carried out using *ImageJ,* an open-source image processing software. A cuboid region of interest (ROI) (50 ×50 mm^3) was selected from the centre of the reconstructed CT images to exclude pores adjacent to the core edges. The ROI was subject to one pass of the 'sharpen' filter followed by one pass of the 'median' filter to enhance the image quality before being transformed to the binary scale using the 'Li' threshold algorithm[35]. A further filter operation ('despeckle') was undertaken on the binary images before analysis. Finally, a wide range of pore morphological properties was collected using the 'analyse particles' function, including pore size and shape. Surface connected porosity was determined in VG Studio Max 2.2.5 by isolating all of the pores connected at the surface down into the sample using the 'flood fill' tool. As the ROI was selected below the immediate surface, it is possible the surface connected porosity has been slightly underestimated.

2.3.2.3 Soil biochemical properties

Soil pH and microbial biomass carbon (MBC) and nitrogen (MBN) were determined for all surface samples (0-10 cm), and total soil carbon and nitrogen (N) was measured on the soil samples down to 50 cm at 10 cm intervals. Soil pH was estimated on a 1:5 soil-water mixture. MBC and MBN were determined using the chloroform fumigation method[36]. For this fumigated (for 24 h) and non-fumigated soils (10.0 ± 0.5 g fresh sample) were extracted with 0.5 M K_2SO_4 followed by analysis using a Shimadzu CN analyser (TOC-V CPH Shimadzu). The value of the coefficient (K_{EC}) to convert 'chloroform-labile' carbon to microbial biomass carbon of 0.45 was used,[37] and for microbial biomass nitrogen (N_M) the value of K_{EN} was taken as 0.54[38]. Total soil C and N

were determined from 20 mg of oven-dried, ball-milled soil combusted using a total element analyser (Flash EA 1112, CE Instruments, Wigan, U.K.).

Five pairs of soils (0-50 cm in 10 cm increments) that had been under zero-till management for 1, 6 and 15 years were randomly chosen for determining organic matter functional chemistry. Fourier transform infrared (FTIR) absorption spectra were obtained with a Bruker Tensor 27 FTIR equipped with N_2 purge gas generator and an MCT detector. A total of 128 scans were performed on each oven-dried, ball-milled sample, and background spectra were run initially and after every eight samples. The spectral range spanned from 550 to 4000 cm^{-1} at a resolution of 1 cm^{-1}. All spectra were standardised, smoothed and baseline corrected (SpectraGryph v.1.2) before statistical analysis in order to allow direct comparison.

2.3.2.4 Potential greenhouse gas fluxes

Potential GHG fluxes were measured following incubation under a controlled environment. This approach was adopted to allow for comparison between management by removing the effects of climate on gas production *in-situ*. Intact soil cores were removed from the 4 °C environment, and water-filled pore space was adjusted to 60% to represent field condition in all soil cores and kept at 15 °C for 48 hours to activate and stabilise the soil microflora. Each soil core was incubated in an airtight plastic jar (1.5 dm^3) with a septum on the top to allow gas sampling. The air in the headspace was mixed via a magnetic stirrer during gas sampling at time intervals 0, 15, 30 and 60 minutes using a 20 mL syringe. The collected gas samples were stored in pre-evacuated 12 mL airtight glass vials. Gas samples were analysed for concentrations of CO_2, CH_4 and N_2O by

gas chromatography (GC-2014, Shimadzu) and potential GHG flux calculations were made from the soil cores using the ideal gas law[17]. During the incubation, we presumed no uptake mechanisms (e.g. photosynthesis) for CO_2 accumulated during the incubation because there was no plant material in the soil cores due to the timing of sampling in the field. Therefore, CO_2 flux represented CO_2 production rates. We expressed N_2O flux as a net change between production and consumption of N_2O. CH_4 flux was also considered to represent the balance between consumption and production processes of CH_4 as CH_4 production by the anaerobic degradation of organic material is minimised under aerobic conditions, and all agricultural soils uptake CH_4 through oxidation by methanotrophic bacteria except for flooded/poorly drained sites[39]. Additionally, the total concentrations of GHGs during the incubation was presented on a 100-yr global warming potential (GWP) weighted basis to compare the relative contribution of each of the gases measured to the potential impacts of net GHG emission on global warming potential by tillage following Myhre *et al*[40].

This incubation method on intact soil cores and scaling of emissions to produce an estimated GWP has been used in several studies, including Lee *et al*[41] to investigate the relationship between GHG emissions and soil indicators in conventional and recently converted zero-tilled soils, by Norberg *et al*[42] to measure CO_2 emissions from intact cores of cultivated peat soils and by Ball[43] to assess how soil structure affected by wheel compaction, animal trampling and tillage managements influence GHG emissions.

2.3.2.5 Estimating carbon stocks

Soil carbon stocks were estimated by an equivalent soil mass (ESM) procedure to calculate carbon stocks in multiple soil layers (Mg C ha^{-1}) within a defined area using calculations from Wendt and Hauser[12]. This method quantifies and corrects for the fixed depth error associated with calculating carbon stocks as the product of soil bulk density, depth and concentration.

2.3.3 Statistical Analysis

Statistical analysis was carried out in R v.3.4.1 and all data checked for normality. If the residuals from fitted models did not appear consistent with a normal distribution, then a Box-Cox transformation was applied, using the BOXCOX procedure in the MASS package for the R platform[44].

2.3.3.1 Linear mixed model

Model-based analyses were used, specifically a linear mixed model (LMM) [45]. Variation of a soil property, z, was treated as a linear combination of fixed effects (categorical or continuous covariates). The variation not accounted for by the fixed effects was modelled as random effects: a spatially correlated Gaussian random field and an independent error, which includes any variation spatially correlated but only at scales too fine to be resolved by the sampling design. The LMM for a set of *n* observations of a property can be written as

$$\mathbf{z} = \mathbf{X}\boldsymbol{\tau} + \mathbf{u} + \boldsymbol{\varepsilon}, \qquad [1]$$

where $\mathbf{z}$ is an n-vector containing n observations of variable z, $\mathbf{X}$ is a $n \times P$ design matrix which includes any covariates, $\boldsymbol{\tau}$ is a vector of fixed effects coefficients, $\mathbf{u}$ is a vector of correlated random variables, and $\boldsymbol{\varepsilon}$ is a vector of independent and identically distributed random variables, the nugget component.

The random effect **u** is a realisation of a spatial random function $U(\mathbf{x})$ where **x** denotes a location in space. We assume that U is second-order stationary[46] with mean zero and variance $\square^2$. This means that the spatial correlation, between $U(\mathbf{x}_1)$ and $U(\mathbf{x}_2)$ depends only on the difference between the locations, or lag vector $\mathbf{x}_1 - \mathbf{x}_2$. In this study, we made a further assumption of isotropy, by which the spatial correlation of the random effect is modelled as a function just of the scalar part of the lag, or lag distance, $h = |\mathbf{x}_1 - \mathbf{x}_2|$.

In this study, we used the Matérn function to describe spatial correlation[47] due to its flexibility[48]. It has a smoothness parameter that directly controls correlation at small distances[49], allowing it to describe a range of spatial processes. The Matérn correlation function[47] is written

$$g(h) = \{2^{\kappa-1}\Gamma(\kappa)\}^{-1}\left(\frac{h}{\phi}\right)^{\kappa} \mathrm{K}_{\kappa}\left(\frac{h}{\phi}\right), \qquad [2]$$

where κ is a smoothness parameter, K_{κ} is a modified Bessel function of the second kind of order κ, Γ is the gamma function, and ϕ is a distance parameter. These have to be estimated in addition to the variance σ^2 and ν^2 the variance of ϵ. This is best done by residual maximum likelihood (REML)[46] which minimises bias in the estimates of parameters of the random effects model by undertaking the likelihood maximization for a generalized filtered version of the data in which the fixed effects are zero.

We fixed the smoothness parameter at a series of values representing a range of degrees of smoothness of the random process and maximizing the residual likelihood with respect to the others. The value of κ for which the residual likelihood was largest was selected. Table 1 shows results for an LMM with

management and duration under zero-tillage as fixed effects for the variable global warming potential and stationary random effects with κ fixed at different values. The value of 0.1 was chosen as it had the smallest negative log residual likelihood.

Table 2.2. Estimated stationary variance parameters for LMM with the variable global warming potential.

κ (fixed)	C_0	C_1	ℓ
0.1	26.943	6.742	-499.65
0.5	29.663	4.030	-499.35
1.0	30.016	3.640	-499.33
1.5	30.185	3.449	-499.33
2.0	30.293	3.328	-499.34

For the estimation of the model variance (random component and spatial dependence) and estimation of the fixed effect parameters (e.g. mean value for zero-till and conventional sites), a covariance matrix is obtained by

$$\hat{\mathbf{C}} = (\mathbf{X}^T\mathbf{V}^{-1}\mathbf{X})^{-1}, \quad [3]$$

where the covariance matrix $\mathbf{V}$ is obtained from the variance parameters estimated by REML and $\mathbf{X}$ is the overall fixed effects design matrix introduced in Equation 1. The standard errors of the estimated parameters are the square roots of the terms on the main diagonal of $\hat{\mathbf{C}}$.

For significance testing, we evaluated the evidence for including particular fixed effects in a model by a log-likelihood ratio test, e.g. the null model which had an overall mean and a more complex model in which there were different mean

values for sites under zero-tillage and those under conventional tillage. The log-likelihood ratio statistic L is written as

$$\boldsymbol{L = 2(l_N - l)}, \qquad [4]$$

where l_N and l are the logs of the maximum likelihoods for the null and more complex models respectively. Note that we used conventional maximum likelihood rather than REML when fitting models for this inference. The significance of the extra term in the more complicated model was tested by comparing L with the Chi-square distribution with degrees of freedom equal to the number of extra parameters in the more complex model.

2.3.3.2 Linear model of co-regionalization and co-kriging

The LMM described above treats the variables as realisations of spatially correlated random processes therefore we can fit models to two or more variables which are based on underlying auto-correlated and cross-correlated random fields. A linear model of co-regionalisation (LMCR) modelled the joint spatial variation of the responses and provided parameters for modelling the auto-semivariograms and cross-semivariograms. These parameters were then used to describe the spatial (co-) variation of the treatment responses and used to map the treatment response and their contrasts by co-kriging. A detailed example of how the LMCR can be used to compare treatments across a region is given by Bishop and Lark[50].

2.4 References

1. Brevik, E. C. Soils and Climate Change: Gas Fluxes and Soil Processes.

Soil Horizons. **53**, 12-23 (2012).

2. Derpsch, R., Friedrich, T., Kassam, A. & Hongwen, L. Current status of adoption of no-till farming in the world and some of its main benefits. *Int. J. Agric. Biol. Eng.* **3,** 1–25 (2010).
3. Stockmann, U. *et al.* The knowns, known unknowns and unknowns of sequestration of soil organic carbon. *Agric. Ecosyst. Environ.* **164,** 80–99 (2013).
4. EPA. Greenhouse gas emissions. *Global greenhouse gas emissions data* (2014).
5. Green, V. S., Stott, D. E., Cruz, J. C. & Curi, N. Tillage impacts on soil biological activity and aggregation in a Brazilian Cerrado Oxisol. *Soil Tillage Res.* **92,** 114–121 (2007).
6. Song, K. *et al.* Influence of tillage practices and straw incorporation on soil aggregates, organic carbon, and crop yields in a rice-wheat rotation system. *Sci. Rep.* **6,** (2016).
7. Powlson, D. S. *et al.* Limited potential of no-till agriculture for climate change mitigation. *Nat. Clim. Chang.* **4,** 678–683 (2014).
8. X.Liu, S.J. Herbert, A.M. Hashemi, X. Zhang, G. D. Effects of agricultural management on soil organic matter and carbon transformation - a review. *Plant Soil Environ.* **52,** 531–543 (2006).
9. Six, J., Bossuyt, H., Degryze, S. & Denef, K. A history of research on the link between (micro)aggregates, soil biota, and soil organic matter dynamics. *Soil Tillage Res.* **79,** 7–31 (2004).
10. Baker, J. M., Ochsner, T. E., Venterea, R. T. & Griffis, T. J. Tillage and soil carbon sequestration - What do we really know? *Agric. Ecosyst. Environ.* **118,** 1–5 (2007).

11. Ellert, B. H. & Bettany, J. R. Calculation of organic matter and nutrients stored in soils under contrasting management regimes. *Can. J. Soil Sci.* **75,** 529–538 (1995).

12. Wendt, J. W. & Hauser, S. An equivalent soil mass procedure for monitoring soil organic carbon in multiple soil layers. *Eur. J. Soil Sci.* **64,** 58–65 (2013).

13. Lugato, E., Leip, A. & Jones, A. Mitigation potential of soil carbon management overestimated by neglecting N2O emissions. *Nat. Clim. Chang.* **8,** 219–223 (2018).

14. Vranova, V., Rejsek, K. & Formanek, P. Aliphatic, cyclic, and aromatic organic acids, vitamins, and carbohydrates in soil: A review. *The Scientific World Journal.* **52,** 1-15 (2013).

15. Mangalassery, S., Mooney, S. J., Sparkes, D. L., Fraser, W. T. & Sjögersten, S. Impacts of zero tillage on soil enzyme activities, microbial characteristics and organic matter functional chemistry in temperate soils. *Eur. J. Soil Biol.* **68**, 9-17 (2015).

16. Mangalassery, S. *et al.* To what extent can zero tillage lead to a reduction in greenhouse gas emissions from temperate soils? *Sci. Rep.* **4,** 4586 (2015).

17. Busari, M. a., Kukal, S. S., Kaur, A., Bhatt, R. & Dulazi, a. a. Conservation tillage impacts on soil, crop and the environment. *Int. Soil Water Conserv. Res.* **3,** 1–11 (2015).

18. Duiker, S. W. & Lal, R. Carbon budget study using CO_2 flux measurements from a no till system in central Ohio. *Soil Tillage Res.* **54, 21-30** (2000).

19. Schaufler, G. *et al.* Greenhouse gas emissions from European soils under

different land use: Effects of soil moisture and temperature. *Eur. J. Soil Sci.* **61,** 1-9 (2010).

20. Bouwman, A. F. Direct emission of nitrous oxide from agricultural soils. *Nutr. Cycl. Agroecosystems.* **46,** 53-70 (1996).
21. Baggs, E. M., Rees, R. M., Smith, K. a. & Vinten, a J. a. Nitrous oxide emission from soils after incorporating crop residues. *Soil Use Manag.* **16,** 1-8 (2000).
22. Alvarez, R. & Steinbach, H. S. A review of the effects of tillage systems on some soil physical properties, water content, nitrate availability and crops yield in the Argentine Pampas. *Soil and Tillage Research*. **104,** 1-15 (2009).
23. Signor, D. & Cerri, C. E. P. Nitrous oxide emissions in agricultural soils: a review. *Pesqui. Agropecuária Trop.* **22,** 426-433 (2013).
24. Bayer, C. *et al.* Methane emission from soil under long-term no-till cropping systems. *Soil Tillage Res.* **124,** 1-7 (2012).
25. Hütsch, B. W. Tillage and land use effects on methane oxidation rates and their vertical profiles in soil. *Biol. Fertil. Soils* **27,** 284-292 (1998).
26. Jacinthe, P. A. & Lal, R. Labile carbon and methane uptake as affected by tillage intensity in a Mollisol. *Soil Tillage Res.***80,** 35-45 (2005).
27. Jacinthe, P. A. & Lal, R. Methane oxidation potential of reclaimed grassland soils as affected by management. *Soil Sci.* **171,** 772-783 (2006).
28. Sainju, U. M. A Global Meta-Analysis on the Impact of Management Practices on Net Global Warming Potential and Greenhouse Gas Intensity from Cropland Soils. *PLoS One* **11,** (2016).
29. Pittelkow, C. M. *et al.* When does no-till yield more? A global meta-analysis. *F. Crop. Res.* **183,** 156–168 (2015).

30. Intergovernmental Panel on Climate Change. *Special Report Global Warming of 1.5 ºC*. (2018).

31. Kertész & Madarász, B. Conservation Agriculture in Europe. *Int. Soil Water Conserv. Res.* **2,** 91-96 (2014).

32. Day, P. . in *Methods of Soil Analysis. Part 1.* 545–567 (1965).

33. Avery, B.W. Classification in the soil survey of England and Wales, *J. Soil Sci.* **24** (1973).

34. Sarki, A., Mirjat, M. S., Mahessar, A. A., Kori, S. M. & Qureshi, A. L. Determination of Saturated Hydraulic Conductivity of Different Soil Texture Materials. *J. Agric. Vet. Sci.***7,** 56-62 (2014).

35. Li, C. H. & Tam, P. K. S. An iterative algorithm for minimum cross entropy thresholding. *Pattern Recognit. Lett.* **19,** 771-776 (1998).

36. Vance, E. D., Brookes, P. C. & Jenkinson, D. S. An extraction method for measuring soil microbial biomass C. *Soil Biol. Biochem.* **19,** 703–707 (1987).

37. Jenkinson, D. S., Brookes, P. C. & Powlson, D. S. Measuring soil microbial biomass. *Soil Biology and Biochemistry* **36,** 5–7 (2004).

38. Brookes, P. C., Landman, A., Pruden, G. & Jenkinson, D. S. Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil. *Soil Biol. Biochem.* **17,** 837–842 (1985).

39. Conrad, R. Soil microorganisms as controllers of atmospheric trace gases (H2, CO, CH_4, OCS, N_2O, and NO). *Microbiol. Rev.* (1996).

40. Myhre, G. *et al.* Anthropogenic and Natural Radiative Forcing. *Clim. Chang. 2013 Phys. Sci. Basis. Contrib. Work. Gr. I to Fifth Assess. Rep. Intergov. Panel Clim. Chang.* 659–740 (2013).

41. Lee, J., Six, J., King, A. P., van Kessel, C. & Rolston, D. E. Tillage and Field Scale Controls on Greenhouse Gas Emissions. *J. Environ. Qual.* **35,** 714 (2006).

42. Norberg, L., Berglund, Ö. & Berglund, K. Impact of drainage and soil properties on carbon dioxide emissions from intact cores of cultivated peat soils. *Mires Peat* **21,** 1-14 (2018).

43. Ball, B. C. Soil structure and greenhouse gas emissions: A synthesis of 20 years of experimentation. *Eur. J. Soil Sci.* **64,** 357–373 (2013).

44. Venables, W. N. & Ripley, B. D. *Modern Applied Statistics with S Fourth edition.* Springer-Verlag. New York (2002).

45. de Gruijter, J., Brus, D. ., Bierkens, M. F. . & Knotters, M. *Sampling for Natural Resource Monitoring*. (2006).

46. Stein, M. L. *Interpolation of Spatial Data*. (1999). Springer-Verlag New York.

47. Matérn, B. Spatial Variation. *Reports For. Res. Inst. Sweden* **49,** 84 (1960).

48. Handcock, M. . & Stein, M. L. A Bayesian analysis of kriging. *Technometrics* **35,** 403–410 (1993).

49. Genton, M. G. & Kleiber, W. Cross-Covariance Functions for Multivariate Geostatistics. *Stat. Sci.* **30,** 147–163 (2015).

50. Bishop, T. F. A. & Lark, R. M. A landscape-scale experiment on the changes in available potassium over a winter wheat cropping season. *Geoderma* **141,** 384-396 (2007).

2.6 Additional information

2.6.1 Site information

Table 2.3 Selected soil and management characteristics of experimental sites.

Site	County	Years in ZT	Soil Texture	Crop management (ZT/CT)	WRB
1	Lincolnshire	6	Sandy Loam	W/W	Gleysols
2	Lincolnshire	6	Sandy Clay	F/WB	Stagnosols
3	Lincolnshire	6	Sandy Clay	W/W	Stagnosols
4	Lincolnshire	6	Clay	F/M	Stagnosols
5	Lincolnshire	6	Sandy Clay Loam	WT/OSR	Stagnosols
6	Lincolnshire	6	Sandy Loam	OSR/OSR	Stagnosols
7	Lincolnshire	6	Sandy Loam	OSR/OSR	Leptosols
8	Lincolnshire	6	Sandy Clay Loam	OSR/OSR	Leptosols
9	Lincolnshire	6	Sandy Clay Loam	OSR/OSR	Leptosols
10	Lincolnshire	5	Sandy Clay Loam	G/P	Cambisols
11	Lincolnshire	6	Sandy Loam	P/P	Cambisols
12	Lincolnshire	3	Sandy Loam	WT/WT	Cambisols
13	Lincolnshire	1	Sandy Loam	OSR/OSR	Leptosols
14	Lincolnshire	3	Sandy Loam	P/P	Leptosols
15	Lincolnshire	2	Sandy Loam	WT/WT	Leptosols
16	Lincolnshire	1	Sandy Loam	W/W	Gleysols
17	Lincolnshire	1	Sandy Silt Loam	W/W	Gleysols
18	Lincolnshire	1	Clay Loam	W/F	Gleysols
19	Lincolnshire	4	Silty Clay	OSR/OSR	Gleysols
20	Lincolnshire	6	Sandy Loam	W/W	Leptosols
21	Lincolnshire	6	Clay Loam	W/OSR	Leptosols
22	Lincolnshire	6	Sandy Clay Loam	W/OSR	Leptosols
23	Lincolnshire	3	Sandy Clay	WB/WB	Stagnosols
24	Lincolnshire	2	Clay	WB/WB	Cambisols
25	Lincolnshire	7	Clay	WB/WB	Cambisols
26	Leicestershire	5	Clay	W/WT	Stagnosols
27	Leicestershire	5	Clay	W/W	Stagnosols
28	Nottinghamshire	1	Clay	F/F	Stagnosols
29	Nottinghamshire	1	Clay Loam	WT/WT	Stagnosols
30	Nottinghamshire	2	Sandy Clay	WT/W	Stagnosols
31	Leicestershire	1	Sandy Loam	G/G	Stagnosols
32	Leicestershire	1	Clay Loam	G/G	Stagnosols
33	Lincolnshire	15	Clay	W/W	Cambisols
34	Lincolnshire	1	Clay	W/W	Cambisols
35	Lincolnshire	15	Clay	G/G	Stagnosols
36	Leicestershire	15	Clay	WT/WT	Stagnosols
37	Leicestershire	15	Clay	W/W	Stagnosols
38	Leicestershire	15	Clay	F/W	Stagnosols
39	Leicestershire	2	Clay	WT/WT	Stagnosols
40	Leicestershire	2	Clay	F/W	Stagnosols
41	Leicestershire	2	Clay	WT/F	Stagnosols
42	Leicestershire	2	Clay Loam	W/W	Stagnosols
43	Leicestershire	2	Clay Loam	OSR/OSR	Cambisols
44	Rutland	15	Sandy Silt Loam	WB/WB	Luvisols
45	Rutland	15	Clay Loam	WB/W	Leptosols

46	Rutland	15	Sandy Silt Loam	WB/W	Leptosols
47	Leicestershire	1	Clay	WB/W	Planosols
48	Leicestershire	1	Sandy Clay Loam	G/G	Planosols
49	Leicestershire	1	Clay	WB/WB	Stagnosols
50	Lincolnshire	7	Sandy Loam	G/G	Leptosols
51	Lincolnshire	7	Clay Loam	F/F	Leptosols
52	Rutland	7	Sandy Silt Loam	OSR/OSR	Leptosols
53	Lincolnshire	1	Clay	F/W	Gleysols
54	Lincolnshire	1	Sandy Clay Loam	F/F	Cambisols
55	Cambridgeshire	4	Clay	WB/WB	Cambisols
56	Cambridgeshire	4	Clay	WB/W	Cambisols
57	Cambridgeshire	4	Clay	WB/WB	Cambisols
58	Bedford	1	Clay Loam	G/G	Cambisols
59	Bedford	1	Clay Loam	W/W	Cambisols
60	Lincolnshire	2	Clay Loam	WT/WT	Stagnosols
61	Lincolnshire	2	Clay	WT/WT	Stagnosols
62	Lincolnshire	2	Clay	M/M	Stagnosols
63	Lincolnshire	2	Sandy Silt Loam	WB/WB	Leptosols
64	Lincolnshire	2	Clay	WT/WT	Leptosols
65	Lincolnshire	2	Sandy Loam	OSR/WT	Leptosols
66	Leicestershire	1	Clay	G/F	Stagnosols
67	Leicestershire	1	Clay	G/F	Stagnosols
68	Leicestershire	3	Clay	W/W	Luvisols
69	Leicestershire	3	Sandy Clay	M/M	Luvisols
70	Leicestershire	5	Clay	W/W	Stagnosols
71	Lincolnshire	7	Clay	WB/WB	Gleysols
72	Lincolnshire	7	Clay	WB/WB	Gleysols
73	Lincolnshire	7	Clay	WT/W	Gleysols
74	Nottinghamshire	1	Sandy Silt Loam	WT/WT	Stagnosols
75	Nottinghamshire	1	Clay	WT/W	Stagnosols
76	Nottinghamshire	1	Sandy Clay	W/W	Stagnosols
77	Leicestershire	3	Clay	W/W	Planosols
78	Leicestershire	3	Clay	W/W	Planosols
79	Leicestershire	3	Clay	WT/WT	Stagnosols
80	Nottinghamshire	2	Sandy Clay Loam	WB/WB	Cambisols

2.6.2 Total and surface connected porosity

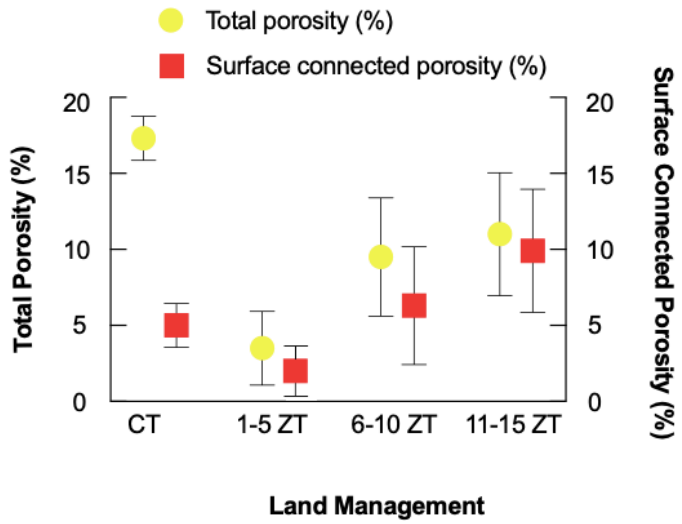


Figure 2.6 The total porosity (left y-axis) and surface connected porosity (right y-axis), as analysed using X-ray Computed Tomography. Soil cores had been in conventional tillage (CT), and between 1 and 5 (1-5 ZT), 6 and 10 (6-10 ZT) and 11-15 (11-15 ZT) years in zero-tillage (ZT).

2.6.3 Graphical abstract

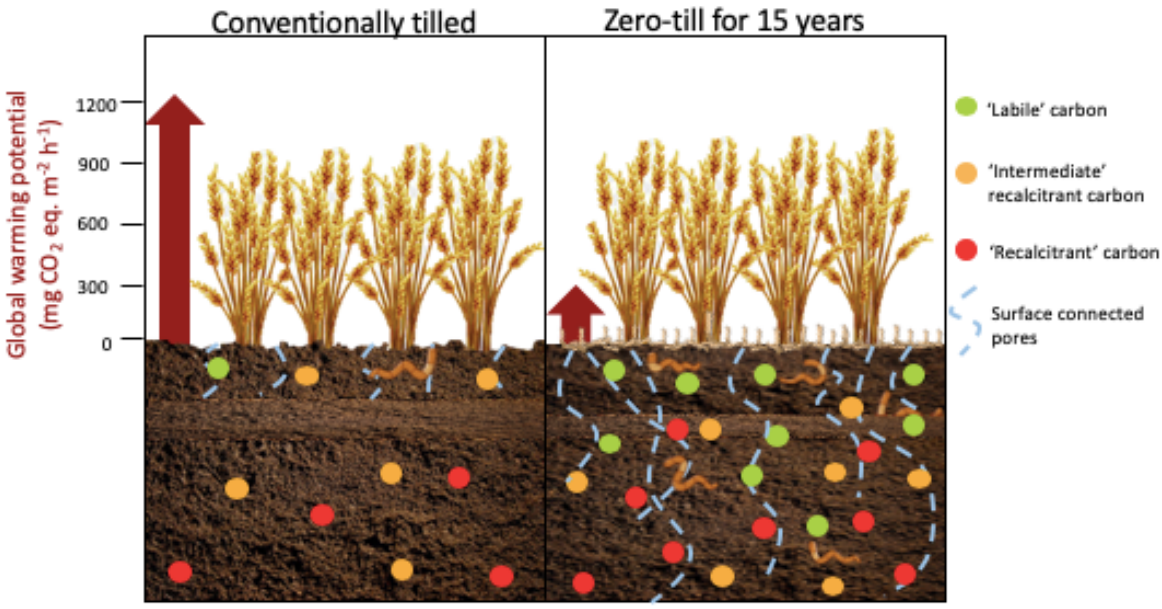


Figure 2.7 A graphical abstract presented at world congress of soil science 2019 illustrating the main conclusions of this paper.

2.6.4 FTIR analysis

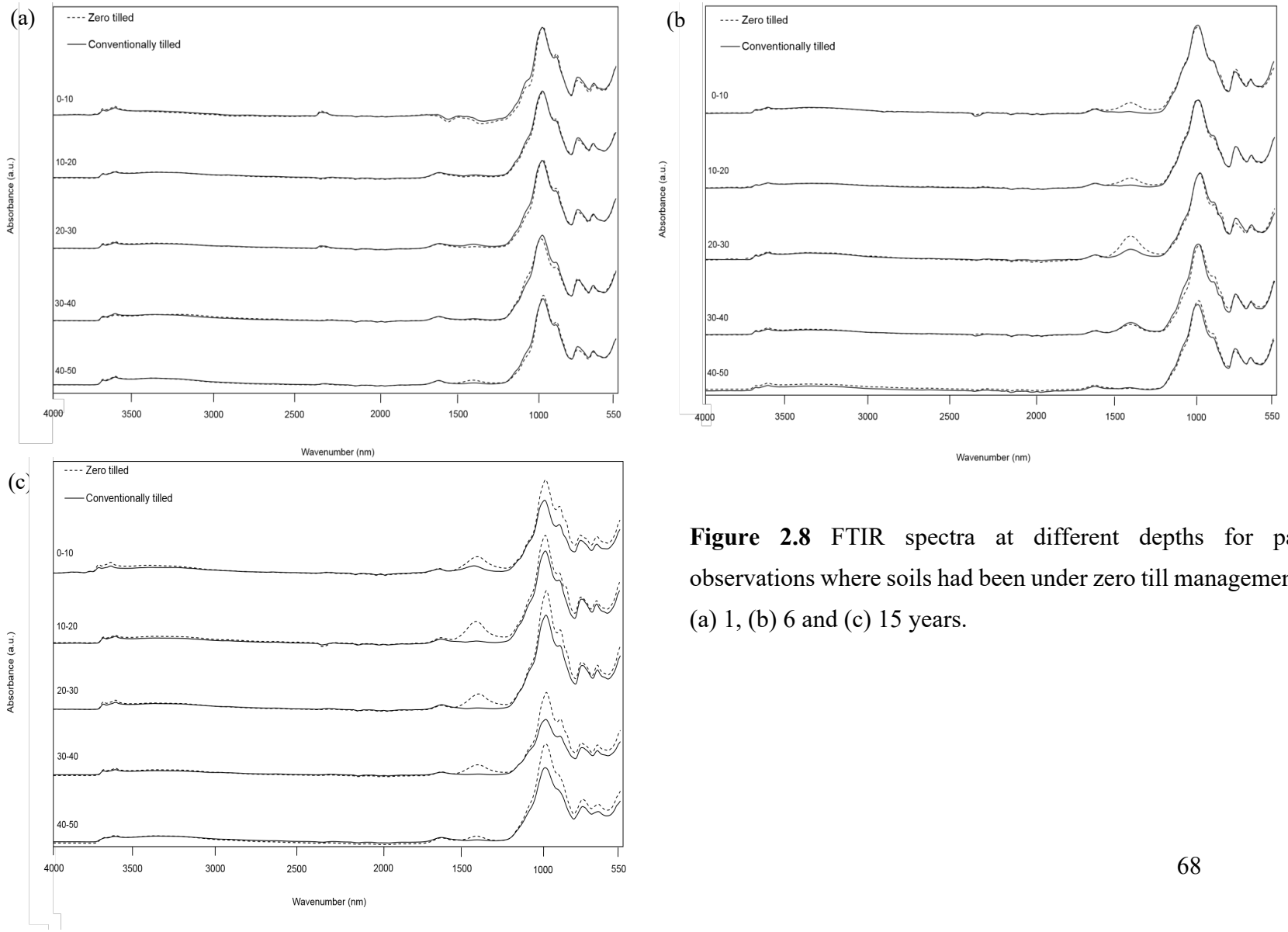


Figure 2.8 FTIR spectra at different depths for paired observations where soils had been under zero till management for (a) 1, (b) 6 and (c) 15 years.

Table 2.4 F statistic from analysis of variance (ANOVA) for the absorbance at different wavelengths from FTIR spectra. *p<0.05**p<0.01***p<0.001.

Wavelength (nm)	Functional group	Tillage	Depth	Tillage x Depth	Tillage	Depth	Tillage x Depth	Tillage	Depth	Tillage x Depth
		Paired 1 year zero-till fields			Paired 6 years zero-till fields			Paired 15 years zero-till fields		
2925	CHn, Aliphatics	0.21	582.3**	0.87	0.18	403.1**	0.5	1.93	172.9**	0.54
2850	CHn, Aliphatics	0.5	1000.56**	1.14	0.03	497.7**	0.89	1.57	208.86**	0.62
1422	Aliphatics	0.39	24.36**	0.83	1.85	2.48	0.67	4.66*	1.03	0.11
1004	Ether	0.05	10.66**	1.46	0.02	18.61**	0.99	8.95**	7.57**	0.09
684	Aromatics	1.45	13.43**	0.08	1.34	52.99**	1.4	6.6**	9.58**	0.17
671	Aromatics	0.5	45.51**	0.08	0.39	54.85**	1.53	5.84*	8.76**	0.22
669	Aromatics	0.39	47.12**	0.08	0.35	55.82**	1.54	6.8*	8.81**	0.16
665	Aromatics	0.92	42.36***	0.26	0.3	57.65**	1.53	6.83*	8.86**	0.16
651	Aromatics	0.83	47.23***	0.24	0.27	69.54**	1.5	6.44*	9.06**	0.13
649	Aromatics	0.23	53.25**	0.06	0.27	71.7**	1.51	6.4*	9.11*	0.12

2.6.5 Variograms

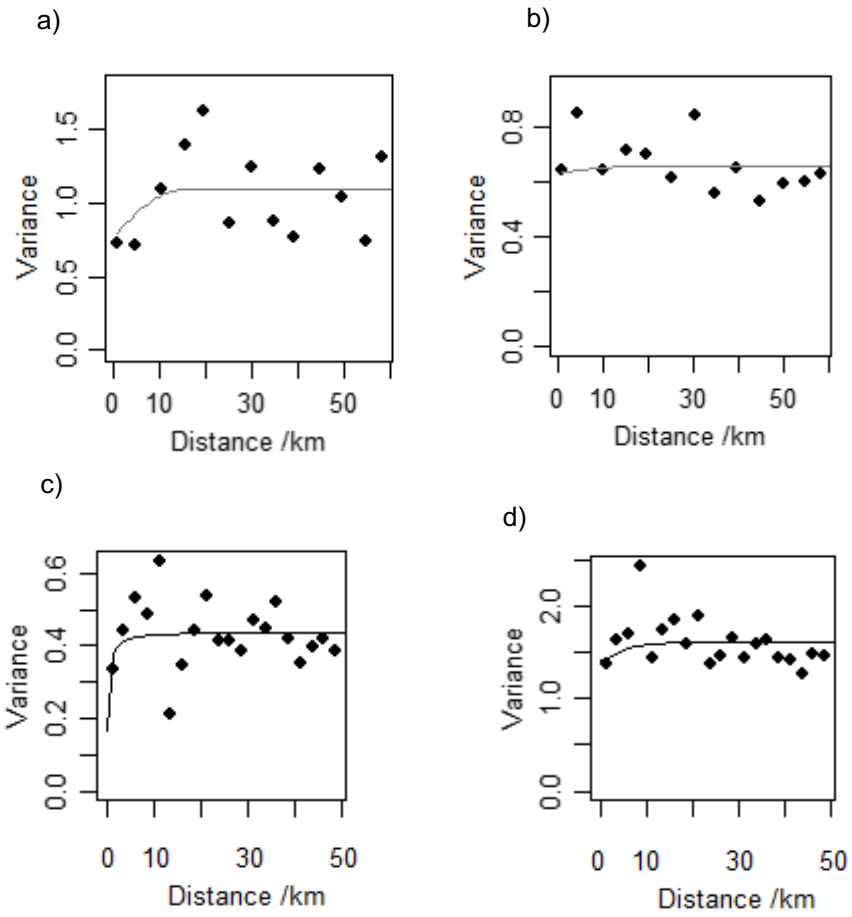


Figure 2.9 Variograms for predicted global warming potential across the sampling area (East Midlands of England) from (a) zero and (b) conventionally tilled soils and the variance for associated predicted carbon stocks from (c) zero and (d) conventionally tilled soils. Note, the variance (y-axis) is logged. Note that the variogram models are estimated by residual maximum likelihood and are not fitted to the point estimates which are shown for illustrative purposes.

3.0 Zero-tillage impacts on soil physical and biological properties in temperate soils

The previous chapter demonstrated the temporal trend of greenhouse gas fluxes from zero-tilled soils. This chapter provides a detailed characterisation of soil quality and microbial activity in the soil from the East Midlands, U.K., assessing whether this temporal trend was observed in other soil characteristics across two seasons. A subset of the samples from Chapter 2 were also used for assessment for one season. A selection of the previously sampled fields was chosen for additional sampling and subsequent analysis in July 2016.

3.1 Introduction

Soil quality is one of the three components that define environmental quality, alongside water and air quality (Andrews et al., 2002). There have been many definitions of soil quality since the term was introduced in 1971 by Mausel, but a comprehensive definition states, it is "the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and support human health and habitation" (Karlen et al., 1997). Soils have both inherent and dynamic qualities (Dominati et al., 2010), where inherent soil qualities relate to a soils natural ability to function and thus not easily changed, e.g. texture and depth of soil profile (Tresch et al., 2018). Dynamic soil qualities depend on how soil is managed, and can be altered, e.g. soil organic matter content, soil structure, water and nutrient holding capacity etc. (Bertrand et al., 2015). Soils

alter differently to management systems depending on the inherent properties of the soil and the neighbouring landscape, and a change in management practice often leads to differences in biological, chemical and physical properties of soil, thereby altering soil functional quality (Bünemann et al., 2018; Celik et al., 2011; Ding et al., 2011).

Conventional tillage is a traditional method for crop production, in which soil is prepared for planting by complete inversion, typically with a mouldboard plough, to a depth of 20–30 cm (OECD, 2001). Many studies report that this practice contributes negatively to soil quality through the degradation of soil structure and decreasing soil organic matter and nutrient content (Kladivko, 2001; Montgomery, 2007). Conventional tillage damages soil pore continuity and aggregate stability resulting in sediment mobilisation, erosion and surface hardening (Dörner et al., 2010; Hamza and Anderson, 2005). In turn, aggregates which are exposed to physical disruption break apart, which enhances the accessibility of organic matter to micro-organisms, stimulating oxidation and the loss of organic matter (Devine et al., 2014). In contrast, zero-tillage, where the seed is sown directly into the soil, causing minimal disturbance and retains surface crop residue, has been adopted worldwide since the 1940s (Derpsch et al., 2010). This management system is perceived by some to be an efficient method of sustainable agriculture that increases soil quality, and saves fuel and time (Triplett and Dick, 2008). Estimates from Basch et al., (2012) suggest a 44.2 L ha^{-1} yr^{-1} saving in fuel, which would equate to 133.9 kg CO_2 ha^{-1} yr^{-1} reduction of CO_2 emissions. Smith et al., (1998) suggested a 100% conversion to zero-tillage in Europe could mitigate all fossil fuel-carbon emissions from

agriculture in Europe. Assessments from the United States Department of Agriculture (Creech., 2017) state if a farmer ploughs 15 acres per hour, they will save approximately 67 hours of work with each eliminated pass over a 1,000-acre field by changing from conventional to zero-tillage.

Soils can react slowly to changes in land use and management, and therefore it can be challenging to detect differences in soil quality before substantial damage has occurred (Bünemann et al., 2018). Traditionally, due to constraints of analytical techniques and sampling strategies, soil quality studies have primarily focused on chemical and physical characteristics of soil (Larson and Pierce, 1994), but in the last decade, it has been reported that biological characteristics of soil act as early and sensitive indicators in response to a change in land use and management (Aziz et al., 2013; Islam and Weil, 2000). Consequently, biological properties alongside soil chemical and physical characteristics are required to evaluate soil quality as affected by changes in tillage operations (Aziz et al., 2013).

Previously, most studies focused on agricultural land under zero-tillage management have considered those that converted ten years ago or fewer (Ding et al., 2011; Kainiemi et al., 2013; Mangalassery et al., 2014), with long-term studies typically viewed as those with five years after conversion from conventional tillage. However, evidence suggests the duration of zero-tillage management impacts on soil quality, with Díaz-Zorita et al., (2004) identifying significant changes in structural properties in soils that had been in zero-tillage for 20 years. Mathew et al., (2012) described a greater soil carbon and nitrogen

content in the soil surface for soil which had been in zero-tillage for 18 years. Nutrient availability and microbial activity can also vary under zero compared with conventional tillage due to surface placement of crop residues rather than mechanical mixing within the plough layer (Franzluebbers and Hons, 1996). In zero-tillage, slower decomposition of surface crop residues may prevent leaching of nutrients down through the soil profile, which is more likely when residues are incorporated into the soil profile (Lupwayi et al., 2007), but the extent to which long-term zero-tillage alters nutrient content and microbial activity throughout the soil profile is less understood.

This chapter presents data from an extensive sampling campaign including agricultural fields which had been in zero-tillage for 1–15 years with adjacent conventionally tilled fields for comparison. The aim was to investigate the differences in soil qualities between two contrasting soil tillage systems, assessing impacts down to 50 cm, and outline the potential implications on soil quality and for farmers. The overarching hypothesis was that long-term zero-tillage practices (implemented for 15 years) would improve soil quality relative to paired conventionally tilled practices and recently converted zero-tilled soils.

3.2 Methods

3.2.1 Site selection and sample collection

A subset of the samples utilised in Chapter 2 was also used for this data chapter. Furthermore, a selection of the previously sampled fields was chosen for additional sampling and subsequent analysis in July 2016 (Table 3.1). As in Chapter 2, all sampling sites comprised pairs of conventionally tilled farms and

farms where zero-tillage practices were followed. GPS coordinates from the previous sampling campaign were used to ensure soil samples were collected from the same sites as previous work. From each location, bulk soil samples were collected using an auger at four depths (0-10, 10-20, 20-30 and 30-40 cm) and intact soils cores were taken down to 30 cm. The sampling was replicated in random locations three times at each site. In the laboratory, the field moist samples were composited by mixing the replicates. They were then separated for different analyses and stored as required. Samples for assessing enzyme activity were frozen at -20 °C and were samples were thawed at 4 °C before analysis over five days. The intact soil cores and a subset of auger soil samples were retained at 4 °C until analysis of soil biophysical analyses.

Table 3.1 The sub-set of paired conventionally and zero-tilled (ZT) fields sampled in July 2016. Site numbers refer to the experiment in chapter 2.

Site	Soil texture	Years in ZT	Site	Soil texture	Years in ZT
31	Sandy Loam	1	**6**	Sandy loam	6
61	Clay	2	**7**	Sandy loam	6
62	Clay	2	**35**	Clay	15
63	Sandy silt loam	2	**36**	Clay	15
66	Clay	1	**37**	Clay	15
67	Clay	1	**38**	Clay	15
1	Sandy loam	6	**44**	Sandy silt loam	15
2	Sandy clay	6	**46**	Sandy silt loam	15
3	Sandy clay	6			
4	Clay	6			

3.2.2 Penetration Resistance

A cone penetrometer (Rimik CP40) was used to measure the soil penetration resistance encountered at various depths, to a maximum depth of 60 cm. The probe was pushed into the ground and the force recorded by the electronic load cell to calculate the cone index, which is a relative indicator of the soil's strength. For accurate readings, the penetrometer was inserted into the ground at a steady speed (c. 30 mm/s). This was repeated in triplicate at each sampling location. This measurement could not be taken during the sampling in July 2016 due to excessive soil hardness.

3.2.3 Soil water content

Field moist samples collected using the auger were used to calculate gravimetric soil moisture content. This was measured by oven drying soil samples in a foil tin at 105 °C for 8 hours:

$$MC\ \% = \frac{[W_2 - W_3]}{[W_3 - W_1]} \times 100, \qquad \text{(Equation 3.1)}$$

where W_1 is the weight of the foil tin (g), W_2 is the weight of field moist soil in the foil tin (g), and W_3 is the weight of the dry soil and foil tin (g). Gravimetric soil moisture was calculated down to 50 cm for Nov-Dec 2015 samples, but due to soil hardness was only calculated to 40 cm for July 2016 samples.

3.2.4 Aggregate stability

Loose soil samples from each field (again, down to 50 cm in 10 cm increments for Nov-Dec 2015 and to 40 cm in July 2016 sampling) were air-dried for several days and gently broken apart by hand into smaller aggregates during drying. The soils were then sieved, and aggregates sized between 3-5 mm retained for analysis. Five g of calibrated soil aggregates were gently immersed in 50 ml deionised water for 10 minutes, and the water was siphoned off using a pipette and the soil material transferred to a 50 μm sieve (which had previously been submerged in methylated spirit to avoid re-aggregation of the soil sample and to reduce the amount of additional breakdown). The 50 μm sieve was gently moved up and down in a beaker containing methylated spirit for 1 minute, and care was taken to avoid the methylated spirit from flowing into the top of the sieve to avoid further disruption of the soil aggregates. This first step separated fragments < 50 μm from those > 50 μm. Once the methylated spirit evaporated, the sieve containing the remaining sample was placed in an oven for 8 hours at 105 °C. After drying, the sample was gently sieved through a stack of six sieves (2000, 1000, 500, 200, 100, and 50 μm). The soil mass remaining on each sieve was recorded, and the mass percentage in each size fraction calculated. The aggregate stability for each breakdown mechanism was expressed as the mean weight diameter (MWD):

$$MWD = \sum_{i=l}^{n} xi\ wi, \qquad \text{(Equation 3.2)}$$

where wi is the proportion of the sample with a mean size of xi μm (van Bavel, 1950).

3.2.5 Water repellency

Water repellency was estimated using the water drop penetration time (WDPT) test. The WDPT test consists of applying ten water drops (100 ± 5 µl) from a consistent height (30 cm), onto the air-dried soil surface, and measuring the amount of time (s) it takes for the water droplet to infiltrate the soil. The amount of time it took for the water droplet to infiltrate was used to estimate water repellency (Table 3.2).

Table 3.2 Waterdrop penetration time (WDPT) test, time (s) it takes for a water droplet to infiltrate the soil can provide water repellency.

Water repellency	WDPT (s)
Wettable	<5
Slightly to moderately repellent	5-60
Strongly water-repellent	60-600
Severely water-repellent	600-3600
Extremely water-repellent	>3600

3.2.6 Particle size analysis

Particle size analysis was performed using the hydrometer method. Soil pre-treatment was carried out by adding 100 ml of 6% H_2O_2 to 50 g of <2 mm sieved air-dry soil and left to stand overnight. The following morning, 100 ml of sodium hexa metaphosphate $(NaPO_3)_6$ was added, and soil suspension made up to 1-litre mark with distilled water. A blank sample with no soil suspension was also included. The temperature of the suspension was measured, and then the plunger

inserted and moved up and down to mix. The hydrometer was lowered and read at 32 seconds to give the value for silt and clay. The contents were mixed again and allowed to stand for 8 hours, before recording clay content. A density correction was applied by subtracting the density of the blank at each reading, from the corresponding density readings for the samples. 0.2 was added to the readings of the samples for every 1ºC above 20 ºC and 0.2 subtracted for every 1 ºC below 20 ºC. Equation 3.3 and 3.4 were used to calculate the percentage of clay and silt.

$$\%\ clay = \frac{Corrected\ hydrometer\ reading\ at\ 8\ hrs}{wt\ of\ soil\ sample\ on\ OD\ basis} x100, \qquad \text{(Equation 3.3)}$$

$$\%\ silt \equiv \left(\frac{Corrected\ hydrometer\ reading\ at\ 32\ seconds}{wt\ of\ soil\ sample\ on\ OD\ basis} x100\right) - \%\ clay, \qquad \text{(Equation 3.4)}$$

3.2.7 Greenhouse gas fluxes

Potential GHG fluxes were measured following incubations under controlled conditions. This approach was adopted to allow comparison between management by removing the effects of climate on gas production *in-situ*. Intact soil cores were removed from the 4 °C environment, and water-filled pore space was adjusted to 60% (following equation 3.5), and then kept at 5 °C for 48 hours to activate and stabilise the soil microflora.

$$WFPS = \frac{(Soil\ water\ content\ x\ Bulk\ density}{(1-(Bulk\ density\ /\ Particle\ density))} \qquad \text{(Equation 3.5)}$$

Each soil core was incubated in a plastic jar (1.5 dm^3) with a septum on the top to allow gas sampling. The air in the headspace was mixed via a magnetic stirrer during gas sampling at time intervals 0, 15, 30 and 60 minutes using a 20 mL syringe. The collected gas samples were stored in pre-evacuated 12 mL airtight glass vials. Gas samples were analysed for concentrations of CO_2, CH_4 and N_2O by gas chromatography (GC-2014, Shimadzu) and potential GHG flux data were converted to mass per volume and mass per weight basis using the ideal gas law (Equation 3.6) (Laugier and Garai, 2009).

$$PV = nRT, \qquad \text{(Equation 3.6)}$$

where P is the atmospheric pressure (≈ 1 atm), V is the volume of headspace (dm^{-3}), n is the number of moles of gas, R is the ideal gas constant (0.08205746 L atm K^{-1} mol^{-1}), and T is the temperature (273 + room temperature in °C).

From this, the potential flux of each gas was calculated (Equation 3.7):

$$E = \frac{nm}{at} \, x \, 1000, \qquad \text{(Equation 3.7)}$$

where E is the flux of each has (mg m^{-2} h^{-1}), n is the number of moles (CO_2, CH_4 or N_2O), m is the molar weight (CO_2 and N_2O is 44.01 and CH_4 is 16.04), a is the area of the soil core measured and t is the time for the incubation, in hours.

Global warming potential (GWP) was calculated in CO_2-equivalents following equation 3.8 (Myhre et al., 2013):

$$GWP = \left\{\left(\frac{CO_2\ x\ 44}{12}\right) + (CH_4\ x\ 23) + (N_2O\ x\ 296)\right\}, \quad \text{(Equation 3.8)}$$

This method was repeated at 10 and 15°C on the intact soil cores collected in Nov-Dec 2015 and at 25 °C on intact soil cores collected in July 2016.

At the end of the incubations Q_{10} values were calculated for the soil cores incubated at 5, 10 and 15 ºC. This value describes the increase in respiration rate with a 10 °C increase in temperature and was calculated using equation 3.9 with k being the rate constant (Davidson et al., 2006).

$$Q_{10} = e^{10k}. \quad \text{(Equation 3.9)}$$

3.2.8 Ammonium and nitrate

For the determination of ammonium (NH_4-N), 6 g field moist surface (0-10 cm) soil was extracted in 40 ml 2M KCl. Samples were placed for one hour on a rotary shaker, centrifuged at 2500 rpm for 5 minutes and then filtered through Whatman 42 filter paper. The concentration of NH_4-N in the extracts was determined colourimetrically (Kempers, 1974) and 8 ml of filtrate was mixed with 1 ml of phenol-nitroprusside, 1 ml of hypochlorite and 2.5 ml of de-ionised water to form a blue indophenol complex in solution. Each solution was placed

in an incubator at 25°C for 1 hour and then absorbance measured in a spectrophotometer at 635 nm.

For the measurement of nitrate-nitrogen (NO_3^--N), 5 g field moist surface (0-10 cm) soil was extracted in 40 ml de-ionised water. Samples were placed for 16 hours on a rotary shaker; centrifuged at 2000 rpm for 15 minutes and then filtered through 22 µm microfilters. NO_3^--N in the extracts was determined through Ion Chromatography.

3.2.9 pH, EC and redox potential

The pH, electrical conductivity (EC) and redox potential were determined by adding 5 g of fresh soil to 12.5 ml Milli-Q water, in triplicate, in a 50 ml centrifuge tube (Fisher Scientific). pH, redox potential (HANNA instrument, 209, pH and redox meter) and EC (HANNA, HI9835 Microprocessor Conductivity/TDS meter) readings were taken after placing on a rotary shaker for 30 minutes and solution heated to 25°C.

3.2.10 Microbial biomass carbon and nitrogen

Field moist soil samples (0–10 cm) were used for the determination of microbial biomass carbon (MBC) and nitrogen (MBN) by the chloroform fumigation method (Vance et al., 1987). In brief, 10 grams (0.5g) of field moist samples were incubated in a chloroform environment for 24 hours. Both fumigated and non-fumigated control soil samples were extracted with 60 ml of 0.5 M K_2SO_4, centrifuged, filtered and then analysed using a Shimadzu CN analyser (TOC-V CPH Shimadzu) and MBC/N was determined using equation 3.10.

$$C_m = \left\{\left(\frac{TOC_F}{W_{soil\,F}}\right) - \left(\frac{TOC_B}{W_{soil\,B}}\right)\right\}\ x\ \left(\frac{V_{ext}}{k_{EC}}\right), \qquad \text{(Equation 3.10)}$$

where C_m is the microbial biomass carbon (mg C kg^{-1} soil), TOC_F is the total organic carbon measured in the fumigated soil extract (µg mL^{-1}), TOC_B is the total organic carbon measured in the blank soil extract (µg mL^{-1}), V_{ext} is the volume of K_2SO_4 extract (mL), and W_{soil} is the dry weight equivalent of soil for fumigated (*soil F)* or blank (*soil B)* soil. The value of the coefficient K_{EC}, to convert 'chloroform-labile' carbon to MBC of 0.45 was used (Jenkinson et al., 2004) and for microbial biomass nitrogen (N_m) the value of K_{EN} was taken as 0.54 (Brookes et al., 1985).

3.2.11 Enzyme assays

Dehydrogenase activity was determined using an approach modified by Ohlinger (1995), originally from Thalman (1968). The activity within the soil samples is measured colourimetrically, by the reduction of 2,3,5-triphenyltetrazolium chloride (TPC) to triphenyl formazan (TPF). The intensity of TPF was measured at 546 nm against known TPF standards and was expressed as µg TPF g^{-1} day^{-1}.

Both cellulase and xylanase activity was measured by the reduction of potassium hexacyanoferrate (III) to potassium hexacyanoferrate (II). For cellulase activity, the reduced potassium hexacyanoferrate (II) was reacted with ferric ammonium sulphate in an acid medium to form a coloured complex of ferric

hexacyanoferrate (II), using carboxy-methyl cellulose as a substrate. For xylanase activity, the same reaction occurred but using xylan as a substrate. Both colour intensities were assessed at 690 nm and expressed as mg GE g^{-1} day^{-1} (Schinner and Vonmersi, 1990).

3.2.12 Statistical analysis

For the samples collected in Nov-Dec 2015, the same model-based statistical approach was used as previously in Chapter 2. For samples collected in July 2016, a two-way analysis of variance was applied to results obtained from laboratory analysis with duration under zero-tillage or soil texture and management as the two factors. The treatment means were compared at $p < 0.05$ and $p < 0.01$. Standard errors of the means were calculated and provided as required. All analysis was conducted in RStudio V 1.2.5019.

3.3 Results

3.3.1 Penetration resistance

Penetration resistance between 0–5 cm was larger in soils under zero-tillage than under conventional cultivation ($F_{1,158} = 32$, $p<0.001$, Fig. 3.1). When the soils under zero-tillage were grouped by length of time under this management, penetration resistance was largest for the soils with the longest history of zero-tillage ($F_{5,154} = 7,7$, $p<0.001$, Fig. 3.1c). The average penetrometer resistance for conventionally-tilled soils was 529 kPa, whereas that for soils under zero-tillage for 11–15 years was 881 kPa. A similar pattern was observed at depth interval 10–15 cm, ($F_{5,154} = 18$, $p<0.001$), and the average penetrometer resistances of soils under conventional and zero-tillage for 11–15 years was 912 kPa and 1481

kPa respectively. At 30–35 cm interval, there was no significant difference between the managements, averaging 1585 kPa ± 59 and 1631 ± 55 in zero and conventionally tilled soils respectively ($F_{5,134}$ = 0.9, p>0.05, Fig. 3.1). At depth intervals below 40 cm, penetration resistance was smallest for the soils with the longest history of zero-tillage, 1409 kPa compared to 1669 kPa in the paired cultivated soils ($F_{5,122}$ = 12, p<0.001, Fig. 3.1d). This analysis could not be carried out in July 2016 due to excessive soil hardness.

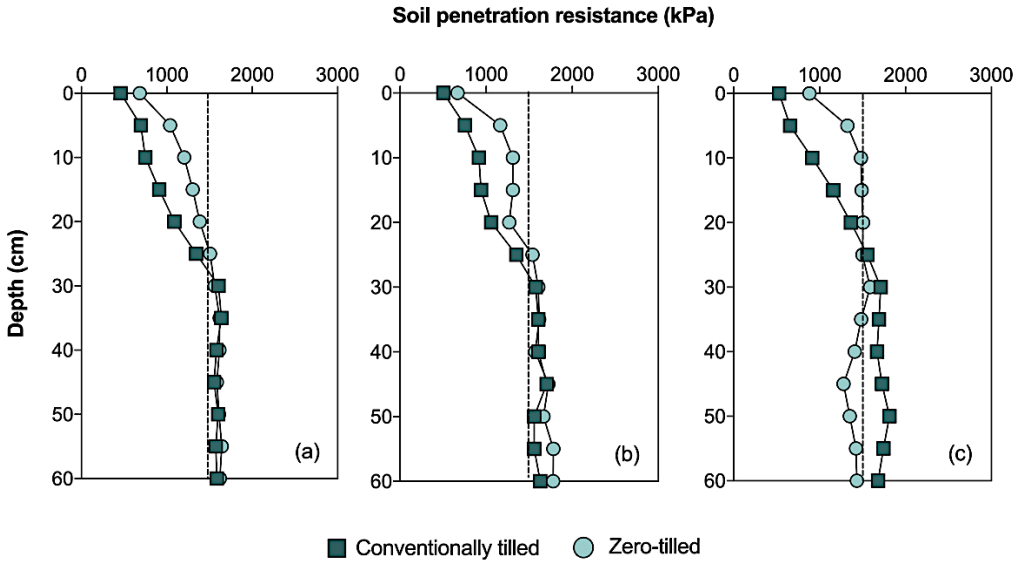


Figure 3.1 The soil penetration resistance (kPa) in zero and conventionally tilled soils to a depth of 60 cm in Nov-Dec 2015. The soils had been in zero-tillage for (a) 1-5 years, (b) 6-10 years, and (c) 10-15 years. The dotted line indicates 1500 kPa. Error bars are not shown as they are smaller than the symbols, see Table B1 in Appendix B for means and standard errors of the means.

3.3.2 Aggregate stability

Aggregate stability in the cultivated and recently adopted zero-tilled soils did not significantly differ from each other for all depth intervals. When the soils,

under zero-tillage, were adopted for longer than six years, aggregate stability was significantly greater than conventionally tilled soils at intervals 0-10 and 10-20 cm ($F_{5,154}$ = 14, p<0.001, Fig. 3.2).

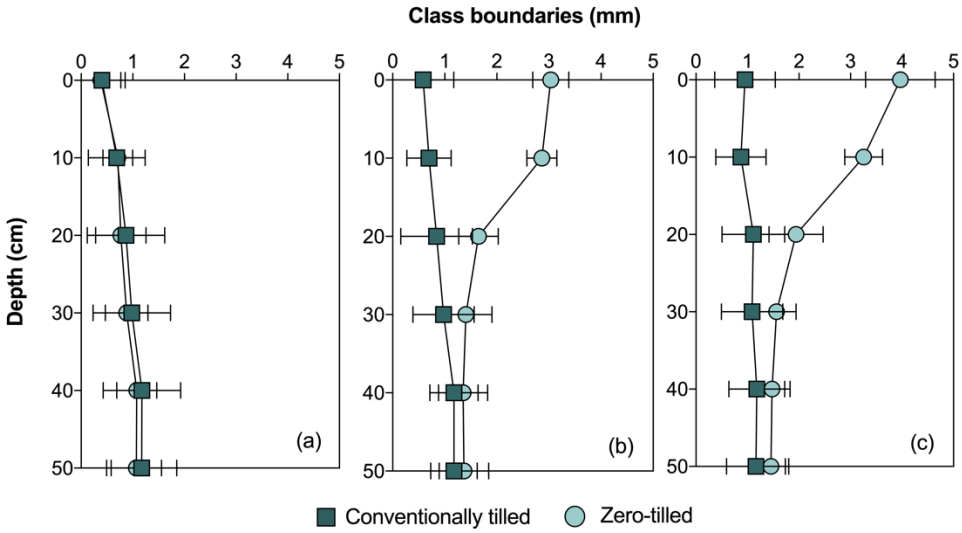


Figure 3.2 Aggregate stability from zero and conventionally tilled soils down to a depth of 50 cm. The soils had been in zero-tillage for (a) 1-5 years, (b) 6-10 years, and (c) 10-15 years. Means ± one standard error.

In the cultivated and recently converted zero-till soils, aggregate stability was classed as very unstable (Table 3.3) in the 0-10 cm interval, but at depths below 10 cm, aggregate stability was classified as being 'median' stability (Table 3.3). Soils which had been in zero-tillage for six years or longer contained very stable aggregates in the top 20 cm. Below 20 cm, these aggregates were classed as either median or stable.

Table 3.3 The boundaries of mean weight diameter indicating aggregate stability.

Class boundaries	Mean weight diameter (mm)
Very unstable	< 0.4
Unstable	0.4 – 0.8
Median	0.8 – 1.3
Stable	1.3 - 2
Very stable	> 2

3.3.3 Water repellency

The duration of tillage management did not have a significant impact on soil water repellency from samples collected in Nov-Dec 2015 or July 2016. However, soil texture significantly impacted water repellency between the two managements (Figure 3.3).

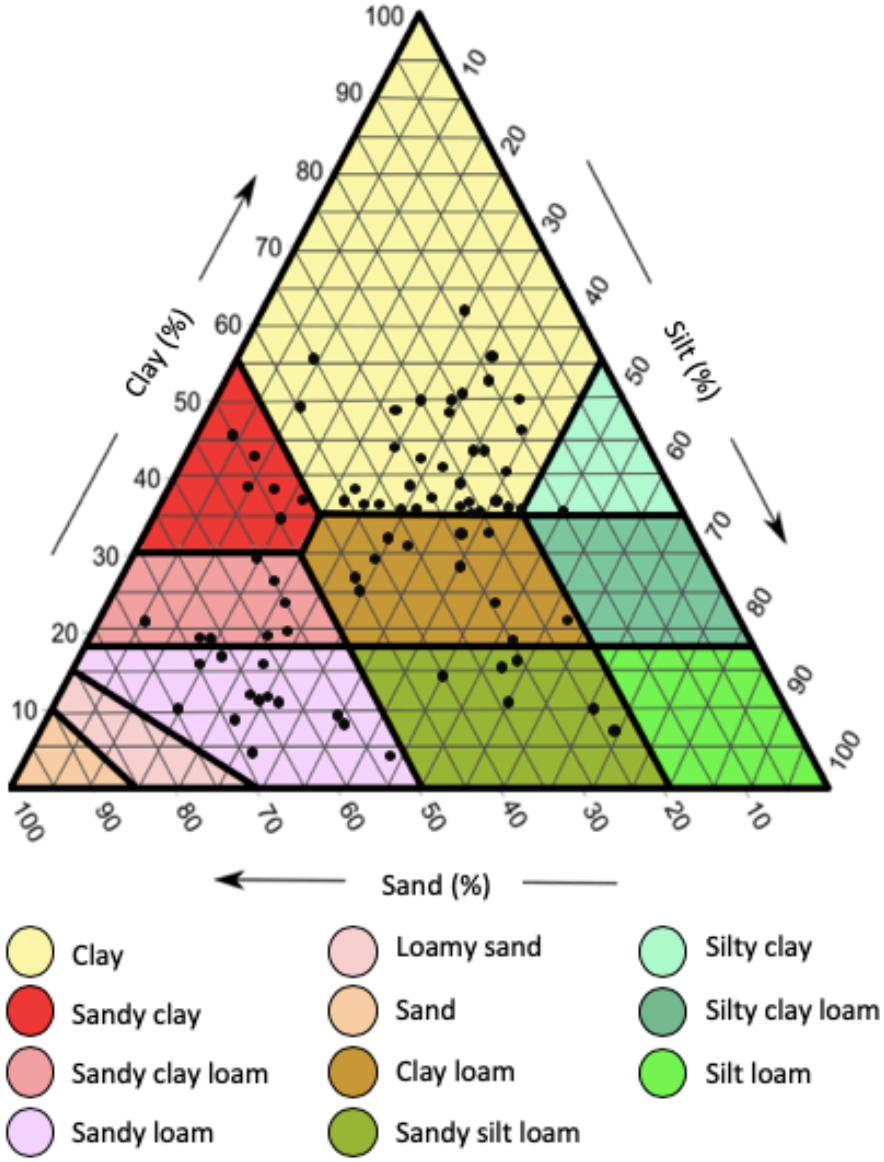


Figure 3.3 Soil texture classes of soil samples across the East Midlands according to the soil survey of England and Wales (Hodgson, 1997). Particle size analysis was measured using the hydrometer method with three replicates per site.

For samples collected in Nov-Dec 2015, there were no significant differences between the tillage managements in the fine and moderately fine-textured soil.

However, there were significant differences between tillage practices in the coarse-textured soils ($F_{7,152}$ = 49, p<0.001, Fig. 3.4a). In the coarse-textured zero-tilled soils (n=20), mean infiltration rate was 42 seconds, compared to 11 seconds for conventionally tilled soils. These measurements suggest that coarse-textured soils (regardless of tillage management) were classed as 'slightly to moderately repellent' compared to being classed as 'wettable' in the fine and moderately fine-textured soils (Table 3.2).

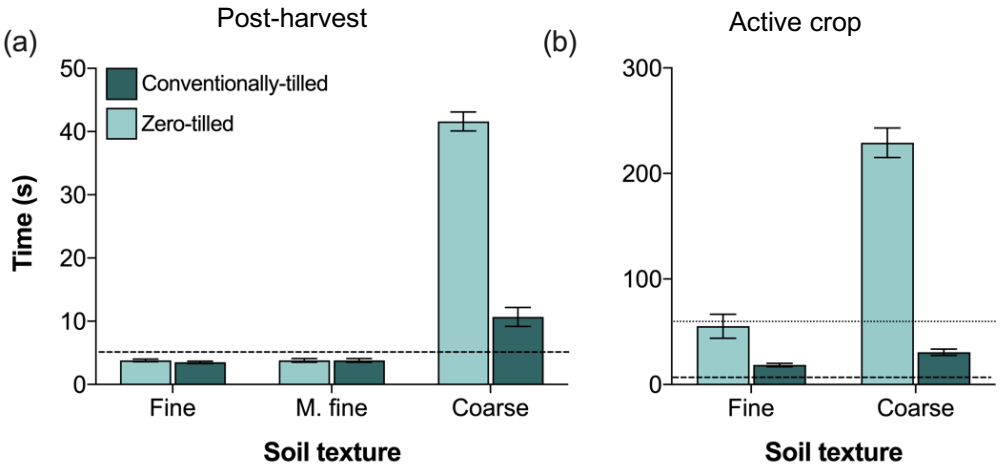


Figure 3.4 Water repellency in soils from (a) Nov-Dec 2015 sampling, n=80 and (b) July 2016 sampling, n=18. Water repellency was estimated using the Water Drop Penetration Time (WDPT) method. Dotted lines denote 5s (≤ 5s – soil is 'wettable') and 60s (5-60s - soil is 'slightly to moderately repellent, and 60-600s – soil is 'strongly water-repellent') and M. fine refers to moderately fine soil texture. Means ± one standard error.

A similar trend occurred in the coarse soils (n=7) which were collected in July 2016, but also in the fine-textured soils (n=11) ($F_{1,16}$ = 69, p<0.001, Fig. 3.4b), with a mean of 19 and 55 seconds for water droplets to infiltrate in the fine zero-

till and cultivated soils respectively. In this soil textural class, both managements were categorised as 'slightly to moderately repellent'. In the coarse-textured soils, conventionally tilled soils were classed as 'slightly to moderately repellent', whereas zero-tilled soils were categorised as 'strongly water-repellent' (Fig. 3.4b).

3.3.4 Greenhouse gas fluxes

At 5 °C, there was no significant differences in GHG emissions or GWP between different tillage managements ($p>0.05$, Fig. 3.6a, e). However, when the soil cores were incubated at 10 °C, soil tillage practices significantly influenced CO_2 fluxes with greater fluxes from soils under conventional tillage (213 mg CO_2 m^{-2} h^{-1}) compared to zero-tillage (117 mg CO_2 m^{-2} h^{-1}) ($F_{1,158} = 6$, $p<0.05$, Fig. 3.5f). Similarly, there was a significant difference in CH_4 fluxes between the two tillage practices ($F_{1,158} = 5$, $p<0.05$). Conventionally tilled soils were a small source of CH_4 (0.0125 mg CH_4 m^{-2} h^{-1}) compared to zero-tilled soils which were a minor sink (-0.0779 mg CH_4 m^{-2} h^{-1}). Overall the net GWP calculated was significantly greater from conventionally tilled soils (807 mg CO_2 eq. m^{-2} h^{-1}) compared to zero-tilled soils (486 mg CO_2 eq. m^{-2} h^{-1}) ($F_{1,158} = 6$, $p<0.05$, Fig. 3.5c).

When the intact soil cores were incubated at 15 °C, N_2O fluxes were significantly greater from zero-tilled soils, averaging 0.118 mg N_2O m^{-2} h^{-1} compared to 0.085 mg N_2O m^{-2} h^{-1} from cultivated soils ($F_{1,158} = 4.8$, $p<0.01$). In contrast, both CH_4 and CO_2 were significantly greater from cultivated soils. When the soils under zero-tillage were grouped by length of time under zero-

tillage, CO_2 fluxes and GWP were lowest for soils with the longest history of zero-tillage. A similar trend was observed at 25 °C, with a mean CO_2 flux of 172 mg CO_2 m^{-2} h^{-1} from zero-tilled soils compared to 307 mg CO_2 m^{-2} h^{-1} from conventionally tilled soils ($F_{2,15}$= 16.8, $p<0.01$, Fig. 3.5h). Total GWP was lowest for the soils with the longest time under zero-tillage ($F_{2,15}$= 42, $p<0.01$, Fig. 3.5d).

Temperature strongly influenced CO_2 production, with potential CO_2 fluxes most temperature-sensitive in the conventionally tilled soils, with a Q_{10} value of 1.6 (±0.4) compared to 1.2 (±0.3) in zero-tilled soils. When the soils under zero-tillage were grouped by length of time under this management, the Q_{10} value for CO_2 fluxes was its lowest at 1.1 (±0.2), compared to the paired conventionally tilled soils which had a Q_{10} value which was almost double at 2.03 (±0.2).

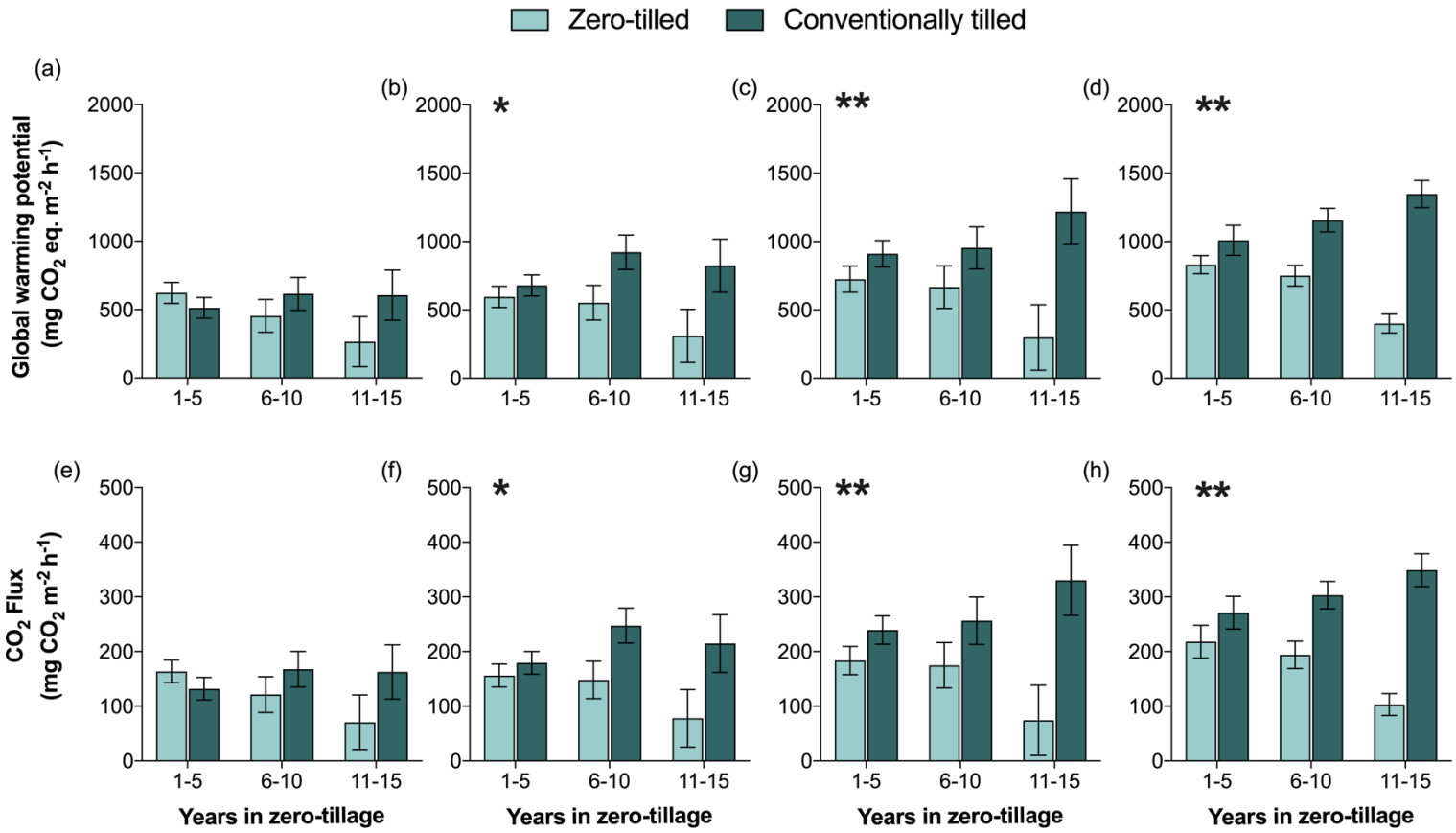


Figure 3.5 Global warming potential (GWP) (a-d), and carbon dioxide (CO_2) emissions (e-f) from zero and conventionally tilled soils sampled in Nov-Dec 2015 and July 2016. Soil cores were incubated at (a&e) 5 °C, (b&f) 10 °C, (c&g) 15 °C and (d&h) 25 °C. Means ± one standard error. * indicates tillage management significance at $p<0.05$ and ** indicates tillage management and duration of management significance at $p<0.05$.

3.3.5 Soil enzyme assays

3.3.5.1 Dehydrogenase

Zero-tilled soils had a greater dehydrogenase activity (29.7 μg TPG g^{-1} day^{-1}) compared to cultivated soils collected in Nov-Dec 2015 (24.2 μg TPG g^{-1} day^{-1}) ($F_{1,158} = 71$, $p<0.001$, Fig. 3.6a). When the soils under zero-tillage were grouped by length of time under this management dehydrogenase activity significantly increased for the soils with the longest history of zero-tillage ($F_{5,154} = 29$, $p<0.001$, Fig. 3.6a). The same trend was observed in soil samples taken in July 2016 ($F_{2,29} = 31$, $p<0.001$, Fig. 3.6b).

3.3.5.2 Cellulase

Similar to dehydrogenase, zero-tilled soils exhibited significantly increased cellulase activity ($F_{1,158} = 23$, $p<0.001$, Fig. 3.6c) with mean values of 0.35 mg GE g^{-1} day^{-1} compared to 0.16 mg GE g^{-1} day^{-1} in cultivated soils. The length of time under management also significantly increased cellulase activity in zero-tilled soils ($F_{5,154} = 8$, $p<0.001$, Fig. 3.6c), increasing from 0.26 mg GE g^{-1} day^{-1} in recently converted soils (1-5 years) compared to 0.73 mg GE g^{-1} day^{-1} in the soils with the longest history of zero-tillage. For soil samples collected in July 2016, tillage management had a significant effect on cellulase activity, with mean values of 0.39 mg GE g^{-1} day^{-1} in cultivated soils compared to 0.67 mg GE g^{-1} day^{-1} in zero-tilled soils ($F_{1,29}=13$, $p<0.001$, Fig. 3.6d). There was no significant effect of length under management from this sampling time point (July 2016).

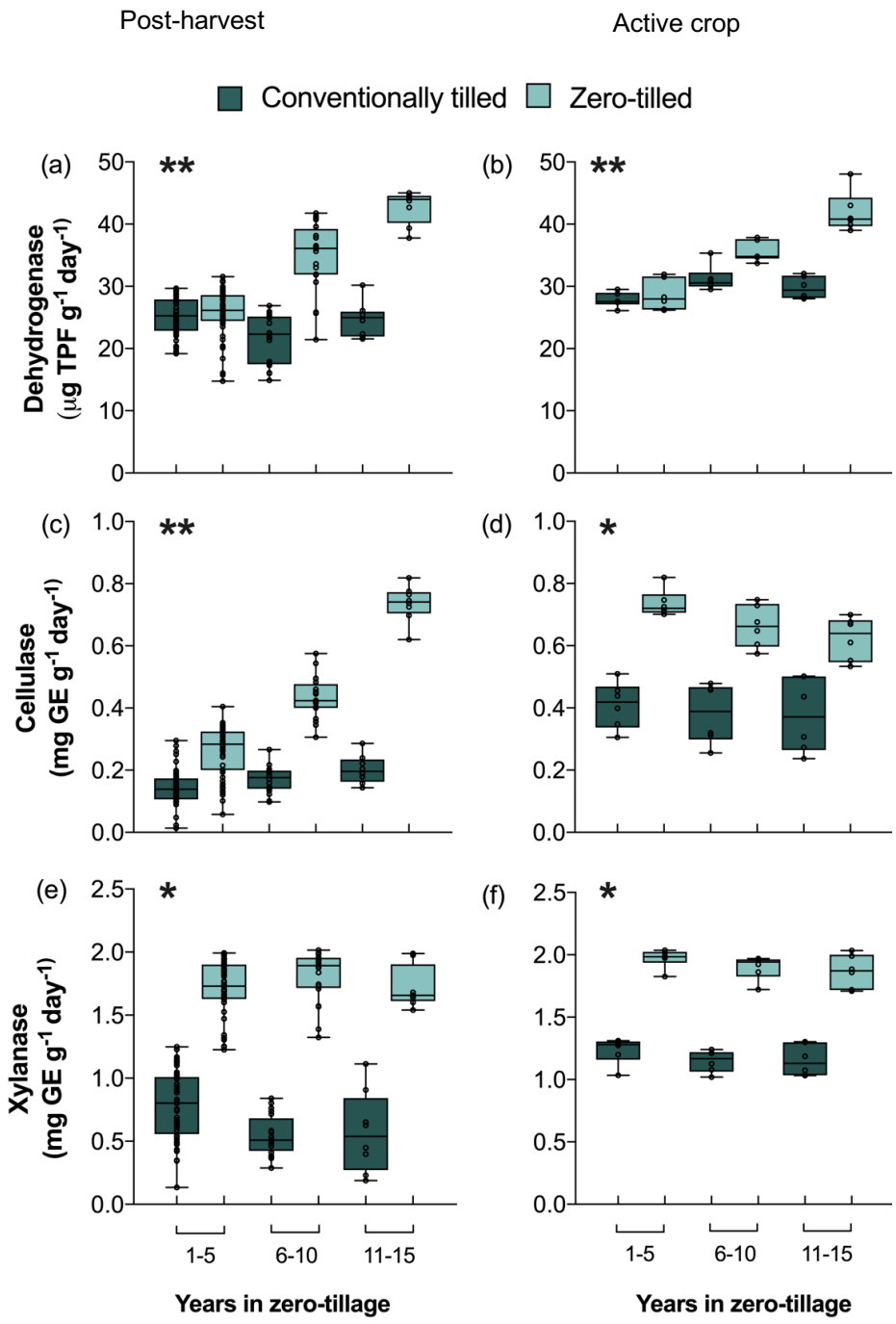


Figure 3.6 Soil enzyme assays at 0-10 cm under zero and conventionally tilled soils which were sampled in Nov-Dec 2015 (a,c,e) and July 2016 (b,d,f). The enzymes measured were (a-b) dehydrogenase, (c-d) cellulase and (e-f) xylanase. Means ± one standard error. * indicates tillage management significance at

$p<0.05$ and ** indicates tillage management and duration of management significance at $p<0.05$.

3.3.5.3 Xylanase

Tillage also significantly influenced the xylanase activity in zero-tilled soils with a mean of 1.74 mg GE g^{-1} day^{-1} compared to 0.7 mg GE g^{-1} day^{-1} in tilled soils ($F_{1,158}$ = 10, $p<0.001$, Fig. 3.6e). The same effect was observed in the soils collected in July 2016, as xylanase activity was 0.7 mg GE g^{-1} day^{-1} greater in zero-tilled soils compared to conventionally tilled soils ($F_{1,15}$=6, $p<0.05$, Fig. 3.6f). There was no significant impact of duration of tillage management on xylanase activity from soils collected at either time point.

Table 3.4: Means, standard errors and statistical output from linear mixed modelling (management and duration, i.e. time since conversion) for the physio-chemical characteristics of soils under zero (ZT) and conventional tillage (CT) from samples taken in (a) Nov-Dec 2015 and (b) July 2016. Mean ± standard error of the mean. ns: not significant, * $p<0.05$, ** $p<0.01$.

(a) Nov-Dec 2015, n=80				
Variable	ZT	CT	Management	Duration
Microbial C (mg kg^{-1} soil)	497 ± 27.1	425.5 ± 27.1	6.1*	5.81*
Microbial N (mg kg^{-1} soil)	46.7 ± 4.2	33.0 ± 4.2	15.4**	ns
pH	7.14 ± 0.14	6.99 ± 0.14	ns	ns
EC (dSm^{-1})	5.81 ± 3.2	4.84 ± 3.2	7.6**	ns
Ammonium (mg/kg^{-1})	32.7 ± 3.38	26.5 ± 3.37	6.5*	5.4*
Nitrate (mg/kg^{-1})	15.4 ± 1.2	10.7 ± 1.1	8.9**	7.2**
GWC (%) (0-10 cm)	38.6 ± 1.57	36.6 ± 1.56	4.7*	4.3*
GWC (%) (10-20 cm)	29.9 ± 6.0	30.2 ± 5.9	ns	ns
GWC (%) (20-30 cm)	27.7 ± 1.3	27.3 ± 1.2	ns	ns
GWC (%) (30-40 cm)	26.7 ± 1.2	26.9 ± 1.2	ns	ns
GWC (%) (40-50 cm)	26.0 ± 2.0	24.6 ± 2.0	ns	ns

(a) July 2016, n=18				
Variable	ZT	CT	Management	Duration
Microbial C (mg kg^{-1} soil)	210 ± 30.1	140 ± 24.3	11.9**	9.3*
Microbial N (mg kg^{-1} soil)	28.6 ± 4.1	18 ± 3.1	9.2*	ns
pH	7.13 ± 0.2	7.00 ± 0.3	ns	ns
EC (dSm^{-1})	6.12 ± 7.4	5.24 ± 5.4	10.2**	ns
Ammonium (mg/kg^{-1})	24.8 ± 2.1	19 ± 1.8	6.2*	ns
Nitrate (mg/kg^{-1})	11.0 ± 1.1	8.2 ± 0.9	4.5*	ns
GWC (%) (0-10 cm)	10.1 ± 2.1	7.0 ± 0.7	10.2**	8.4**
GWC (%) (10-20 cm)	13.9 ± 1.6	6.7 ± 1.6	12.1**	11.2**
GWC (%) (20-30 cm)	13.2 ± 2.5	6.5 ± 2.0	12.0**	ns
GWC (%) (30-40 cm)	12.4 ± 1.2	6.0 ± 1.5	11.2**	ns

ns: not significant

* $p<0.05$

** $p<0.01$

3.4 Discussion

3.4.1 Long-term zero-tillage improves soil physical characteristics

Changes in soil physical characteristics associated with the adoption of zero-tillage generally resulted in improved soil quality, especially when implemented for at least six years across the East Midlands. Figure 3.1 shows the effect of different tillage systems on penetration resistance down to 60 cm. In general, crop root growth starts to become restricted when the penetration resistance exceeds 1500 kPa and is severely restricted at 2500 kPa or more (Coelho et al., 2000). Penetration resistance values for both tillage managements in the 0-20 cm layer were below the critical 1500 kPa level. At a 20-30 cm depth, where the potential for a plough pan during tillage operations is high, increased resistance down through the soil profile was very similar between conventionally tilled soils, and soils which had been converted from conventional to zero-tillage for up to ten years prior, and both managements had a resistance greater than 1500 kPa (Fig. 3.1a,b). Distinct differences in root distribution and total yield in compacted vs uncompacted layers has been previously shown (Burr-Hersey et al., 2017; Pierret et al., 2007).

In contrast, soils which had been in zero-tillage for the greatest length of time (11-15 years, Fig. 3.1c), had significantly reduced penetration resistance between depth intervals 35-60 cm compared to conventionally tilled soils. Increased stubble residue on the surface of zero-tilled soils provides organic matter and an energy source for soil macrofauna, particularly for earthworms, which loosen the soil through burrowing activities (Blanco-Canqui et al., 2005). Pelosi et al., (2009) reported three to seven times more anecic and epigeic (litter

feeders found within the top 3 cm of the soil) earthworms in zero-tilled systems than those in cultivated soils. The anecic species are highly sensitive to tillage operations due to their large size, and they are vulnerable to mechanical damage (Briones and Schmidt, 2017). However, in zero-till systems this species can build permanent deep vertical tunnels (up to 2 m) through the soil profile; increasing macroporosity (Capowiez et al., 2009), reducing penetration resistance (Ruiz et al., 2015) and encouraging deeper rooting growth (Fischer et al., 2014). Similar results were reported by Yvan et al., (2012) who observed that recolonization of anecic earthworms took between 12 and 24 months after a compaction event and the recovery of bulk density took more than four years in a silt loam soil.

The advantage of tillage, as it briefly loosens the soil surface, is still not overcome by its long-term weakening of the soil structure. As early as the 1980s, Tisdall and Oades stated that soil structural stability could be controlled by management. Soil aggregates are the building blocks of soil structure (Zheng et al., 2018) and the quantity and stability of soil aggregates directly regulate soil quality and fertility (Al-Kaisi et al., 2014). The stability of soil aggregates also controls the capacity of soil aggregates to resist exogenic action and to remain stable when exposed to changing environments. Therefore aggregate stability is closely correlated with soil erodibility (Zheng et al., 2018). Our results indicate that long-term zero-tillage improved aggregate stability in the top 20 cm compared to cultivated soils (Fig. 3.2), through reducing disturbance by mechanical tillage, maintaining surface residue cover, increasing organic

binding agents (Man et al., 2019), therefore preventing soil erosion in high wind and heavy rain events.

3.4.2 Impacts of tillage management and soil texture on soil hydraulic behaviour

Across the two sampling campaigns (post-harvest and active crop), zero-tilled soils had a greater soil water content in the top 10 cm compared to conventionally tilled soils (Table 3.4a, b). Crop residues left on the soil surface in zero-tillage systems reduces evapotranspiration (Brunel et al., 2013) and decreases soil temperature (Shen et al., 2018). A lower volume of macropores and a greater volume of medium-sized (0.5-50 μm) water-holding soil pores are also possible explanations for higher water content in the soil after the adoption of zero-tillage (Busari et al., 2015).

Soil water content is a highly variable parameter which depends on the microclimate and soil conditions. In the soils sampled in Nov-Dec 2015 which was post-harvest, where high precipitation was recorded (a mean of 113.5 mm across the Midlands [Met Office, 2019]), there was no difference in the soil water content between 10 – 50 cm soil depth for the contrasting tillage managements (Table 3.4a). However, in the warmer sampling period in which there was an active crop (July 2016, when mean rainfall was only 35.1 mm [Met Office, 2019]), a greater soil water content was found in zero-tillage at each depth interval compared to cultivated soils (Table 3.4b). Similar results were reported by Małecka et al., (2012) and McVay et al., (2006). In addition, it was observed that the soil water content was much greater in the top 20 cm in soils

which had been in zero-tillage for 15 years compared to the recently converted zero-tilled soils in both post-harvest and active crop soils. Similarly, Pagliai et al., (2004) reported that converting from conventional tillage to minimum tillage improved the soil pore system through increasing the micro porosity (0.5-50 mm) and the elongated transmissions pores (50-500mm) which they related to an increase in soil water content and consequently, to an increase of available water for plants after 24 years of minimum tillage. Future studies could assess the water release characteristics between zero and conventionally tilled soils, to investigate the plant available water between these two management systems as a greater water content does not always translate into more plant available water (Whalley et al., 2013).

In the 'active crop' soils the water repellency increased in zero, compared to conventionally tilled soils. This has been attributed to increases in concentrations of hydrophobic compounds associated with increasing surface soil organic carbon (Roper et al., 2013). Water repellency also occurs at low to moderate moisture contents (Dekker and Ritsema, 2000), in the July 2016 sampling, the surface moisture content was between 7 and 10% (Table 3.4b). However, this increase in water repellency isn't a level to be concerned at and may have beneficial effects on the soil health in the summer with the slow absorption of water into the zero-tilled soils, compared to quicker absorption by conventionally tilled soils. This study showed that the magnitude of zero-till induced changes in soil water repellency varied across seasons and soil textures. In July 2016, soil water-repellency was 5 and 3 times greater under zero-till than under conventional tillage in coarse and fine-textured soils respectively (Fig.

3.4b). However, in Nov-Dec 2015, zero-tilled soils were only 3 and 1.1 times more water-repelling compared to conventional tillage in coarse and fine-textured soils, respectively (Fig. 3.4a). Blanco-Canqui (2011) reported in summer that the soil water-repellency was 40 times greater compared to 2 times greater in the winter than that of conventional tillage. Similarily, Hallett et al., (2001) reported zero-till soils were 1.6 to 1.8 times more water repellent than conventionally tilled soils in a silt loam in Scotland. Although not measured in this study, management will also impact on soil water sorptivity, and efficient crop production substantially depends on the capturing and storing of adequate soil water (Villarreal et al., 2017).

Soils textures with a small surface area e.g. sandy soils, are quicker to become water repellent as it takes less hydrophobic material to coat individual grains compared to silt or clay particles (Wallis and Horne, 1992). However, it is important to note that only the coarse-textured zero-tilled soils (from July 2016) had a water repellency classed as 'strongly-water repellent' in this study. The remaining zero-till soils were categorised as either 'wettable' or 'slightly to moderately repellent', which studies have suggested may have beneficial impacts on soil physical characteristics and carbon sequestration, through slow water entry into the soil reducing rapid aggregate slaking, thus stabilising soil aggregates and promoting long-tern carbon sequestration inside soil aggregates (Blanco-Canqui, 2011). The smaller Q_{10} value reported for zero-tilled soils suggests that zero-tillage may mitigate CO_2 emissions in response to increasing temperatures compared to conventionally tilled soils (Meyer et al., 2018).

3.4.3 Microbial mechanisms may reduce carbon dioxide production

The absence of mechanical turnover in zero-tillage is favourable in providing a constant supply of organic materials to soil microorganisms (Balota et al., 2003), and this is evident in the increased levels of microbial biomass carbon and nitrogen in zero-tilled soils, throughout winter and summer months (Table 3.4a,b). An increase in microbial activities in zero-till soils was also evident in enzyme activities, which were greater in zero-till soils than conventionally tilled soils, and some enzymes showed a significant increase with duration of zero-tillage (Fig. 3.6). Soil enzymes have been proposed as suitable soil quality indicators owing to their relationship to soil biology and quick responses to changes in soil management practices (Doran et al., 1996; Mathew et al., 2012). This study demonstrated a consistent increase in cellulase, xylanase and dehydrogenase in zero-tilled soils compared to conventionally tilled soils. This has also been observed by Balota et al., (2004) for cellulase in south Brazil in a 21-year zero-till experiment.

Through a lack of mechanical disturbance and reduced aeration in zero-tilled soils, the soil matrix is less oxidising compared to conventionally tilled soils (Dinnes et al., 2002). The surface accumulation of crop residues and subsurface supply of organic matter through either root biomass or anecic earthworms transporting surface organic material deeper into the soil profile further enhances enzyme activity. The enzymes involved in the metabolism of carbon (cellulase and xylanase) degrade cellulose and other polysaccharides in crop residues and root biomass, and in an oxygen limiting environment, the organic

material is retained as humus, and the recalcitrant carbon is encapsulated in soil aggregates and thus helps to sequester carbon in soil and reduce CO_2 emissions.

Even though microbial activities and surface crop residues were greater in zero-tilled soils, potential CO_2 fluxes, which should reflect the respiration status of soil, were smaller under zero-tillage compared to conventional tillage (Fig. 3.5). Under conventionally tilled conditions, with continuous soil disturbance, the soil microbial community may potentially be in a state of stress compared to the more stabilised and consistent environment under zero-till conditions (Giller et al., 1997) and therefore the microbial community will function more effectively i.e. their respiration relative to their biomass is reduced (Mangalassery et al., 2015). These conditions may enhance CO_2 fluxes from conventionally tilled soils. Also, the formation and stabilisation of macroaggregates in zero-tillage represents an important mechanism for the incorporation and protection of soil organic matter that would otherwise be oxidised to CO_2 and lost, as in conventionally till practices.

CH_4 production did not significantly differ between tillage managements (Fig. 3.5); however, N_2O emissions increased in zero-tilled soils. This may be due to a lack of mechanical disturbance and greater water content, resulting in microsites of anoxic and aerobic conditions, simultaneously allowing nitrification and denitrification (Hermle et al., 2008). The large difference in CO_2 emissions between the managements, however, was the key driver of the greater global warming potential from conventionally tilled soils.

3.5 Conclusions

These results suggest that the zero-tillage systems in this study led to progressive improvement in several soil parameters, including decreasing soil penetration resistance and increasing aggregate stability in the top 20 cm. This will play an important role in building a soil's architecture which will reduce soil erosion, increase water infiltration and alter soil porosity. In this study, it has also been demonstrated that tillage practices have the potential to strongly influence CO_2 fluxes, alongside the other crucial biogenic gases, CH_4 and N_2O, and it was identified that long-term zero-tillage in UK agricultural soils reduces CO_2 emissions, which could be partially due to an accumulation of carbon in recalcitrant forms as a result of increased cellulase and xylanase activities. Even with increased emissions of CH_4 and N_2O, the net GWP was significantly reduced under zero-tilled soils which were in this management for the longest period of time. This study suggests that soil tillage management have significant effects on soil structure and microbial biomass which influences the GHG release and water transport through the soil.

4.0 The impact of long-term zero-tillage on greenhouse gas emissions and soil biophysical properties in tropical soils

This chapter focusses on how zero and conventional tillage affects GHG production and mechanisms of carbon storage in Brazil, in a contrasting climate to the previous studies in chapter 2 and 3. This study also uses a paired sampling design but implemented on a smaller scale due to site, time and transportation limitations. An interesting aspect of Brazilian agriculture compared to the UK is that zero-tillage has been a popular method of farming since the late 1950s. As a result, soil samples were collected from fields which had been in zero-tillage for >30 years. GHG production and carbon content were measured in these samples to assess whether the temporal trend observed in zero-tilled soils was observed within tropical climates, when in continuous zero-tillage for periods greater than 15 years.

4.1 Introduction

Zero-tillage has been widely adopted since the 1940s, with 111 million ha globally managed using this approach (Derpsch et al., 2010). In Brazil, the adoption of zero-tillage increased from 1 million ha in 1990 to 23.6 million in 2005, accounting for c. 45% of all agriculture, thereby making it a critical agricultural management practice across South America.

Adoption of zero-tillage has generally been driven in Brazil by its ability to reduce soil erosion. Between 1990 and 2005 zero-tillage is estimated to have reduced soil erosion from 3.4-8.0 t/ha under conventional tillage compared to 0.4 t/ha in zero-tillage (Derpsch, 2008). Studies on the effects of zero-tillage on soil properties have previously shown decreases in surface runoff during heavy rain and high wind events (DeLaune et al., 2013), increases in water infiltration rate and greater storage capacity compared to conventionally tilled soils (Copec et al., 2015). These differences have been ascribed to several factors, including an increase in aggregate stability (Zheng et al., 2018), a greater number of biopores originating from former root growth and earthworm burrowing (Briones and Schmidt, 2017) and a decrease in the frequency of machinery traffic passing over the soil, collectively altering soil architecture and porosity (Galdos et al., 2019).

Soil porosity is a critical physical attribute of a soil's structural matrix, as it determines oxygen and water availabilities, which in turn governs gas exchange, crop yield and microbial biomass and abundance (de Araújo Santos et al., 2019). However, relatively few studies have assessed the combined impact of zero, and conventional tillage on the gas exchange (of all three major biogenic gases), microbial activity and soil physical properties (Mangalassery et al., 2014), and even fewer studies have been conducted in the tropics as opposed to temperate climates. For example, a meta-analysis by Pittelkow et al., (2015) on the differences in crop yield between zero and conventionally tilled soils included 6005 observations, of which only 7% was from tropical climates. Similarly, Ogle et al., (2005), identified 80 peer-reviewed articles which assessed soil

organic carbon differences between zero and conventionally tilled soils, of the 80 articles, of which fewer than ten were from the tropics.

Beyond the sparsity of studies, most have focused on agricultural land under zero-tillage management for ten years or fewer, with long-term studies typically viewed as those with five years beyond conversion. However, evidence suggests the duration of zero-tillage management impacts on crop yield, soil physical properties, biological activity and GHG emissions (de Araújo Santos et al., 2019; Díaz-Zorita et al., 2004; Lal et al., 1994).

With atmospheric concentrations of GHGs increasing globally, there is an urgent need to assess the potential role of soil as a carbon sink and regulating and mitigating GHG production across different agricultural managements. GHG emissions in tropical soils differ significantly from other climates due to changes in oxidation rates of soil organic matter. High temperatures and moisture throughout most of the year results in rapid decomposition of crop residues incorporated in the soil or maintained on the surface (Bolliger et al., 2006; Castro et al., 2015). Some studies have shown fluxes of CO_2, CH_4 and N_2O may have antagonistic responses to zero-tillage (Mangalassery et al., 2015), for example, reduced soil CO_2 and CH_4 fluxes but increased N_2O fluxes (Liu et al., 2006), resulting in a net increase in global warming potential (GWP) compared to conventionally tilled soils (Abdalla et al., 2013). However, this relationship has not been conclusively demonstrated in tropical agriculture.

To address these critical knowledge gaps, the key questions which are addressed here are: 1) how does the conversion from conventional to zero-tillage alter GHG emissions, soil physical properties and microbial activity across a range of tropical agricultural soils? and 2) how do these effects vary over time under zero-tillage? To address these questions, we conducted a field study, based on several closely located pairs of zero and conventionally tilled fields, across difference soil textures and micro-climates in Brazil, where zero-tillage had been implemented for 2-30 years. In line with question 1 it was hypothesised that i) long-term zero-tillage will increase shear strength due to reduced disturbance and increase total soil porosity due to the development of biopores and ii) CO_2 fluxes will be greater from conventionally tilled soils due to mechanical disturbance breaking apart aggregates and exposing organic C to mineralisation. iii) N_2O fluxes will be larger from zero-tilled soils due to increased moisture content, and substrate availability and iv) conventionally tilled soils will be a net source of CH_4 compared to zero-tilled soils which will be a net sink due to increased methanotrophic activity in undisturbed soils. For question 2, it was hypothesised v) microbial activity will be greater in long-term zero-tilled soils due to increased organic matter from surface crop residues and reduced disturbance.

4.2 Methods

4.2.1 Site selection and sample collection

This study was conducted in Brazil, with samples collected from experimental farms from four regional States: Maranhão, Goiãs, São Paulo and Paranã (Fig. 4.1). Soil samples were collected in November 2016 from locations 1 and 2 and in November 2017 from locations 3 and 4 (Fig. 4.1). Expect for location 2, soil samples were taken from twelve fields, of which six had been zero-tilled for a varying length of time, and six paired conventionally tilled fields (detailed are outlined in Table 4.1). In location 2, it was not possible to have paired conventional and zero-tilled comparisons due to the limited availability of conventionally tilled fields. All zero-tilled soils had been managed for between 2 and 30 years, whereas the conventionally tilled soils were subjected to annual mechanical turnover to a depth of at least 10 cm.

From each eperimental field, using a randomised sampling method, four intact soil cores (5 cm diameter x 30 cm depth) were collected using a manual core sampler that used transparent sample liner tubes (Van Walk Lts, Haslemere, UK) for X-ray Computed Tomography imaging and measurements of greenhouse gas fluxes. The soil cores were first sealed with paraffin wax at the top and bottom to prevent movement during transit. To account for changes in bulk density between managements, soil samples were collected to a depth of 50 cm at 5 cm increments using a stainless-steel cutting cylinder (7 diameter cm × 4 cm height). Simultaneously, bulk soil samples were collected at each increment for measurements of soil organic matter, aggregate size distribution and soil enzyme activities.

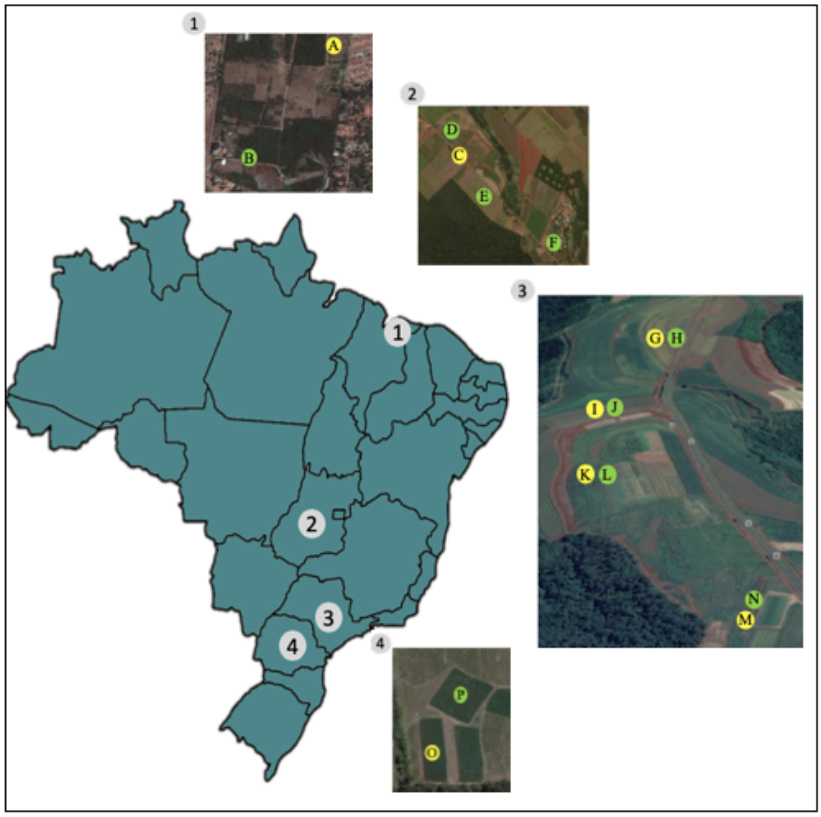


Figure 4.1 The location of the 16 sampled fields across experimental farms in Brazil. Soil samples were taken from one pair of zero and conventionally tilled fields from locations 1 and 4, four pairs from location 3 and one conventionally tilled and three zero-tilled fields from location 2. The yellow circles indicate conventional management and green circles indicate zero-till management. Additional soil and management characteristics of the experimental fields are in Table 4.1.

Table 4.1 Selected soil and management characteristics of experimental fields. ZT and CT refer to zero and conventional tillage, respectively.

Field	Management	Crop rotation	% clay	Location	WRB
A	15-20 cm CT	*	10	2°35'12.3"S 44°12'29.6"W	Arenosols
B	15 years ZT	*	10	2°35'34.9"S 44°12'43.5"W	Arenosols
C	15-20 cm CT	Soybeans/fallow	65	16°29'16.7"S 49°17'59.6"W	Ferralsols
D	2 years ZT	Maize/beans	65	16°29'56.1"S 49°17'02.1"W	Ferralsols
E	22 years ZT	*Urochloa spp*	65	16°29'29.9"S 49°17'48.1"W	Ferralsols
F	6 years ZT	*Urochloa spp*	65	16°29'21.0"S 49°18'10.4"W	Ferralsols
G	15-20 cm CT	Triticale/maize	65	22°48'42.7"S 48°25'43.4"W	Nitisols
H	16 years ZT	Triticale/soybean	65	22°48'43.0"S 48°25'40.3"W	Nitisols
I	15-20 cm CT	Maize/soybean	62	22°49'10.6"S 48°25'51.3"W	Nitisols
J	30 years ZT	Maize/soybean	62	22°49'10.9"S 48°25'46.3"W	Nitisols
K	10-15 cm CT	Grasses/maize	62	22°49'14.5"S 48°25'44.4"W	Nitisols
L	3 years ZT	Grasses/maize	62	22°49'16.3"S 48°25'47.7"W	Nitisols
M	15-20 cm CT	Maize/black oat	35	22°49'47.7"S 48°25'33.5"W	Nitisols
N	11 years ZT	Soybean/maize	35	22°49'46.6"S 48°25'32.6"W	Nitisols
O	20-25 cm CT	Black oat/corn	63	26°21'30.0"S 51°20'10.0"W	Alisols
P	28 years ZT	Black oat/corn	62	26°21'30.7"S 51°20'11.8"W	Alisols

*Information not available at the time of writing.

4.2.2 Soil physical properties

4.2.2.1 Soil porosity

Following sealing, samples were transported to the Hounsfield Facility at the University of Nottingham for X-ray CT scanning. Previously, a pilot study using X-ray CT scanning of soil cores in Brazil and the UK demonstrated the efficacy of paraffin wax as an approach for preventing disturbance during transit (Galdos et al., 2019).

Soil cores were scanned using a Phoneix v|tome|x M scanner (GE Measurement and Control Solution, Wunstorf, Germany). This allowed visualisation and quantification of the soil porous architecture. Voxel resolution was set to 50 µm, with a potential energy of 160 kV and a current of 180 µA. Soil cores were scanned in three sections (i.e. depths, where each section was 8cm in length, thereby each soil core was scanned to a total length of 24 cm), with a total scan time of 30 minutes per soil core. A total of 2998 image projections were captured for each core.

Analysis of soil pore morphology was carried out using ImageJ, an open-source image processing software. A cuboid region of interest (ROI) (50 ×50 mm^3) was selected from the centre of the reconstructed CT images to exclude pores adjacent to the core edges. The ROI was subject to one pass of the 'sharpen' filter followed by one pass of the 'median' filter to enhance the image quality before being converted to the binary scale using the 'MinError' threshold algorithm. A further filter operation ('despeckle') was undertaken on the binary images before analysis. Finally, a wide range of pore morphological properties

was collected using the ‘analyse particles’ function, including pore size and shape.

4.2.2.2 Shear strength

Surface (5 cm) soil strength was recorded in the field using a Pilcon 120 kPa hand vane. The vane was rotated carefully at a uniform speed until the soil failed and the force was recorded. Twelve readings were collected from each field.

4.2.2.3 Aggregate stability

Loose soil samples from each field (down to 50 cm in 10 cm increments) were air-dried for several days and gently broken apart by hand into smaller aggregates during drying. The soils were then sieved, and aggregates sized between 3-5 mm retained for analysis. Five g of calibrated soil aggregates were gently immersed in 50 ml deionised water for 10 minutes; the water was siphoned off using a pipette and the soil material transferred to a 50 μm sieve (which had previously been submerged in methylated spirit to avoid re-aggregation of the soil sample and to reduce the amount of additional breakdown). The 50 μm sieve was gently moved up and down in a beaker containing methylated spirit for 1 minute, and care was taken to avoid the methylated spirit from flowing into the top of the sieve to avoid further disruption of the soil aggregates. This first step separated fragments < 50 μm from those > 50 μm. Once the methylated spirit evaporated, the sieve containing the remaining sample was placed in an oven for 8 hours at 105 °C. After drying, the sample was gently sieved through a stack of six sieves (2000, 1000, 500, 200, 100, and 50 μm). The soil mass remaining on each sieve was recorded, and

the mass percentage in each size fraction calculated. The aggregate stability for each breakdown mechanism was expressed as the mean weight diameter (MWD):

$$MWD = \sum_{i=l}^{n} xi\ wi, \qquad \text{(Equation 4.1)}$$

where wi is the proportion of the sample with a mean size of xi μm (van Bavel, 1950).

4.2.3 Soil biochemical properties

4.2.3.1 Microbial biomass carbon

Soil microbial biomass carbon (MBC) was determined for all surface samples (0-10 cm) and subsurface samples (40-50 cm) using the chloroform fumigation method (Vance et al., 1985). For this, fumigated (for 24 h) and non-fumigated soils (10.0 ± 0.5 g fresh sample) were extracted with 0.5 M K_2SO_4 followed by analysis using a Shimadzu CN analyser (TOC-V CPH Shimadzu). The value of the coefficient (K_{EC}) to convert 'chloroform-labile' carbon to microbial biomass carbon of 0.45 was used (Jenkinson et al., 2004).

4.2.3.2 Soil organic matter

Soil organic carbon content was assessed using the loss on ignition method (LOI), by igniting the humus content of oven-dried soils at 550ºC in a muffle furnace. Soil organic carbon stock was then recorded by multiplying organic carbon content with the thickness of the soil core and bulk density (Batjes, 2014).

4.2.3.3 Greenhouse gas fluxes

Potential GHG fluxes were measured following a four-week incubation under controlled environments. This approach was adopted to allow comparison between management by removing the effects of climate on gas production *in-situ*. Intact soil cores were removed from the 4 °C environment, and water-filled pore space was adjusted to 60% (equation 4.2), and then kept at 25 °C for 48 hours to activate and stabilise the soil microflora.

$$WFPS = \frac{(Soil\ water\ content\ x\ Bulk\ density)}{(1-(Bulk\ density\ /\ Particle\ density))} \quad \text{(Equation 4.2)}$$

Each soil core was incubated in a plastic jar (1.5 dm^3) with a septum on the top to allow gas sampling. The air in the headspace was mixed via a magnetic stirrer during gas sampling at time intervals 0, 15, 30 and 60 minutes using a 20 mL syringe. The collected gas samples were stored in pre-evacuated 12 mL airtight glass vials. Gas samples were analysed for concentrations of CO_2, CH_4 and N_2O by gas chromatography (GC-2014, Shimadzu) and potential GHG flux data was converted to mass per volume and mass per weight basis using the ideal gas law (Equation 4.3) (Laugier and Garai, 2009).

$$PV = nRT, \quad \text{(Equation 4.3)}$$

where P is the atmospheric pressure (≈ 1 atm), V is the volume of headspace (dm^{-3}), n is the number of moles of gas, R is the ideal gas constant (0.08205746 L atm K^{-1} mol^{-1}), and T is the temperature (273 + room temperature in °C).

From this, the potential flux of each gas was calculated (Equation 4.4):

$$E = \frac{nm}{at}\ x\ 1000, \qquad \text{(Equation 4.4)}$$

where E is the flux of each has (mg m^{-2} h^{-1}), n is the number of moles (CO_2, CH_4 or N_2O), m is the molar weight (CO_2 and N_2O is 44.01 and CH_4 is 16.04), a is the area of the soil core measured and t is the time period for the incubation, in hours.

Global warming potential (GWP) was calculated in CO_2-equivalents following equation 4.5 (Myhre et al., 2013):

$$GWP = \left\{\left(\frac{CO_2\ x\ 44}{12}\right) + (CH_4\ x\ 23) + (N_2O\ x\ 296)\right\}, \qquad \text{(Equation 4.5)}$$

4.2.3.4 Enzyme assays

Dehydrogenase activity was determined using an approach modified by Ohlinger (1995), originally from Thalman (1968). The activity within the soil samples is measured colourimetrically, by the reduction of 2,3,5-triphenyltetrazolium chloride (TPC) to triphenyl formazan (TPF). The intensity of TPF was measured at 546 nm against known TPF standards and was expressed as μg TPF g^{-1} h^{-1}.

Both cellulase and xylanase activity was measured by the reduction of potassium hexacyanoferrate (III) to potassium hexacyanoferrate (II). For cellulase activity, the reduced potassium hexacyanoferrate (II) was reacted with ferric ammonium sulphate in an acid medium to form a coloured complex of ferric hexacyanoferrate (II), using carboxy-methyl cellulose as a substrate. For xylanase activity, the same reaction occurred but using xylan as a substrate. Both colour intensities were assessed at 690 nm and expressed as mg GE g^{-1} day^{-1} (Schinner and Vonmersi, 1990).

4.2.3.5 pH, EC and redox potential

The pH, electrical conductivity (EC) and redox potential were determined by adding 5 g of fresh soil to 12.5 ml Milli-Q water, in triplicate, in a 50 ml centrifuge tube (Fisher Scientific). pH, redox potential (HANNA instrument, 209, pH and redox meter) and EC (HANNA, HI9835 Microprocessor Conductivity/TDS meter) readings were taken after placing on a rotary shaker for 30 minutes and solution heated to 25°C.

4.2.4 Statistical analyses

Differences in soil properties between zero-tilled and conventionally tilled fields were assessed using a linear mixed-effects model fitted using Residual Maximum Likelihood (REML). Differences in cellulose, dehydrogenase, xylanase, microbial biomass carbon, ammonium (NH_4^+), nitrate (NO_3^-), electrical conductivity (EC) and pH included depth, treatment and duration as fixed effects, and site and pair (surface or subsurface) as random effects. The

interaction between depth and treatment, and depth and duration were also included in models. Analyses of GHG production and GWP, and surface organic carbon, porosity, shear vane and aggregate stability included only treatment and duration as fixed effects and the sites as a random effect. All statistical analyses were carried out in Genstat v17.0.

4.3 Results

4.3.1 Soil physical properties

Soil management had a significant effect on surface soil strength ($F_{1,51}$ = 163, $p<0.001$, Fig 4.2), with zero-till soils exhibiting a greater soil strength compared to conventionally tilled soils. Shear strength also significantly increased in the top 5 cm, in soils which had been in zero-tillage for the longest period of time compared to conventionally tilled soils ($F_{9,54}$ = 156, $p<0.001$, Fig 4.2). For example, across the conventionally tilled fields at location 3 (Fig 4.2c), the mean shear strength was 9.3 kPa. Soils which had been in zero-tillage for 3, 11, 16 and 30 years the mean shear strength was 9.8, 15, 21 and 27 kPa, respectively.



Figure 4.2 Shear strength measured in the top 5 cm from different locations in

Brazil. From (a) Maranhão, (b) Goiãs, (c) São Paulo and (d) Paranã. CT refers to conventional tillage and numbers on the x-axis refers to the number of years in zero-tillage.

Soil management had a significant effect on total soil porosity ($F_{1,51}$ = 237, p<0.001, Fig 4.3), with the smallest total porosity measured in the conventionally tilled soils. The greatest porosity was measured at Maranhão (Fig 4.3a), with a mean porosity of 47 and 50% in conventional and zero-tilled soils. These samples had the largest proportion of sand (~80%, Table 4.1). Total porosity also significantly increased in the top 16 cm, in soils which had been in zero-tillage for the longest period of time compared to conventionally tilled soils ($F_{8,51}$ = 55, p<0.001, Fig 4.3). The soils which had been in zero-tillage for the longest length of time (30 years) had a mean porosity of 22.0 % in the top 16 cm, compared to 11.9 % in the paired conventionally tilled soil (Fig 4.3c).

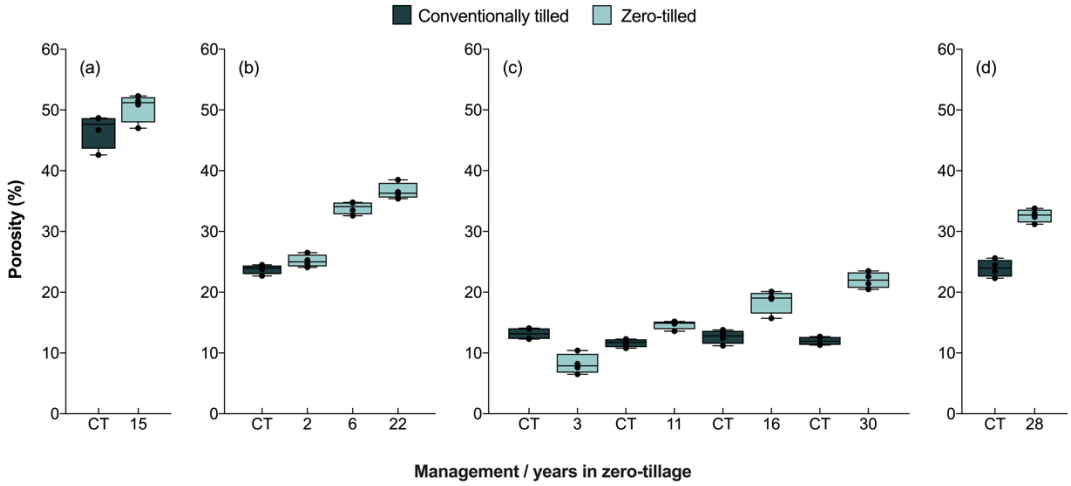


Figure 4.3 Total soil porosity measured by X-ray Computed Tomography in the top 16 cm from different locations in Brazil. From (a) Maranhão, (b) Goiãs, (c)

São Paulo and (d) Paranã. CT refers to conventional tillage and numbers on the x-axis refers to the number of years in zero-tillage.

Soil management had a significant effect on aggregate stability in the top 10 cm ($F_{1,45} = 23$, $p<0.001$, Fig 4.4). Samples from São Paulo and Paranã had the largest aggregate stability in soils which had been in zero-tillage for 30 and 28 years, with a mean weight diameter >2 cm (Fig 4.4c&d). These aggregates are classed as 'very stable'. At Goiãs, the soils which were under zero-tillage for 22 years had a mean weight diameter of 1.4 mm, indicating that the aggregates were 'stable'. However, the aggregates from the conventionally tilled soil, and soils which had been under zero-tillage for 2 and 6 years had a mean weight diameter 0.73 - 0.84 mm, indicating that these aggregates were 'unstable/median' (Fig. 4.4b).

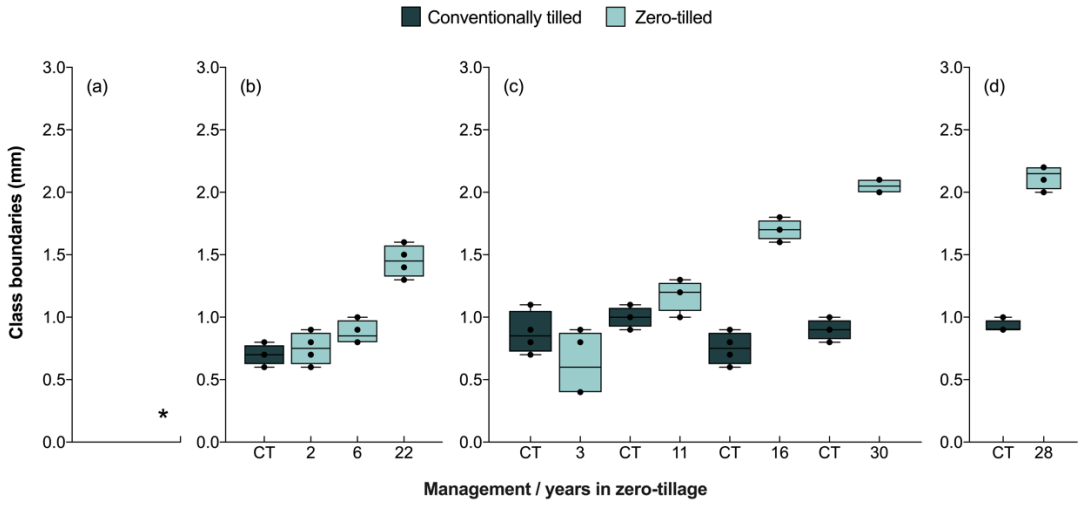


Figure 4.4 Aggregate stability from zero and conventionally tilled soils in the top 10 cm. From (a) Maranhão, (b) Goiãs, (c) São Paulo and (d) Paranã. CT refers to conventional tillage and numbers on the x-axis refers to the number of years in zero-tillage. * Aggregate stability could not be measured in these soils.

4.3.2 Soil biochemical properties

Microbial biomass carbon (MBC) was significantly greater in the surface zero-tilled soils compared to surface conventionally tilled soils, but the opposite was observed in the sub-surface soils ($F_{1,103} = 128$, $p<0.001$, Table 4.2). Soils in zero-tillage for the greatest length of time had a significantly greater MBC compared to conventionally tilled and recently converted soils ($F_{8,103} = 9.0$, $p<0.001$, Table 4.2).

Organic carbon content was significantly greater in the top 10 cm in zero-till compared to conventionally tilled soils ($F_{1,51} = 86$, $p<0.001$, Table 4.2a). The largest increase in organic carbon content was observed at Paranã (Table 4.2a), whereby concentrations increased from 30.9 g kg^{-1} in conventionally tilled soil to 73.6 g kg^{-1} in soils which were in zero-tillage for 28 years.

Electrical conductivity (EC) was significantly greater in the surface (0-10 cm) compared to sub-surface (40 -50 cm) soils ($F_{1,92} = 91$, $p<0.001$, Table 4.2), however, there was no significant treatment effect ($p>0.05$). Similarly, for ammonium (NH_4-N), there were significantly greater concentrations in the surface compared to subsurface soils ($F_{1,50} = 26$, $p<0.001$, Table 4.2), but no treatment effect ($p>0.05$). There was no significant depth or treatment effect observed for nitrate (NO_3-N) ($p>0.05$).

Table 4.2 Means and standard errors for the bio-chemical properties of soils under zero (ZT) and conventional tillage (CT) from soil samples collected in (a) the surface, 0-10 cm and (b) the sub-surface, 40-50 cm. Blank entries: data not available. EC is electrical conductivity.

Location	Field	Microbial C (mg kg^{-1} soil)	Organic C (g kg^{-1})	EC (dSm^{-1})	Ammonium (mg kg^{-1})	Nitrate (mg kg^{-1})
(a) Surface, 0 – 10 cm						
Maranhão	A, CT	85.5 ± 26.5	4.9 ± 0.8	2.20 ± 2.7	39.5 ± 5.6	2.0 ± 0.4
	B, ZT	96.2 ± 18.1	10.7 ± 0.7	3.93 ± 1.2	40.3 ± 7.1	1.3 ± 0.5
Goiãs	C, CT	90.7 ± 20.9	17.7 ± 1.9	2.41 ± 3.4	35.6 ± 3.3	1.1 ± 0.3
	D, ZT	151 ± 29.3	23.2 ± 0.6	2.53 ± 4.3	31.0 ± 3.1	2.5 ± 0.6
	E, ZT	211 ± 45.7	32.5 ± 2.4	4.18 ± 1.3	37.6 ± 5.2	3.5 ± 0.6
	F, ZT	328 ± 53.4	50.5 ± 2.6	4.55 ± 2.4	41.7 ± 4.2	7.9 ± 0.9
São Paulo	G, CT	341 ± 7.0	20.1 ± 0.1	2.57 ± 1.6	-	-
	H, ZT	360 ± 6.9	20.9 ± 0.6	2.56 ± 4.3	-	-
	I, CT	300 ± 4.3	19.8 ± 0.6	2.52 ± 4.0	-	-
	J, ZT	410 ± 4.1	31.8 ± 1.0	2.87 ± 7.9	-	-
	K, CT	342 ± 2.2	19.5 ± 1.0	2.56 ± 6.6	-	-
	L, ZT	467 ± 6.3	38.9 ± 0.6	4.29 ± 3.6	-	-
	M, CT	345 ± 6.8	17.9 ± 0.6	2.48 ± 4.5	-	-
	N, ZT	500 ± 7.4	46.3 ± 0.5	4.39 ± 4.0	-	-
Paranã	O, CT	108 ± 5.4	30.9 ± 0.6	2.57 ± 3.5	34.2 ± 3.0	3.1 ± 0.4
	P, ZT	240 ± 5.3	73.6 ± 1.3	3.94 ± 1.0	41.0 ± 3.2	7.7 ± 0.7
(b) Sub-surface, 40 – 50 cm						
Maranhão	A, CT	38.1 ± 11.6	-	1.81 ± 1.3	28.5 ± 2.2	0.3 ± 0.1
	B, ZT	26.7 ± 8.8	-	1.03 ± 6.0	22.3 ± 1.6	0.4 ± 0.1
Goiãs	C, CT	158 ± 49.7	-	2.40 ± 5.2	29.0 ± 1.8	4.4 ± 0.1
	D, ZT	78.9 ± 2.9	-	3.67 ± 2.2	27.3 ± 1.9	4.4 ± 0.5
	E, ZT	87.7 ± 34.5	-	2.58 ± 3.7	27.3 ± 2.2	2.0 ± 0.3
	F, ZT	188 ± 34.8	-	2.92 ± 5.1	33.1 ± 3.3	2.3 ± 0.3
São Paulo	G, CT	336 ± 12.8	-	2.00 ± 0.7	-	-
	H, ZT	103 ± 2.2	-	2.17 ± 0.6	-	-
	I, CT	291 ± 3.4	-	2.46 ± 0.8	-	-
	J, ZT	157 ± 3.1	-	1.97 ± 0.5	-	-
	K, CT	322 ± 7.7	-	2.31± 2.1	-	-
	L, ZT	181 ± 4.3	-	2.70 ± 2.0	-	-
	M, CT	345 ± 4.4	-	2.11 ± 1.8	-	-
	N, ZT	205 ± 6.5	-	2.66 ± 2.2	-	-
Paranã	O, CT	51.5 ± 2.8	-	2.07 ± 0.9	26.9 ± 1.0	2.3 ± 0.4
	P, ZT	103 ± 2.6	-	2.34 ± 1.9	33.8 ± 3.0	5.2 ± 0.9

4.3.2.1 Greenhouse gas fluxes

CO_2 fluxes from conventionally tilled soils were significantly greater than those from zero-tilled soils ($F_{1,50} = 210$, $p<0.001$, Fig 4.5). When the soils under zero-tillage were grouped by length of time under this management, CO_2 fluxes were smallest for the soils with the longest history of zero-tillage ($F_{8,38} = 18$, $p<0.001$, Fig 4.5). The smallest difference observed was at Maranhão (Fig 4.5a), where CO_2 fluxes from conventionally tilled soils averaged 270 mg CO_2 m^{-2} h^{-1} compared to 180 mg CO_2 m^{-2} h^{-1} in the adjacent zero-tilled soils. The largest difference observed was at São Paulo (Fig 4.5c), where CO_2 fluxes from conventionally tilled soils averaged 253 mg CO_2 m^{-2} h^{-1} compared to 53 mg CO_2 m^{-2} h^{-1} in the zero-tilled soils which were under this management for 30 years. There were no significant differences in N_2O or CH_4 fluxes between zero and conventionally tilled soils ($p>0.05$, Fig 4.5).

The global warming potential (GWP) was significantly greater from conventional compared to zero-tilled soils ($F_{1,51} = 246$, $p<0.001$, Fig 4.6). The smallest difference in GWP was observed at Maranhão (Fig 4.6a), with a mean value of 1004 ± 130 mg CO_2 eq. m^{-2} h^{-1} in conventionally tilled soils compared to 630 ± 62 mg CO_2 eq. m^{-2} h^{-1} in the paired zero-tilled soils. When the soils under zero-tillage were grouped by length of time under this management, GWP was significantly smaller for the soils with the longest history of zero-tillage ($F_{8,53} = 16$, $p<0.001$, Fig 4.6). For example, at Goiãs (Fig 4.6b), the mean GWP for the conventionally tilled soils was 820 mg CO_2 eq. m^{-2} h^{-1}, compared to a GWP of 369 and 165 mg CO_2 eq. m^{-2} h^{-1} in soils which had been in zero-tillage for 6 and 22 years, respectively.

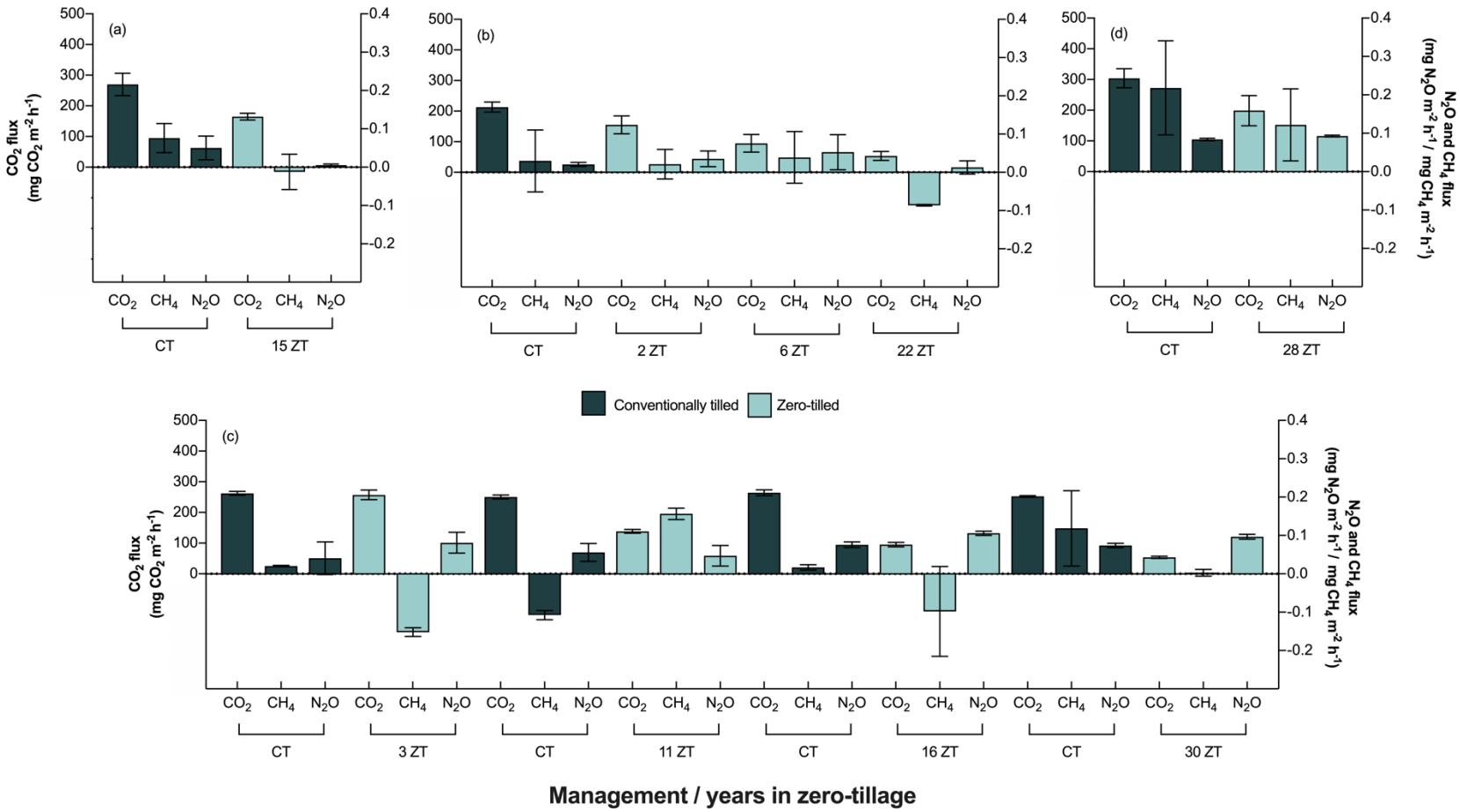


Figure 4.5 Carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O) fluxes from conventional (CT) and zero-tilled (ZT) soils across different locations in Brazil. From (a) Maranhão, (b) Goiás, (c) São Paulo and (d) Paranã. CT refers to conventional tillage and numbers on the x-axis refers to the number of years in zero-tillage. Mean ± standard error are shown, n=4.

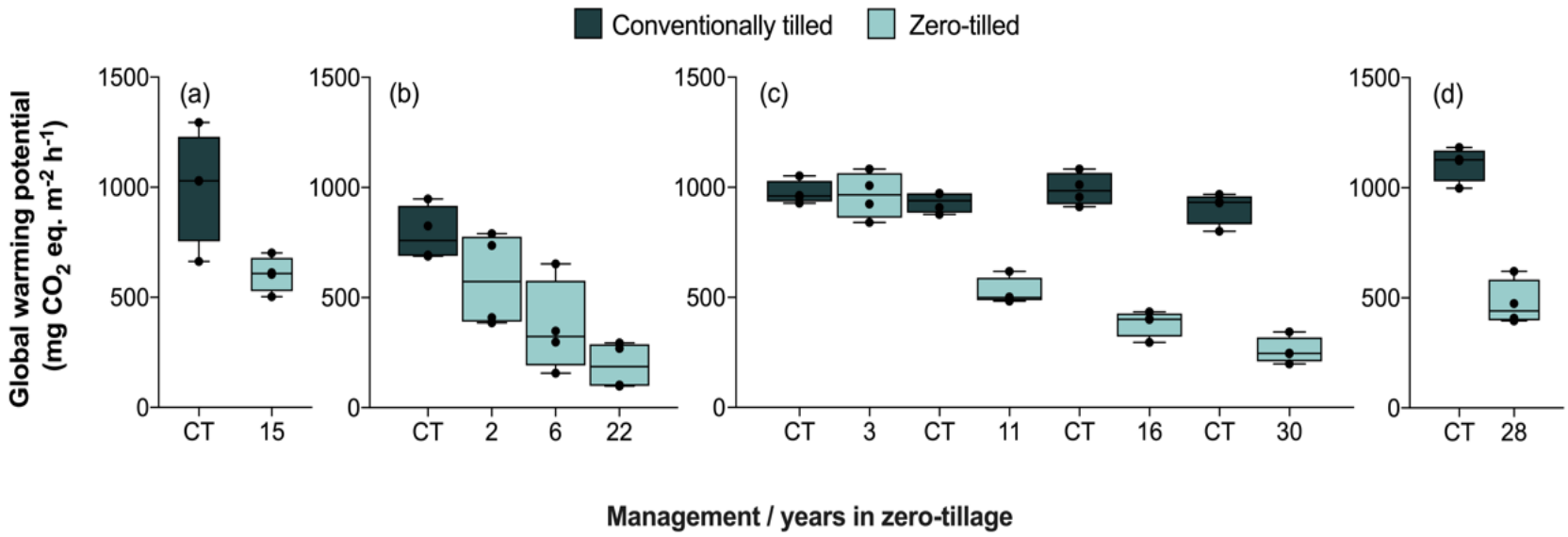


Figure 4.6 Global warming potential from conventional (CT) and zero-tilled (ZT) soils across different locations in Brazil. From (a) Maranhão, (b) Goiãs, (c) São Paulo and (d) Paranã. CT refers to conventional tillage and numbers on the x-axis refers to the number of years in zero-tillage.

4.3.2.1 Enzyme assays

Dehydrogenase activity was significantly greater in the surface (0-10 cm) compared to subsurface (40-50 cm) soils ($F_{1,103}$ = 59, $p<0.001$, Fig 4.7a-d). Dehydrogenase activity was also significantly greater in zero compared to conventionally tilled soils ($F_{1,88}$ = 370, $p<0.001$, Fig 4.7a-d), and activity significantly increased with duration in zero-tillage ($F_{8,45}$ = 36, $p<0.001$, Fig 4.7a-d). For example, at Paranã (Fig 4.7d) activity increased from 2.14 μg TPG g^{-1} h^{-1} in conventionally tilled soils to 2.79 μg TPG g^{-1} h^{-1} from the adjacent zero-tilled soils.

Similar significant observations were made for cellulase (C) and xylanase (X) activity. Activity was significantly greater at the surface compared to sub surface soils (C - $F_{1,103}$ = 67, $p<0.001$, Fig 4.7e-h) (X - $F_{1,103}$ = 113, $p<0.001$, Fig 4.7i-l) and activity was significantly greater in zero compared to conventionally tilled soils (C - $F_{1,75}$ = 118, $p<0.001$, Fig 4.7e-h) (X - $F_{1,75}$ = 37, $p<0.001$, Fig 4.7i-l).

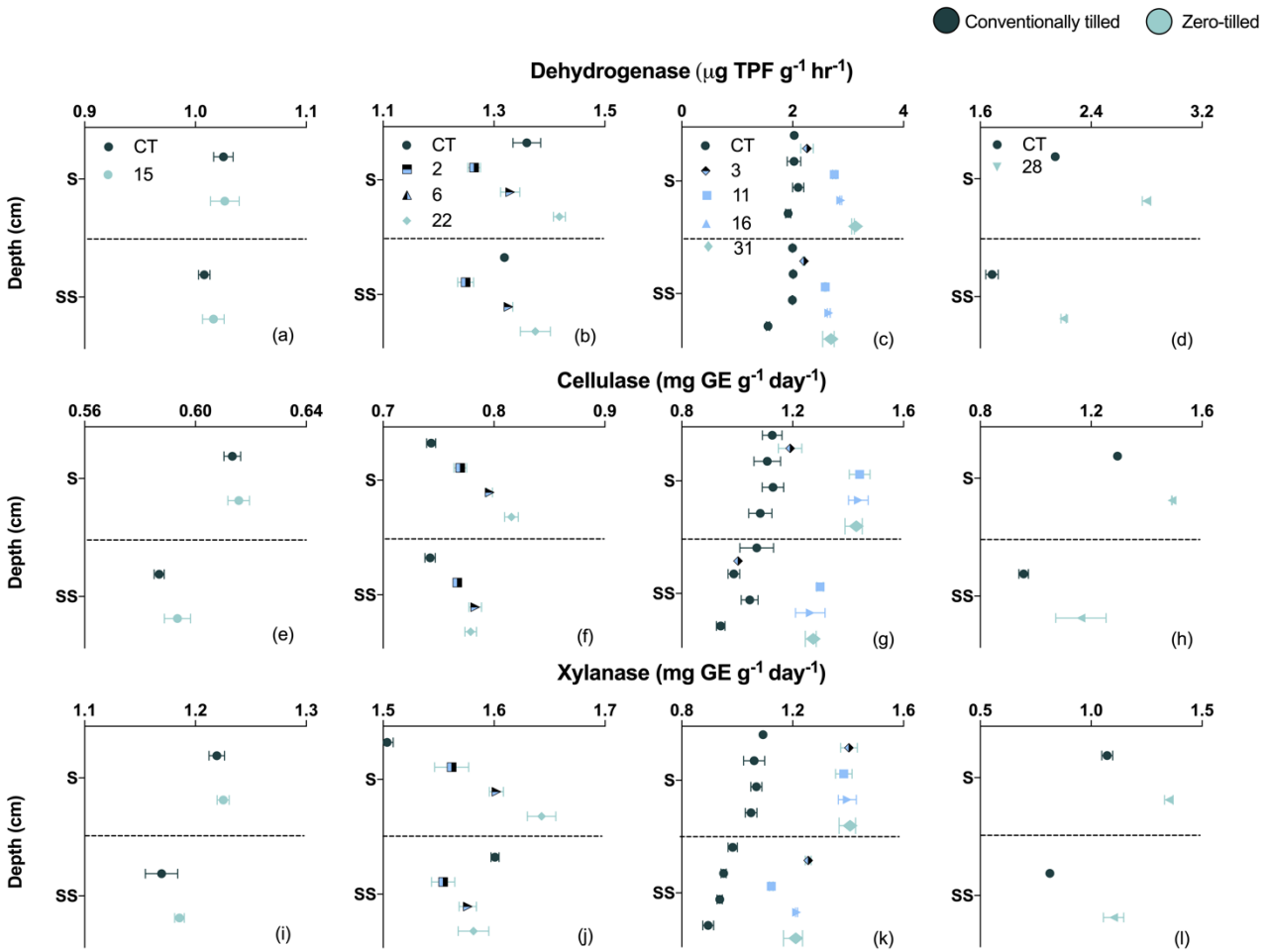


Figure 4.7 Soil enzyme assays at 0-10 cm (S) and 40-50 cm (SS) under zero and conventionally tilled soils which were sampled from (a,e,i) Maranhão, (b,f,j) Goiãs, (c,g,k) São Paulo, and (d,h,l) Paranã. The enzymes measured were (a-d) dehydrogenase, (e-h) cellulase and (i-l) xylanase. The dotted line illustrates a split between surface and subsurface samples.

4.4 Discussion

4.4.1 Crop residue retention in zero-tillage improves soil structural properties

Changes in soil physical properties associated with the adoption of zero-tillage resulted in improved soil structural properties, especially when implemented for at least three to five years. In line with the first hypothesis, shear strength increased at Goiãs, São Paulo, and Paranã in zero compared to conventionally tilled soils (Figure 4.2). The conversion from conventional to zero-tillage leads to soil consolidation in the surface and a subsequent decrease in volumes of macropores (Peigné et al., 2007). However, these recorded levels of shear strength are not likely to prevent root growth, especially if pore continuity is enhanced by the creation of biopores (Coelho et al., 2000). Surface cover through increased retention of crop residues alongside surface soil consolidation can act to limit soil particle detachment by shattering following raindrop impact, reducing the raindrop mass, and decreasing velocity (Montgomery, 2007).

Similarly, to shear strength, total porosity increased at Goiãs, São Paulo, and Paranã in zero compared to conventionally tilled soils (Figure 4.3). In addition, the largest soil porosity was measured in soils which had been in zero-tillage for the greatest length of time. This effect is likely driven by the changes in soil structure caused by conventional tillage operations, which contributes to the disaggregation of continuous pores above the plough layer (Bauer et al., 2015), but also the creation of medium-sized pores (0.5-50 mm) that are important for water storage. However, at depths below ~10-15 cm, conventional tillage reduces porosity, due to the formation of a plough pan which slows the

infiltration rate of water and reduces root growth (Tuzzin de Moraes et al., 2016). An important consideration in this study is the duration of time that the soils had been managed under zero-tillage. Very few studies have examined the soil structure, via non-destructive methods, following as long as 30 years of zero-tillage and it is most likely this long-time period is the driver of the significant increase in porosity, which is generally decreased in the short-term following adoption (Da Veiga et al., 2008). Increased crop residue on the surface of zero-tilled soils provides organic matter for soil macrofauna, which creates biopores through burrowing activities and these typically develop over long time scales (>10 years) in soils (Palm et al., 2014).

4.4.2 Long-term zero-tillage reduces global warming potential

The greater CO_2 release observed in response to conventional tillage (Fig 4.5) supports the second hypothesis and emphasises the role of ploughing in the turnover of soil aggregates and exposure of organic materials for microbial decomposition. The greater porosity observed in zero-tilled managements will increase water and gas retention and exchange but, the surface consolidation results in a less oxidising environment in comparison to conventionally tilled soils (Dinnes et al., 2002). In contrast to the hypothesis, that 'N_2O fluxes will be larger from zero-tilled soils due to increased moisture content and substrate availability', we did not find any significant differences in N_2O fluxes between tillage systems (Fig. 4.5). The majority of studies have shown greater N_2O emissions from zero compared to conventionally tilled soils, which is ascribed to increased rates of nitrification, and greater bulk density and soil water content (Hermle et al., 2008). However, the impact of zero-tillage on N_2O emissions also

depends on soil temperature, texture and duration of the period in zero-tillage, and some studies have described similar fluxes between the two tillage systems. For example, Jantalia et al., (2008) reported no significant difference in N_2O fluxes between zero and conventional tillage in clay soils in Rondôia, Brazil. There was also no management effect on CH_4 fluxes between the tillage managements. The large differences in CO_2 emissions between the managements, however, was the key driver of the lower GWP in the zero-till soils that had been under this management for the greatest length of time (Fig 4.6).

In line with the third hypothesis, greater microbial activity in long-term zero-tilled soils was observed (Fig 4.7). Crop residue accumulation at the soil surface, coupled with a continuous sub-surface supply of organic matter derived from root necromass and earthworm transport both enhance microbial activity (Brown et al., 2003; Mathew et al., 2012). Both cellulose and xylanase are key enzymes in soil carbon cycling, including the decomposition of cellulose and xylan, key structural components of soil organic matter. In low oxygen environments, the organic material is retained as humus, and the recalcitrant carbon is encapsulated in soil aggregates and thus helps to sequester carbon in soil and reduce CO_2 emissions (S. Mangalassery et al., 2015).

Even though microbial activity and surface crop residues were greater in zero-tilled soils, potential CO_2 fluxes, which broadly reflect the respiration status of the soil, were reduced under zero-tillage compared to conventional tillage as previously outlined above. Under conventionally tilled conditions, ongoing soil

disturbance can stress microbial communities, preventing them from reaching a steady-state, (Giller et al., 1997). Therefore the lack of disturbance in zero-tilled soils allows them to continually develop function more effectively i.e. their respiration relative to their biomass is reduced (Mangalassery et al., 2015). In addition, the formation and stabilisation of macroaggregates in zero-tillage represents an important mechanism for the incorporation and protection of soil organic matter that would otherwise be oxidised to CO_2 and lost, as in conventionally till practices.

4.5 Conclusions

This study demonstrates that, when combining all three GHG fluxes, potential GWP from zero-tilled soils was 50% lower than that of conventionally tilled soils. By extending the duration of zero-tillage it is possible to significantly reduced GWP further. This effect was primarily driven by decreased CO_2 fluxes which may be ascribed to an improvement in soil structural characteristics, increased microbial activity and increased aggregate stability. In many scenarios, zero-till managements can represent a 'win-win' situation, where there is potential for CO_2 mitigation, simultaneously, other important benefits are achieved, including improved soil quality through increases in microbial biomass and abundance, protection of land against soil erosion, and reduced cost and labour savings. These added benefits often result in financial benefits for farmers and will be the leading drivers in adoption and continual participation of zero-tillage systems throughout Brazil, especially as no economic mechanisms are available to promote the implementation of zero-tillage as a mitigation strategy of global warming. This study was limited to the number of

sites available under long-term zero-tillage (>20 years) and the number of samples which could be transported to the University of Nottingham. In order to inform policy and evaluate the potential of zero-tillage as a tool for mitigation of climate change, further assessment, across a wider geographical area, including seasonal measurements is needed, but these initial data suggest much promise in the approach.

5.0 Long-term zero-tillage enhances the physical and chemical protection of soil carbon

This chapter extends on the work from Chapter 4 and investigates the mechanisms in relation to carbon storage in Brazilian soils. This chapter focusses in greater detail on soil structure at the microscale and assesses the relationship between soil physical protection of carbon in different aggregate size classes. Also, this chapter identifies the potential for further accumulation or release under climate warming by assessing carbon thermostability using Rock-Eval pyrolysis.

5.1 Introduction

The amount of CO_2 released to the atmosphere from agricultural soils is mainly dependent the rate of soil organic carbon (SOC) formation versus decomposition (Trumbore, 1997). Conventional agricultural practices, such as ploughing/conventional tillage accelerate the loss of SOC by increasing the oxygen concentration in the soil profile, destroying soil aggregates and exposing organic carbon for mineralisation (Liu et al., 2006). Zero-tillage, an agricultural practice which minimises soil disturbance, can increase soil aggregation and may accumulate soil organic carbon (Liu et al., 2006; Luo et al., 2010a; West and Post, 2002), which is critical in meeting global targets for soil carbon sequestration (Minasny et al., 2017). However, results from studies on the impact of zero-tillage on soil carbon storage to date have been inconsistent,

finding both significant increases and decreases compared to conventional tillage following adoption (Luo et al., 2010b). In addition, few studies have characterised the chemical composition and thermostability of organic carbon in long-term zero-tilled soils. Therefore, there remains a large gap in our current understanding of the mechanisms which control carbon storage and determine future susceptibility to mineralisation or accumulation (Bongiorno et al., 2019; Sainepo et al., 2018).

The dynamic processes involved in the transformation of organic matter are highly sensitive to environmental conditions. In wet sub-tropical regions, organic matter is estimated to decompose at an annual rate of 3.2%, approximately three times faster than in temperate zones (1.0%) (Nogueirol et al., 2014). Changing from conventional tillage to zero-tillage alters the soil physical structure and influences the arrangement of solid particles (mineral and organic matter) and pores in which microbial decomposers, gases and soluble compounds are located (Mangalassery et al., 2014). The interactions between the organic matter and decomposing microorganisms, or their enzymes, drives organic matter mineralisation at the micrometre scale. X-ray Computed Tomography (CT) offers a way to non-destructively, measure the size and shape of the pores and their degree of connectivity enabling assessment of the susceptibility of organic matter to mineralisation (Galdos et al., 2019).

In addition, the carbon storage potential of a soil depends upon the level of aggregation. Zero-tillage has been hypothesised not only to increase the proportion of macroaggregates but also the number of microaggregates formed within macroaggregates (Six et al., 1999). Previous studies have identified that microaggregate formation within macroaggregates is crucial for the long-term sequestration of carbon as microaggregates have a greater capacity to protect carbon against decomposition compared to macroaggregates (Coleman et al., 2007; Kumar et al., 2013). It is important not only to assess the quantity of organic carbon but also the thermostability within different aggregates. Labile organic matter turnover occurs within a day to a year and is highly influenced by soil management practices, while stable organic matter turnover occurs on timescales ranging from decades to centuries (Feng et al., 2014). Both fractions can be found in aggregates of all sizes and contribute to the regulation of organic carbon storage duration (Bongiorno et al., 2019). Advances in analytical techniques now allow us to provide a detailed description of the organic carbon structure. Rock-Eval pyrolysis has previously been used to characterise organic matter thermostability in forest soils (Soucémarianadin et al., 2018), mountainous soils (Saenger et al., 2013) and tropical peatlands (Cooper et al., 2019; Girkin et al., 2019) but to date has not been used in agricultural soils under contrasting management to understand carbon dynamics.

This study aims to understand the relationship between soil physical protection of carbon in different aggregate size classes and identify the potential for further accumulation or release by assessing carbon thermostability under two different

agricultural systems. It was hypothesised that, (i) the minimised mechanical disturbance associated with zero-tillage will regenerate the soil porous architecture and increase soil porosity through the development of continuous pores. (ii) This will result in a greater number of stable macroaggregates, which will increase in longer-term under zero-tilled soils, (iii) there will be a greater fraction of microaggregates formed within macroaggregates in zero-tilled soils, as the rate of macroaggregate formation and degradation is reduced under zero-tillage and (iv) zero-tilled aggregates will contain more organic carbon than conventionally tilled aggregates, with an increase in the "labile" proportion due to a reduction in aggregate porosity, therefore, limiting microbial access.

5.2 Materials and methods

5.2.1 Sites and soil sample collection

Soil samples were collected in November 2017 from experimental fields at the São Paulo State University (UNESP) at Botucatu, São Paulo, Brazil (22°46'S, 48°25'W). The climate is classified as mesothermal with dry winters, and the dry season is well defined from May to September, with a yearly average rainfall of 1450 mm, distributed typically between October and April. Soil samples were taken from six fields, of which three had been zero-tilled for varying lengths of time, and three paired conventionally tilled fields (details are outlined in Table 5.1). All zero-tilled soils had been managed this way for between 2 and 31 years, whereas the conventionally tilled soils were subjected to annual mechanical turnover to a depth of at least 10 cm.

From each field, using a randomised design, six intact soil cores (5 cm diameter × 30 cm depth) were collected using a manual core sampler that used transparent sample liner tubes (Van Walt Lts, Haslemere, UK) for X-ray Computed Tomography imaging. The soil cores were first sealed with paraffin wax at the top and bottom to prevent movement during transit. To account for changes in bulk density between managements, soil samples were collected to a depth of 50 cm at 5 cm increments using a stainless-steel cutting cylinder (7 diameter cm × 4 cm height). Simultaneously, bulk soil samples were collected at each increment for measurements of soil organic carbon, aggregate size distribution and soil moisture content.

Table 5.1 Selected soil and management characteristics of experimental fields. ZT and CT refer to zero and conventional tillage, respectively.

Field	Management	Soil texture (% clay)	Crop rotation (2017/18)	Grain yield (Mg ha^{-1})
1	15–20 cm CT	65	Triticale/Maize	2.5 / 10.2
2	15 years ZT	65	Triticale/Soybean	2.0 / 1.8
3	15–20 cm CT	62	Maize/Soybean	8.5 / 2.7
4	31 years ZT	62	Maize/Soybean	8.5 / 2.9
5	10–15 cm CT	62	Grasses/Maize	6.0
6	2 years ZT	62	Grasses/Maize	6.0

5.2.2 X-ray Computed Tomography

Following the sealing, samples were transported to the Hounsfield Facility at the University of Nottingham for X-ray CT scanning. Before this, a pilot study had been conducted which involved preserving the samples as described here and then transporting soil samples for X-ray CT scanning initially in Brazil and then again at the Hounsfield Facility, in the U.K. This confirmed that the addition of paraffin wax was an excellent approach for preventing sample disturbance during transit (Galdos et al., 2019).

Soil cores were scanned using a Phoenix v|tome|x M scanner (GE Measurement and Control Solution, Wunstorf, Germany). This allowed visualisation and quantification of the soil porous architecture. Voxel resolution was set to 50 µm, with a potential energy of 160 kV and a current of 180 µA. Soil cores were scanned in three sections (i.e. depths), with a total scan time of 30 minutes per soil core. A total of 2998 image projections were captured for each core. After scanning, each soil core was dismantled into two depths, 0 – 20 cm and >20 cm and the soil passed through a series of sieves of 2, 0.25- and 0.053-mm aperture while subjected to horizontal shaking for four minutes at 250 rotations $minute^{-1}$. Three randomly-selected aggregates, retained between the 2 and 0.25 mm per core at each depth, were scanned using a Phoenix Nanotom (GE Measurement and Control Solution, Wunstorf, Germany), where voxel resolution was 1.51 µm, with potential energy set at 90 kV and a current of 65 µA. A total of 1440 projection images were collected with a total scan time per sample of 69 minutes.

Note a longer scan time was required to achieve a higher image quality at the higher resolution.

Scanned images were reconstructed at 32-bit using Phoenix Datos x 2 reconstruction software. Due to the irregularity of soil surface between cores, the first 50 image slices from the top of each soil core was excluded from the analysis. Scanned images were optimised to correct for any movement of the sample during scan and noise was reduced using the beam hardening algorithm in Datos x 2, set at level 8. As a multi-scan routine was performed for the soil cores, to enable a higher spatial resolution than scanning the whole column in a single scan with a larger field of view, VG StudioMax 2.2.5 was used to merge the three scans to obtain a single 3D volume for the complete core. A total of 36 aggregates (6 from each treatment) were randomly selected from between the 2- and 0.25-mm sieves and conserved in sealed containers in the dark at room temperature.

5.2.3 Image analysis

Initial image analysis of the soil pore morphology was performed using *Image J* (Schneider et al., 2012). A uniform region of interest (ROI) was defined for each soil core and aggregate; 50 × 50 × 50 mm and 0.500 × 0.650 × 0.480 mm, respectively. Soil core ROIs were centrally positioned to limit inclusion of stones and any cracks that may have been introduced during the sampling procedure. Cubic ROIs for the aggregates were not possible because of their

inconstant geometry, so the same ROI, the largest possible in all aggregates was chosen, and the coordinates of these areas were adapted for each image volume/sequence. Once ROIs had been determined, several stages of image processing occurred; (i) cropping image volume to the ROI; (ii) enhancing the contrast/brightness by 0.35%; (iii) application of a 2-pixel median filter; (iv) converting the image volume to 8-bit format and (v) saving the new image volume. Analysis of pore morphology within the soil cores and individual aggregates was determined using the bin bi-threshold approach by Vogel and Kretzschmar (1996) using the open-source software Quantlm (http://www.quantim/ufz.de/). For a more detailed methodology, refer to Bacq-Labreuil et al., (2018).

5.2.4 Dry aggregate size distribution

Subsamples of 100 g of field moist soils were sieved with an 8 mm sieve, air-dried and stored at room temperature. The samples were placed on top of a nest of sieves, comprising of 4, 2, 0.250- and 0.053-mm. Aggregates were slowly separated by horizontal oscillations (150 rounds per minute) for 30 seconds. The aggregates remaining on each sieve were weighed and the dry mean weight diameter (dMWD) calculated:

$$dMWD = \sum_{i=l}^{n} xi\ wi\ , \qquad \text{(Equation 5.1)}$$

where wi is the weight percentage of each aggregate size class with respect to the total sample and xi is the mean diameter of each aggregate size class.

The proportion of microaggregate (0.053-0.250 mm) weight within macroaggregates (0.250-2 mm) was calculated following Six et al., (2000). In brief, macroaggregates (10 g) were immersed in deionised water on top of a 0.25 mm screen and gently shaken with 50 glass beads (dia. = 3 mm). Continuous and steady water flow through the mesh ensured that microaggregates were immediately flushed onto a 0.053 mm sieve. Sub-samples of the sieved aggregates and the microaggregates within macroaggregates (50 mg) were taken for analysis using Rock-Eval pyrolysis.

5.2.5 Rock-Eval 6 pyrolysis

Rock-Eval pyrolysis is a technique used to trace bulk changes in organic matter composition and degree of composition (Disnar et al., 2003; Newell et al., 2016). It predicts soil carbon contents reliably and is an appropriate tool for assessing the vulnerability of soil organic carbon stocks to microbial degradation (Saenger et al., 2013; Soucémarianadin et al., 2018; Upton et al., 2018). Surface (0-20 cm) and subsurface (20-50 cm) soil samples were analysed using a Rock-Eval (v6) analyser. Freeze-dried powdered soil samples (60 mg) were heated at 300 °C for three minutes before an increase in temperature to 650 °C at a rate of 25 °C per minute in an inert N_2 atmosphere. Residual carbon was subsequently oxidized from 300 °C to 850 °C at a rate of 20 °C per minute. The release of hydrocarbons during the two-stage pyrolysis process was detected by a flame

ionisation detector, with an infrared cell detecting the release of CO and CO_2 during the thermal cracking of the organic matter. The Rock-Eval analysis generated a range of standard parameters, including:

- Total organic carbon (TOC_{RE}) is calculated from the sum of the carbon moieties (HC, CO and CO_2).
- The Hydrogen Index (HI mg HC g^{-1} TOC), a measure of hydrocarbons released relative to TOC, was calculated from S2 × 100 / TOC_{RE}.
- The Oxygen Index (OI mg O_2 g^{-1} TOC), corresponding to the amount of oxygen released as CO and CO_2 relative to TOC_{RE}, was calculated from S3 × 100 /TOC_{RE}.

For estimating fractions of organic matter, areas are calculated between the following respective bounds (or nodes): 200–340 °C for A1 (labile biopolymers), 340–400 °C for A2 (resistant biopolymers), and 400–460 °C for A3 (immature geopolymers). A fourth area is calculated, A4, which stands for refractory geopolymers (> 460 °C). Soil organic matter thermal stability was analysed through the calculation of the R and I indices with the I-index assessing the preservation of thermally labile immature SOM (I for immature) and the R-index assessing the contribution of thermally stable SOM corresponding to the most persistent or refractory SOM fraction (R for refractory). Both indices are calculated according to the following equations (Sebag et al., 2016):

$$R - index = (A3 + A4 + A5)/100, \quad \text{(Equation 5.2)}$$

$$I - index = log_{10}\left(\frac{A1+A2}{A3}\right). \quad \text{(Equation 5.3)}$$

5.2.6 Estimating carbon stocks

Total carbon of the bulk soil was determined from 20 mg of oven-dried, ball-milled soil combusted using a total element analyser (Flash EA 1112, CE Instruments, Wigan, UK). Soil carbon stocks were estimated by an equivalent soil mass (ESM) procedure to calculate stocks in multiple soil layers (Mg C ha^{-1}) within a defined area using calculations from Wendt and Hauser (2013). This method quantifies and corrects for the fixed depth error associated with calculating carbon stocks as the product of soil bulk density, depth and concentration.

5.2.7 Statistical analysis

All statistical analysis was performed using the R software (R version 3.4.1) and all data checked for normality. If the residuals from fitted models did not appear consistent with a normal distribution, then a Box-Cox transformation was applied, using the BOXCOX procedure in the MASS package for the R platform (Venables and Ripley, 2002). Analysis of variance (ANOVA) was performed on all primary variables with land management, fields and depth as factors. Due to the nature of the field sampling, and the limitations of the number of fields under different ages under zero-tillage, the soil cores represented pseudo-replicates when assessing the length of time under zero-tillage on soil properties.

5.3 Results

5.3.1 Effect of management on soil pore structure

5.3.1.1 3D image assessment at the core scale

There was a significant difference in pore size characteristics between zero-tilled fields (Fig. 5.1a, b). In soils that had been under zero-tillage for 31 years, a significantly larger proportion of porosity were smaller pores (<0.49 mm) compared to soils under zero-tillage for 2 and 15 years. In these soils, a significantly larger proportion of porosity was accounted for by larger pores (>1.2 mm) ($F_{2,222} = 3.8$, $p<0.05$, Fig. 5.1a and $F_{2,222} = 5.3$, $p<0.01$, Fig. 5.1b).

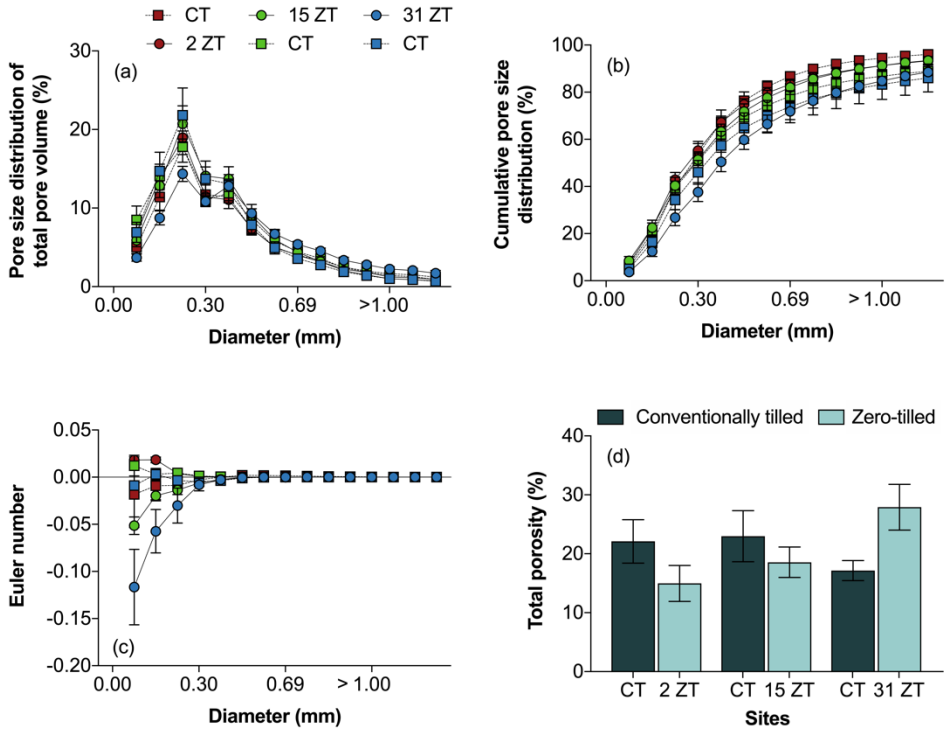


Figure 5.1 Soil pore characteristics of soils under different tillage systems at the core scale (50 µm resolution). (a) The pore size distribution of total pore volume, (b) cumulative pore size distribution, (c) the Euler number and (d) total porosity measured by X-ray Computed Tomography of each management. Means ± 1

SEM (n = 6). Whereby, CT is conventional tillage, and 2 ZT, 15 ZT and 31 ZT represent 2, 15 and 31 years in zero-tillage, respectively.

There was a significant difference in pore connectivity between zero and conventionally tilled management (Fig. 1c, $F_{2,444} = 8.2$, $p<0.01$) with zero-tilled soils having a higher degree of connected pores. There was also a significant difference in pore connectivity between the zero-tilled soils ($F_{2,222} = 10.2$, $p<0.01$) where soils that had been in zero-tillage for 15 and 31 years had more connected pores than soils in zero-tillage for 2 years. There was no significant difference between the conventionally tilled soils ($p>0.05$).

The total porosity for recently converted zero-tilled soils was 14% and significantly increased to 18 and 27% in long-term zero-tilled soils (15 and 31 years, respectively) (Fig 5.1d, $F_{2,12} = 8.2$, $p<0.01$) compared to a mean soil porosity of 21% across all conventionally tilled soils.

5.3.2 Soil aggregate properties

There was a significant difference in mean weight diameter (MWD) between zero and conventionally tilled management associated with sampling depth (Fig. 5.2a, $F_{2,168} = 256$, $p<0.01$) with zero-tilled soils having a larger MWD, which decreased with depth. When taking different fields into count, there was no significant difference in MWD between conventionally tilled fields, but there was a significant difference between the zero-tilled fields. The MWD was

significantly greater in the fields that had been under zero-tillage for 15 and 31 years (Fig. 5.2a, $F_{2,84} = 610$, $p<0.01$).

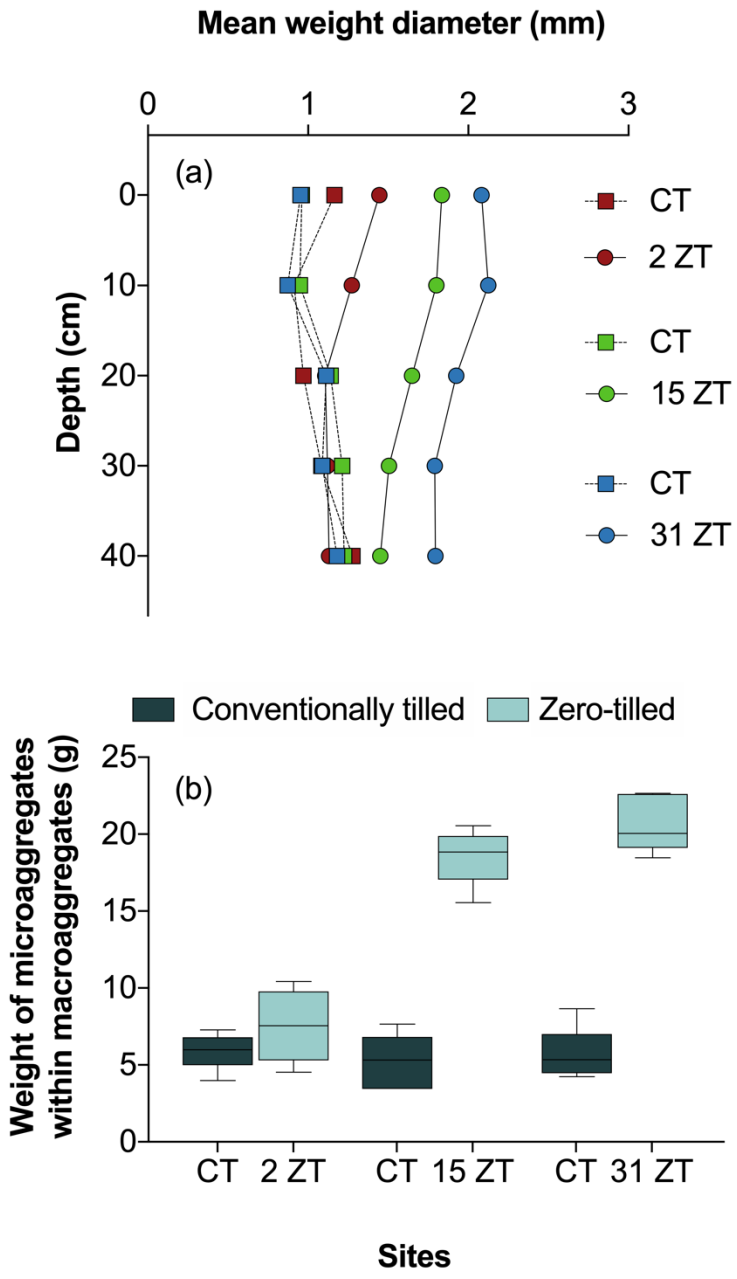


Figure 5.2 Soil aggregate characteristics under conventional and zero-tillage. (a) Mean weight diameter throughout the soil profile to a depth of 50 cm (SEMs ranged from 0.007 to 0.06mm, Table C1) and (b) the weight of microaggregates

(53-250 μm) within macroaggregates (250-2000 μm). Means ± 1 SEM (n = 6). Whereby, CT is conventional tillage, and 2 ZT, 15 ZT and 31 ZT represent 2, 15 and 31 years in zero-tillage, respectively.

There was a significant difference in mM (the proportion of microaggregates within macroaggregates) between zero and conventionally tilled management (Fig. 5.2b, $F_{2,30} = 291$, $p<0.01$) with a greater proportion in zero-tilled sites. There was no significant difference between the conventionally tilled fields ($p>0.05$), but there was a significant difference between zero-tilled fields ($F_{2,15} = 77.3$, $p<0.01$). The proportion of mM was greater at the sites that had been under zero-tillage for 15 and 31 years.

5.3.2.1 3D image assessment at the aggregate scale

Porosity in macroaggregates significantly differed between the two managements ($F_{2,12} = 43.9$, $p<0.01$, Fig. 5.3a, b) with conventionally tilled macroaggregates having an average porosity of 11.0% compared to 1.8% in zero-tilled macroaggregates. There was no significant difference between the three conventionally tilled fields ($p>0.05$) but there was a significant difference between the three zero-tilled sites ($F_{2,12} = 6.3$, $p<0.05$). The porosity in macroaggregates decreased from 6 to 4 to 2 % in macroaggregates that had been in zero-tillage for 2, 15 and 31 years, respectively.

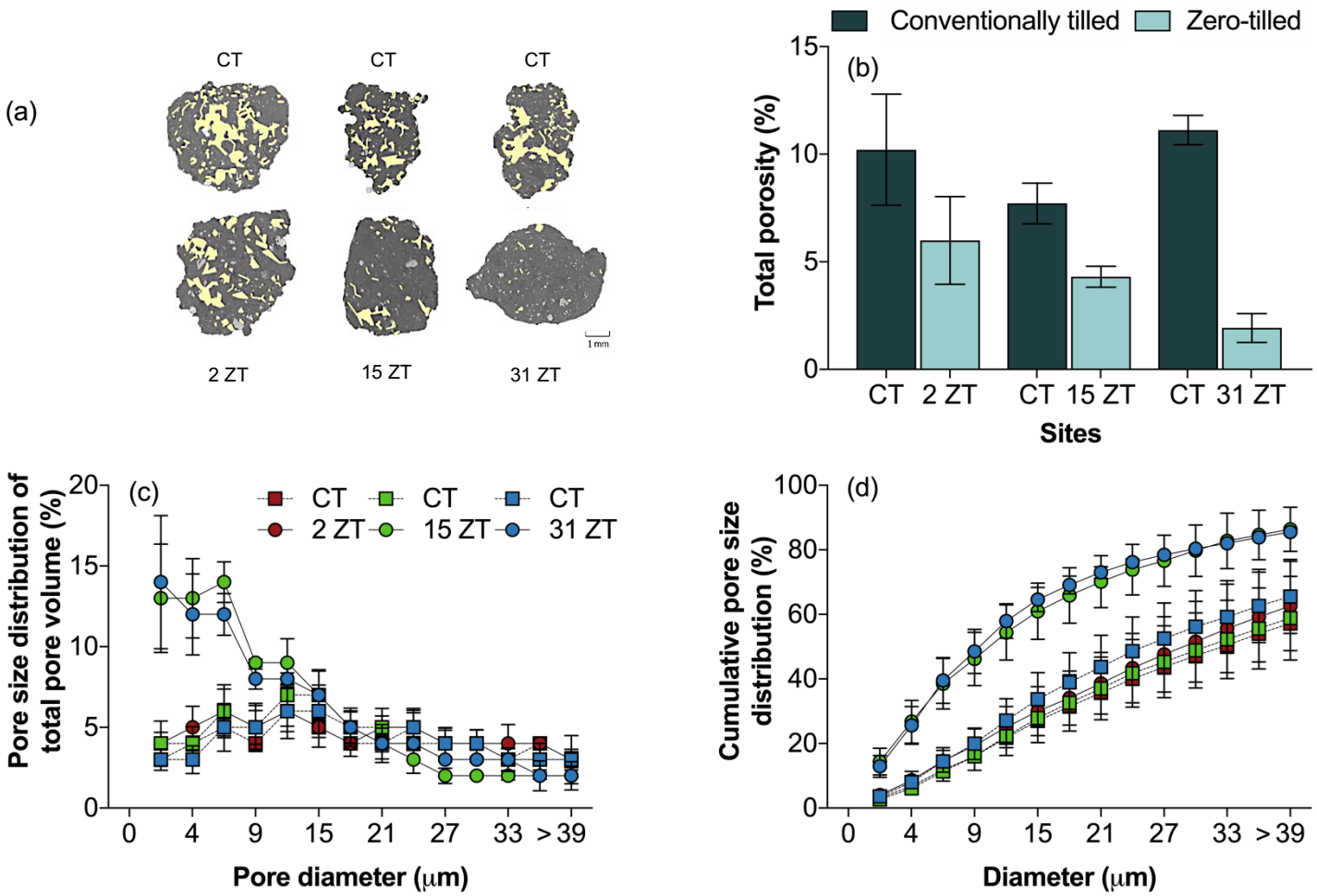


Figure 5.3 Soil pore characteristics of soils under different tillage systems in macroaggregates at 1.5 μm resolution in the top 20 cm. (a) Representative microtomographic images for soil macroaggregates under different tillage systems where yellow is identified as the pore space, (b) total porosity, (c) pore size distribution of total pore volume and (d) cumulative pore size distribution. Means ± 1 SEM (n = 3). Whereby, CT is conventional tillage, and 2 ZT, 15 ZT and 31 ZT represent 2, 15 and 31 years in zero-tillage, respectively.

Soil aggregates under zero-tillage for 15- and 31-years treatment had a larger percentage of total porosity accounted for by smaller pores (<21 μm) compared to conventionally tilled soils, where a larger percentage of total porosity was accounted for by larger pores (> 39 μm). There was a significant difference in the pore size distribution of total pore volume between the two managements in pores smaller than 21 μm ($F_{2,138}$ = 29.8, $p<0.01$, Fig. 5.3c). There was no significant difference among the three conventionally tilled sites ($p>0.05$, Fig. 5.3c) but there was a significant difference among the three zero-tilled treatments ($F_{2,69}$ = 9.5, $p<0.01$, Fig 5.3c). The same trend emerged for cumulative pore size distribution (Fig. 5.3d). There was no significant difference in macroaggregate pore characteristics from 20-50 cm depth between managements or with time under zero-tillage management ($p>0.05$).

5.3.3 Effect of management on soil carbon storage

5.3.3.1 Land use effects on aggregate associated carbon

Soil carbon stocks, on an equivalent soil mass basis, were significantly larger in zero-tilled soils compared to conventionally tilled soils (Fig. 4a, $F_{1,96}$ =318, $p<0.01$). There was also a significant difference between the zero-tilled fields, where carbon stocks significantly increased in soils under zero-tillage for 15 and 31 years compared to 2 years (Fig. 4a, $F_{2,48}$ = 172, $p<0.01$).

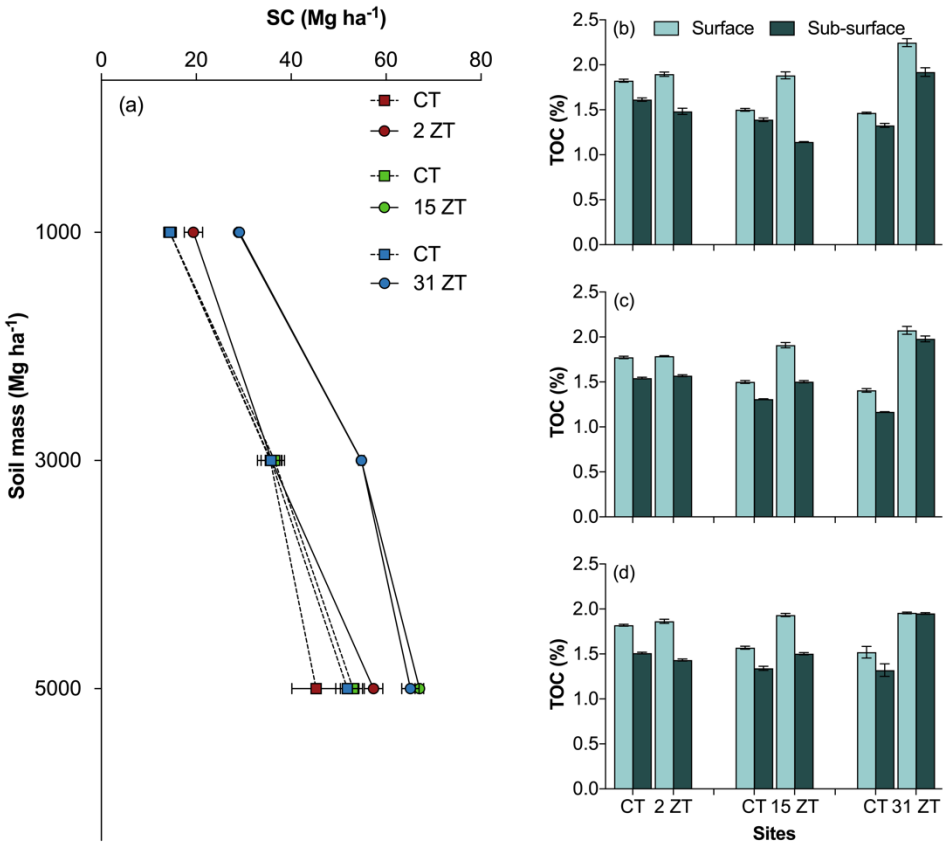


Figure 5.4 (a) Carbon stocks in conventional and zero-tilled soils to a depth of 50 cm and aggregate associated organic carbon under different tillage systems in a) macroaggregates, (b) microaggregates and (c) microaggregates within macroaggregates. Means ± 1 SEM (n = 3). Whereby, CT is conventional tillage and, 2 ZT, 15 ZT and 31 ZT represent 2, 15 and 31 years in zero-tillage, respectively.

In all three aggregate size classes, TOC was significantly larger in zero-tillage compared to conventionally tilled aggregates. Surface TOC was significantly larger in all aggregate size classes compared to sub-surface aggregates. In surface aggregates, the largest increase in TOC was in 31-year zero-tilled soils,

where TOC increased from 1.4 to 2.2% in the macroaggregates (Fig. 5.4b), from 1.4 to 2.1% in the microaggregates (Fig. 5.4c) and from 1.5 to 2.0% in the microaggregates formed within macroaggregates (Fig, 5.4d).

5.3.3.2 Influence of tillage methods on the thermostability of soil organic carbon

Surface macroaggregates in recently converted zero-tilled soils (2 years) had a significantly greater proportion of thermolabile carbon compared to conventionally tilled soils (Fig. 5a, $F_{1,24} = 104$, $p<0.01$).

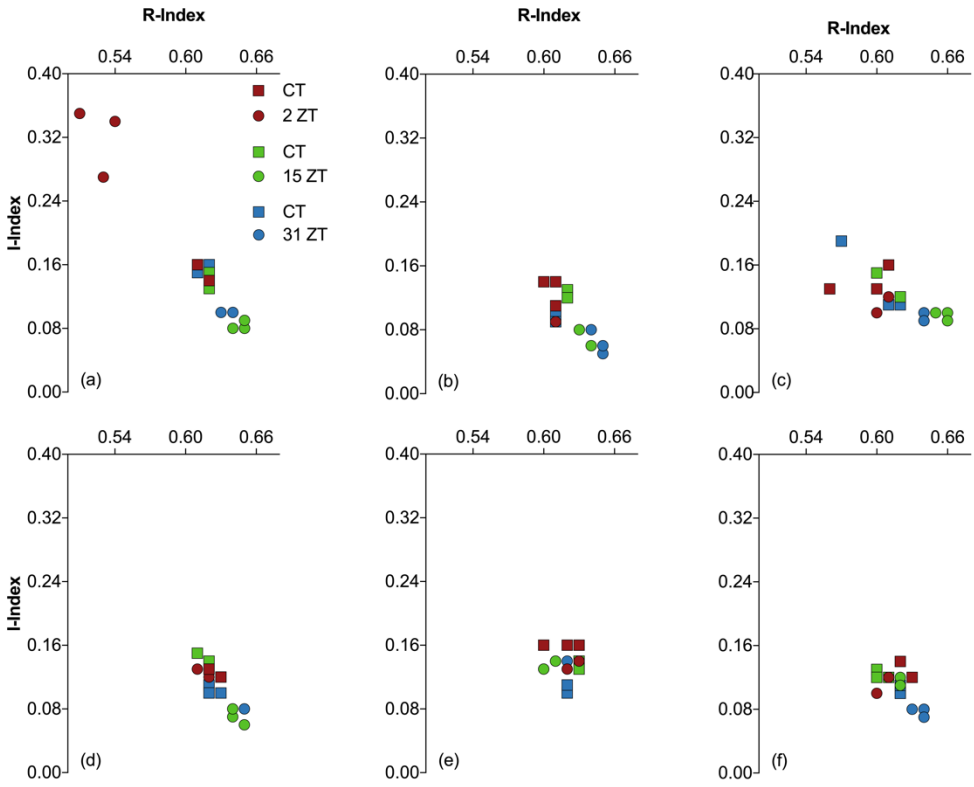


Figure 5.5 I and R indices for (a,d) macroaggregates, (b,e) microaggregates and (c,f) microaggregates within macroaggregates (mM) in (a-c) surface 0-20 cm and (d-f) sub-surface 20-50 cm aggregates. Means ± 1 SEM (n = 3).

There were significant differences in the R and I indices among all aggregate size classes in zero-tilled soils, with more humified carbon measured in aggregates that had been in zero-tillage for 15 and 31 years. (Fig 5.5a, $F_{2,12}$ =155, $p<0.01$ & Fig 5.5b, $F_{2,12}$ = 19, $p<0.01$ & Fig 5.5c, $F_{2,12}$ = 5.3, $p<0.05$). Similarly, in sub-surface aggregate classes, the longer-term zero-tilled macroaggregates had a significantly greater proportion of thermostable organic carbon compared to conventionally tilled and recently converted zero-tilled aggregates (Fig. 5.5d, $F_{1,24}$ = 46, $p<0.01$). There was no significant difference in thermostability of organic carbon between land managements in sub-surface microaggregates or mM aggregates (Fig 5.5e-f, $p>0.05$).

5.4 Discussion

5.4.1 Influence of tillage management on soil structural properties

The absence of mechanical disturbance to invert topsoil increased soil surface compaction reduced soil porosity and increased bulk density (Fig. 5.1d) when soils had been in zero-tillage for two years. This was also found by Matula, (2003) and Romaneckas et al., (2009) who observed that the reduced total soil porosity decreased water infiltration rates and noted a significantly lower root mass measured compared with conventionally tilled soils. The surface soil compaction under zero-tillage can impede root development and the growth of the primary root axes (Martínez et al., 2008).

However, in line with the first hypothesise, ‘minimised mechanical disturbance associated with zero-tillage will regenerate the soil pore architecture through the development of continuous pores’, it was showed that soil structure under long-term zero-tillage (15 and 31 years) improved considerably with a significant increase in soil porosity and pore connectivity (Fig. 5.1c, d). These findings are in line with those from Galdos et al., (2019) who showed that soil under zero-tillage for 30 years had more connected pores than neighbouring conventionally tilled soils in Brazil. The absence of soil inversion and disturbance in zero-tilled systems increases bioturbation in which soil organisms, including microbes, rooting plants and burrowing animals alter the soil structure and contribute to the development of a system of continuous pores (Dignac et al., 2017; Piron et al., 2017).

Total porosity, which influences the water holding capacity, gas exchange and microbial activity, is usually greater in soils under conventional tillage than under recently converted zero-till soils (Skaalsveen et al., 2019). However, such studies generally only consider the topsoil and do not include measurements below the plough layer. This study considers the differences in soil structural properties down to 50 cm. Indeed, in recently converted zero-tilled soils (two years), the total porosity decreased by 7% in the top 50 cm, however, after 31 years in zero-tillage, the total porosity was 13% greater than the paired conventionally tilled soils (Fig. 5.1d). Studies by Hangen et al., (2002) and He et al., (2009) demonstrated that long-term zero-tilled soils had much deeper percolation, due to more favourable conditions for burrowing soil animals providing deep vertical macropores. They are, therefore increasing

macroporosity and total porosity. The results here indicate that soil structure regenerates under long-term zero-tillage and increases connectivity of pores which will positively impact root growth, water dynamics and soil gas fluxes.

5.4.2 Effect of tillage management on soil aggregate formation and structure

Changes in the soil physical characteristics associated with the adoption of zero-tillage generally resulted in the improved aggregate formation, especially when implemented for at least 15 years across the soils in Botucatu, Brazil. Figure 5.2a shows the effect of different tillage systems on aggregate stability down to 50 cm and supports the second hypothesis, that 'zero-tillage will result in a greater number of stable macroaggregates, which will increase in long-term zero-tilled soils'. In general, aggregates are considered 'stable' if the mean weight diameter is greater than 1.3 mm and stable aggregates play a central role in protecting pools of carbon and nitrogen, providing microhabitats for microorganisms and are important in ensuring the soil is resilient against wind and water erosion (Blaud et al., 2017). The results here indicate that long-term zero-tillage improved aggregate stability in the top 50 cm compared to conventionally tilled soils (Fig. 5.2), through reducing disturbance by tillage, maintaining surface residue cover, increasing organic binding agents and therefore preventing soil erosion. It was observed in the conventionally tilled soils that the aggregates were either 'unstable' or 'median'. The exposure of the soil surface in conventionally tilled soils results in aggregates breaking apart due to continuous weathering and drying and wetting cycles (Devine et al., 2014;

Kou et al., 2012). Under zero-tillage, crop residue decomposes at a slower rate compared to conventionally tilled soils, leading to a gradual accumulation and increase in the soil organic carbon. The resulting microbial substrate from residue decomposition contributes to stabilising soil aggregates, and this effect is enhanced over time under zero-tillage. A study by Zheng et al., (2018) assessed the effects of tillage practices on aggregate formation after 20 years and concluded that zero-tillage increased aggregate stability by 58% compared to conventionally tilled soils and this resulted in greater aggregate-associated carbon in the zero than in the conventionally tilled treatments.

In contrast to soil porosity at the soil core scale, whereby porosity increased with time under zero-tillage (Fig. 5.1d), porosity significantly decreased at the macroaggregate scale with time under zero-tillage (Fig. 5.3a, b). Very few studies have assessed the impact of zero-tillage at the aggregate scale, focussing primarily on the larger soil 'core' scale, but a significant reduction in aggregate porosity would provide a crucial mechanism for soil macroaggregates to provide physical protection of organic matter by minimising microorganism mineralisation. The pore size distribution (Fig. 5.3d) showed larger differences at the aggregate scale compared to the soil core scale (Fig. 5.1a, b). Long-term zero-tilled soil aggregates (15 and 31 years) had a larger proportion of porosity accounted for by smaller pores (<0.40 μm). This implies that the soil core scale may be suboptimal to observe differences between treatments, but a combination of both scales may be best suited to observe the structural changes associated with different managements.

5.4.3 Effect of tillage management on aggregate associated soil organic carbon

Zero-tilled aggregates had a significantly larger TOC content compared to conventionally tilled aggregates (Fig 5.4), supporting the fourth hypothesis, that 'zero-tilled aggregates contain more organic carbon than conventionally tilled aggregates'. During conventional tillage, the disruptive shearing breaks down the soil aggregates (of any class size), and the organic material that was held within them is mineralised by microbial organisms. Therefore, decreasing the organic carbon content of the soil. It was also observed that the larger carbon content within zero-tilled aggregates translated into a greater total carbon content throughout the soil profile (0-50 cm) (Fig. 5.4a). Beare et al., (2010) and Six et al., (2002) also found higher carbon concentrations in surface samples from zero-tilled soils than from conventionally tilled soils. However, they also observed no significant differences in carbon content between zero and conventional tillage in deeper soil layers (>20 cm). Some researchers have proposed there is an even distribution of carbon throughout the soil profile in conventionally tilled soils, compared to larger carbon concentration found only in surface soils in zero-tillage due to deposition of crop residues, with carbon content decreasing with depth (Powlson et al., 2014). In line with the results, that a greater carbon content was measured in zero-tilled soils which had been in this management for the greatest length of time (Fig 5.4b-d), Álvaro-Fuentes et al. (2008) and Sisti et al. (2004) concluded that long-term zero-tillage can significantly increase the organic carbon content of soil. Moreover, this effect was significantly greater in zero-tilled than in conventionally tilled soils at 0-

100 cm depths. An increase in organic carbon throughout the soil profile in zero-tilled soils has previously been attributed to the increase in soil organic carbon in stable macroaggregates (Song et al., 2016), and partly due to bioturbation, specifically via the surface-deposition of soil by anecic earthworms, which are known to move material to lower soil horizons over time (Blouin et al., 2013).

It has been proposed that rates of soil carbon sequestration become smaller as the soil carbon stock approaches a new steady-state, i.e. when soil carbon inputs approximate soil carbon outputs. At this steady state, the soil carbon sink is saturated (Paustian et al., 1997; West and Six, 2007). The proposed period necessary for soil organic carbon to attain steady-state ranges between studies, with some suggesting as few as ten years (Qin et al., 2016) to 100 years (Sauerbeck, 2001), depending on climate and soil type. In this study, the data suggest that a steady-state was reached after 15 years under zero-tillage (Fig. 5.4a).

Highlighting the importance of long-term management strategies, Six and Paustian (2014) postulated that the proportion of microaggregates formed within macroaggregates (mM) could serve as a robust indicator for changes in soil organic carbon under different management practices over decadal time scale. Significantly greater soil organic carbon in the mM fraction of zero-tilled soils have been identified in numerous experiments across varying soil types and climatic regions, signifying that a considerable proportion of the difference in zero versus conventional tillage could be driven by carbon associated within the mM fraction (Arshad et al., 1990). These results confirm our third hypothesis,

that ‘there will be a greater fraction of microaggregates formed within macroaggregates in zero-tilled soils, as the rate of macroaggregate formation and degradation is reduced under zero-tillage’. The majority of macroaggregates in conventionally tilled soils are destroyed upon mechanical disturbance. Therefore, any formation of microaggregates within macroaggregates is also destroyed. The increased amount of soil organic carbon in the mM fraction in long-term zero-tilled soils, in addition to the increase in mean weight diameter, indicates the strong potential for soil aggregation and accumulation of soil organic carbon under zero-tillage compared to conventional tillage.

5.4.4 Impact of tillage methods on the thermal stability of soil organic carbon

It has been speculated that the additional carbon found in zero-tilled soils is predominantly in labile forms (plant residues and particulate organic carbon) that could be decomposed if zero-tillage practices ceased and a farmer reverted to conventional tillage (Powlson et al., 2014). This was the case for the soil from the surface from zero-tillage for two years. Soils in zero-tillage are generally cooler in temperature, wetter, and receive less mechanical disturbance, thus leading to lower macroaggregate turnover rates and more labile soil organic matter in macroaggregates (Salem et al., 2015). In contrast with the fourth hypothesis, ‘zero-tilled aggregates will increase the labile proportion due to a reduction in aggregate porosity and therefore limiting microbial access’, for soils that had been managed as zero-tillage for 15 and 31 years, there was a significantly higher proportion of more recalcitrant and humified organic

carbon. The chemical composition of this type of carbon is more resistant to decomposition by microorganisms, and so tends to turn over more slowly (decades to centuries).

In this study, the thermostability of organic carbon in zero-tilled soils was more resistant to decomposition in microaggregates compared to macroaggregates. According to the aggregate hierarchy concept, macroaggregates form first by primary particles, i.e. crop residues (Tisdall and Oades, 1982). Then, microaggregates are developed within macroaggregates by roots and fungal hyphae fragments. Consequently, macroaggregates generally have a higher proportion of younger soil organic matter (labile material), while microaggregates tend to contain more stable soil organic matter (more humic material).

5.5 Conclusions

Zero-till improved the soil structure through an increase in total porosity and pore connectivity (as revealed by X-ray CT imagery), and increased the stability of soil aggregates. In addition, zero-tillage increased the soil organic carbon content in all aggregate size classes in comparison to conventionally tilled soils. An increase in soil disturbance caused by conventional tillage accelerates soil organic carbon decomposition by altering physical protection and mineralising the labile forms of carbon in soil aggregates. Furthermore, long-term zero-tillage (15 and 31 years) increases carbon stocks further and alters the chemical

composition of the organic carbon stored, which reduces its susceptibility to mineralisation upon any disturbance. Taken together, our results demonstrate the potential of zero-tillage as a management practice to increase carbon sequestration in the longer term. This is important because enhancing carbon sequestration is a key priority is mitigating CO_2 emissions.

Chapter 6 – General discussions and conclusions

6.1 General introduction

Agricultural soils are a significant source of GHG emissions, contributing one fifth of CO_2 emissions globally, one third of CH_4 emissions and two thirds of N_2O emissions (EPA, 2014). Changes in the management of agricultural soils can affect their role as a source or sink in the global carbon cycle, and the scale and composition of their GHG emissions. Therefore, understanding the role of different crop cultivation regimes in regulating these processes is of particular importance. Our current understanding concerning the extent to which zero-tillage reduces GHG emissions and increases soil carbon storage compared to conventional tillage is extensively debated in the literature (Powlson et al., 2014), and there remain several critical knowledge gaps including the temporal trends of management, lability of additional soil carbon in zero-tilled soils and the global warming potential (GWP) emitted from each management when considering all three key biogenic gases.

The first two data chapters of this thesis are based on a temperate, arable agricultural area and is advancing science and our practical understanding in three particular respects. First, we provide a regional-scale assessment of differences between two treatments using model-based statistics and extensive, closely co-located, paired samples of these treatments on commercial, rather than experimental farms. This study therefore, through substantial sample effort and appropriate statistics, provides a unique assessment at scale of the impacts of real-world practices. Second, we integrate this regional-scale assessment with

studies on *in-situ* imaging of soil structure using X-ray Computed Tomography. Finally, we assess differences in soil carbon storage whilst addressing key methodological concerns about past work. There is no other published paper describing research of this kind, at the same time the scope for climate change mitigation via agricultural management has never been more topical.

In Chapter 2, a unique large-scale field study was performed, based on 80 closely located pairs of zero and conventionally tilled fields, covering an area of 6,504 km^2 in the East Midlands, U.K, where zero-tillage had been implemented for between 1–15 years. The results from this study assessed regional scale differences between the two contrasting tillage managements using model-based statistics and paired sampling. The study elucidated the key soil properties affected by zero-tillage management, which in turn regulates GHG emissions and governs the concentration and composition of carbon in soils. In Chapter 3, using a sub-set of the same samples as in Chapter 2, a selection of previously visited fields were sampled for further analysis in July 2016, providing a detailed characterisation of soil quality and microbial activity over two seasons.

The subsequent experiments (Chapter 4 and 5) focussed on zero and conventional tillage impacts on GHG production and mechanisms of carbon storage in Brazil, in a contrasting climate to the previous study in the U.K. This study also used a paired sampling design but implemented on a smaller scale due to site, time and transportation limitations. A contrasting aspect of Brazilian agriculture compared to the UK is that zero-tillage has been a popular method of farming since the late 1950s. As a result, soil samples were collected from

fields which had been in zero-tillage for >30 years in some cases. GHG production and carbon content were measured in these samples to assess whether the temporal trend observed in zero-tilled soils held true within tropical climates. Chapter 5 focussed in greater detail on soil structure at the microscale and assessed the relationship between soil physical protection of carbon in different aggregate size classes and identified potential for further accumulation or release under climate warming by assessing carbon thermostability.

This chapter provides a synthesis of key findings (Table 6.2), in a wider global context, as well as some potential future directions of research to further elucidate the role of zero-tillage in minimising GHG emissions whilst simultaneously increasing soil carbon stocks and improving soil quality.

6.2 Key findings

Table 6.1 The research questions and key findings developed throughout the project.

Research questions	Key findings
Chapter 2: Zero-tillage could offer a long-term strategy to mitigate climate change	
2.1 How does conversion from conventional to zero-tillage alter the GHG balance and potential C storage? 2.2 How do responses vary according to length of time under zero-tillage? 2.3 How do these processes vary at a regional scale?	- Zero-tillage significantly increased N_2O production but this was offset by the large reductions in CO_2 production, reducing GWP by 30% compared to conventionally tilled soils. - When comparing 80 zero-tilled fields against 80 conventionally tilled fields, there are no differences in soil carbon stock. However, this study provides evidence that carbon stocks significantly increased in the top 20 cm, and the difference between the two managements increased with duration in zero-tillage. Simultaneously, net GWP potential of zero-tillage systems decreased further with time. - Surface connected porosity significantly increased to 9.9% compared to 5.0% in conventionally tilled soils. - The model-based analysis suggests that management effects were highly significant, therefore regional scale differences could not be inferred.
Chapter 3: Zero-tillage impacts on soil physical and biological properties in temperate soils	
3.1 How long does it take zero-tillage in temperate soils to improve soil quality? 3.2 Does zero-tillage detrimentally impact soil structure? 3.3 Are GHG emission reductions microbially mediated?	- Changes in soil physical characteristics associated with the adoption of zero-tillage generally resulted in improved soil quality, especially when implemented for at least six years across the East Midlands. - Zero-tillage increases surface consolidation, to 900kPa, but not to a level which restricts root growth or water infiltration (>1500 kPa). Long-term zero-tillage reduces compaction below 30 cm compared to conventionally tilled soils in which a plough pan is often formed. - Microbial biomass and enzyme activity increased in line with decreased CO_2 emissions.

Research questions	Key findings
Chapter 4: The impact of long-term zero-tillage on greenhouse gas emissions and soil bio-physical properties in tropical soils	
4.1 How does conversion to zero-tillage alter GHG emissions and microbial activity across tropical agricultural soils? 4.2 How do these effects vary over time under zero-tillage?	- No difference was observed between N_2O or CH_4 fluxes, but there was significant reduction in CO_2 emissions from zero-tilled soils. Microbial activity was greater in zero compared to conventionally tilled soils. - GWP significantly decreased and microbial activity increased with duration in zero-tillage at Goiãs, São Paulo and Paranã but not at Maranhão.
Chapter 5: Long-term zero-tillage enhances the physical and chemical protection of soil carbon	
5.1 How does the chemical characterisation of organic carbon stored in aggregates differ? 5.2 Is organic carbon thermostability greater in mM within macroaggregates under zero-tillage?	- A greater proportion of more recalcitrant and humified organic carbon was measured in zero-tilled soils. The chemical composition of this type of carbon is more resistant to decomposition by microorganisms, and so tends to turn over more slowly (decades to centuries). - Zero-tilled soils had a greater proportion of microaggregates within macroaggregates, of which the organic carbon had greater thermostability compared to conventionally tilled soils.

6.3 Effect of contrasting tillage systems on soil physical properties and soil hydraulic behaviour

After five years in zero tillage, it is clear that the soil surface becomes more consolidated compared to conventionally tilled soils, as shown by penetration resistance, shear strength and X-ray Computed Tomography data. The increased surface soil consolidation following adoption of zero-tillage has been widely reported. For example, Sasal et all, (2006) concluded that soil porosity in conventionally tilled soils was 3.5% greater than in zero-tilled surface soils (0-15 cm). In addition, through X-ray Computed Tomography analysis, they demonstrated zero-tilled soils had a porosity of 14% in the top 10 cm, compared to 20% in conventionally tilled soils. However, the degree of surface consolidation reported in chapter 3 and 4 in zero-tilled soils suggests that crop root growth will not be restricted as penetration resistance is less than 1500 kPa, and the increased consolidation combined with the retention of surface crop residues will act to considerably reduce soil erosion (Montgomery, 2007). Approximately 2.2 million tonnes of topsoil is eroded annually in the UK and over 17% of arable land shows evident signs of erosion (Graves et al., 2015). Currently, only 8% of cultivated land is in zero-tillage in the U.K. (Eurostat, 2008), but an increase of this would reduce soil erosion. In Brazil, at least 45% of cultivated land is in continuous zero-tillage and it is estimated to have reduced soil erosion from 3.4-8.0 t/ha under conventional tillage compared to 0.4 t/ha in zero-tillage (Derpsch, 2008).

When soils had been in zero-tillage for at least 11 years, penetration resistance decreased between 35-60 cm, compared to adjacent conventionally tilled soils in which a plough pan was evident. Simultaneously, surface connected porosity

significantly increased to 9.9% compared to 5.5% in conventionally tilled soils. We demonstrated that soil porosity initially declined by 13.8 %, after the adoption of zero-tillage (1–5 years) due to consolidation via a lack of disturbance. Importantly however, this did not limit water storage or transport down through the soil profile as significantly greater soil moisture content and saturated hydraulic conductivity was observed in long-term zero-tilled soils compared to conventionally tilled soils. In zero-tilled soils the reduction in the total soil porosity will reduce the risk of runoff during heavy rainfall through the development of a highly effective, surface-connected porosity. Greater water content in zero-tilled soils may be influenced by enhanced retention of crop residues which will limit surface runoff and reduce evapotranspiration losses (Liu et al., 2013). Moreover, the increased surface residue on the surface provides organic matter for soil macrofauna, particularly earthworms, which loosen the soil through burrowing activities and create continuous macropores (biopores), which are otherwise destroyed by tillage operations (Soane et al., 2012). These biopores can represent as low as 0.23-2.00% of the total soil volume, but may account for 74-100% of the total water flux (Alaoui and Helbling, 2006) and can potentially reduce runoff by 48% in the UK (Harris et al., 2004). Similarily, in the state of Parana, Brazil, research showed a reduction of 50% in water loss from zero-tilled soils through reduced evaporation (de Freitas and Landers, 2014).

6.4 The role of microbial activity and soil aggregates in transforming, protecting and increasing soil carbon stocks

Chapters 2 and 5 highlight increased soil organic carbon stocks in zero-tilled compared to conventionally tilled soils. These increases are likely to be both microbially and physically mediated. Chapter 2 demonstrates strong shifts in soil functional organic chemistry with conversion to zero-tillage, indicating an increase in the proportion of sequestered recalcitrant carbon compared to conventionally tilled soils. The increased crop residue retention on the surface of zero-tilled soils provides a continuous supply of organic materials to soil microorganisms, and this is evident in the increased levels of microbial biomass. For examples, across the East Midland samples, conventionally tilled soils had a mean microbial biomass carbon of 425 mg kg^{-1} soil compared to 497 mg kg^{-1} soil across the zero-tilled soil (chapter 3 and 4). Furthermore, the carbon stored in zero-tilled soils is less susceptible to oxidation on disturbance compared to the more labile carbon found in conventionally tilled soils. This in turn increases the longevity of the carbon stored in zero-tilled soils. The increase in recalcitrant compounds e.g. aromatics, in 15 year zero-tilled soils suggests greater preservation of lignin during decomposition of crop residues, and/or enhanced microbial stabilization of organic materials, with both mechanisms contributing to further increasing longevity of stored soil carbon (Mangalassery et al., 2015).

Furthermore, in zero-tilled soils, the turnover of soil aggregates is reduced compared to conventionally tilled soils which may physically protect carbon

within aggregates (Six et al., 2000). X-ray Computed Tomography showed significantly increased soil porosity and pore connectivity in the long-term, but decreased inter aggregate porosity, providing a potential physical mechanism for protection of soil organic carbon in the 0-20 cm layer. Rock Eval pyrolysis indicated that zero-tilled macroaggregates exhibit considerable heterogeneity in organic carbon properties and show contrasting trends with conventionally tilled soils. Measurements of organic carbon thermostability indicate that zero-tilled soils feature a large labile pool comprising increased presence of lignin and polysaccharides. Moreover, soils which had been in zero-tillage for 31 years also showed a shift in chemical composition of organic carbon to a more recalcitrant fraction following long-term zero-tillage, suggesting aggregates were accumulating rather than mineralising soil organic carbon.

In comparison to the zero-tilled soils in the UK, which showed a continual increase in soil carbon throughout the 1-15 years, the zero-tilled soils in Brazil suggest that a steady state was reached after 15 years in zero-tillage. The proposed time period necessary for soil organic carbon to attain a steady state ranges between studies, with some suggesting as few as 10 years (Qin et al., 2016) to 100 years (Sauerbeck, 2001), depending on climate and soil type.

6.5 Climate change mitigation potential under zero-tillage

Six et al. (2004) highlight that climate change mitigation is only possible if the overall impact of zero-till adoption reduces the net global warming potential (GWP) of all three major biogenic gases (CO_2, CH_4 and N_2O). The majority of studies focus primarily on either CO_2 or N_2O fluxes, but rarely all three

combined. In chapter 2, all three gases were measured from 160 intact soil cores, and a significantly greater net GWP under conventional tillage systems (30 % higher than zero-tillage systems) was reported, primarily due to decreases in CO_2 emissions. Results also provided evidence that the net GWP of zero-tillage systems decreased further with time in management. Chapter 4 showed a similar temporal trend from intact soil cores in Brazilian soils, although the magnitude of difference between zero and conventional tillage was considerable smaller in the sandy soils in Maranhão.

Across both the UK and Brazil soils, evidence suggests it is reduced CO_2 emissions that drive the reduction in GWP under zero-tillage. Even though microbial activity and surface crop residues were greater in zero-tilled soils, potential CO_2 fluxes, which broadly reflect the respiration status of the soil, were reduced under zero-tillage compared to conventional tillage. Under conventionally tilled conditions, ongoing soil disturbance can stress microbial communities, preventing them from reaching a steady state, (Giller et al., 1997). Therefore the lack of disturbance in zero-tilled soils allows them to continually develop and function more effectively i.e. their respiration relative to their biomass is reduced (Mangalassery et al., 2015). In addition, the formation and stabilisation of macroaggregates in zero-tillage represents an important mechanism for the incorporation and protection of soil organic matter that would otherwise be oxidised to CO_2 and lost, as in conventionally till practices.

In the scenarios presented throughout this project, zero-till can potentially represent a ‘win-win’ situation, where in addition to CO_2 mitigation, other

important benefits are achieved. Chapter 3 highlights the role of long-term zero-tillage in improving soil quality through increases in microbial biomass, improving soil structure and the protection of land against soil erosion across all soil textures. In addition to these benefits, previous studies have shown that zero-tillage reduces costs and labour requirements. For example, Smith et al., (1998) suggested a 100% conversion to zero-tillage in Europe could mitigate all fossil fuel-carbon emissions from agriculture in Europe and assessments from the United States Department of Agriculture (Creech., 2017) show zero-tillage reduces the number of field operations from an average of seven to one, translating into 8-12 hours per ha saved in tractor time (a 60-90% saving). These additional benefits often result in economic gains for farmers and will be the leading drivers in adoption and continual participation of zero-tillage systems throughout the UK and Brazil, especially as long as no economic mechanisms are available to promote the implementation of zero-tillage as a mitigation strategy of global warming.

The climate mitigation potential of zero-tillage also needs to be considered in the context of short- and long-term impacts on yield because mitigation benefits at one site are of no value if reduced production is compensated for by cultivating more land. To date, the majority of studies report little or no difference in yield between zero and conventional tillage management (Pittelkow et al., 2015). Similarily, a main barrier for widespread adoption of zero-tillage is the potential increase in occurrence of blackgrass and other weeds. There are conflicting reports regarding the use of herbicides under zero-tillage, with some studies reporting an increase of herbicide application (Romaneckas

et al., 2009), whilst others observe the emergence of black-grass becomes less prevalent with time under zero-tillage (Godwin, 2014).

In conclusion, the studies within this thesis highlight the critical role that zero-tillage could play in both temperate and tropical climates, in not only improving soil quality but simultaneously reducing the climate impact of agricultural production. Within the temperate study region, converting to zero-tillage could reduce the GWP by as much as 30% in long-term studies, and in the tropics by 50% after 30 years in zero-tillage. Simultaneously, long-term zero-tillage also increased the amount of carbon stored within the soil profile, when measured on an equivalent soil mass basis, by an average of 0.3 Mg yr^{-1} across all sites. A decrease in aggregate porosity but an increase in aggregate stability was shown in the tropical soils and would be a critical mechanism to store and protect soil carbon. The results also suggest that the zero-tillage systems in this study lead to progressive improvements in several soil parameters, including decreasing soil penetration resistance and increasing aggregate stability in the top 20 cm, these elements are crucial for a successful active crop and good soil quality, by reducing soil erosion, increasing water infiltration and altering soil porosity. Given the urgent need for climate change mitigation to meet the 1.5 °C warming target and avoid almost certain strong negative impacts on yield from climate change, we suggest in our context zero-tillage is one of many important tools to reduce the carbon footprint of food productions and incentives should be incorporated into policy for farmers to be able to fund the initial transition.

6.6 Future research directions

6.6.1 In-situ studies assessing seasonal variability

The work undertaken in this project was primarily carried out in the laboratory, although based on intact soil cores from the field. The results from this study need to be extended to the field, to i) assess the temporal variability of GHG emissions in relation to different seasons and ii) the effect of cropping rotations. These approaches will allow the study of climate change mitigation potential of zero-tillage all year round. Encompassing cropping rotations will also be beneficial to allow in-depth characterisation of organic matter cycling and GHG dynamics which is useful for informing farm managements, and potential future incentive payments.

6.6.2 Studies on aggregate characteristics and nutrient availability

One of the key findings in this work showed the conversion from conventional to long-term zero-tillage significantly decreased porosity within macroaggregates. An important question that arises from this finding is 'will the reduced aggregate porosity limit nutrient bioavailability for plant uptake?' It is currently difficult to predict nutrient availability throughout different seasons and micro climates owing to the complex pathways by which the nutrients in the surface residues are recycled, converted to plant-available forms and incorporated into soil aggregates.

6.6.3 Isotopic labelling of carbon

To further illuminate the myriad of factors that control soil organic carbon dynamics, stable isotopic labelling could be applied to zero and conventionally

tilled soils. Pulse labelling of crops using $^{13}CO_2$ *in-situ* would provide realistic estimates of above and belowground carbon inputs without disturbing root-soil interactions. Moreover, this approach would also isotopically label microbial communities responsible for short and long-term carbon cycling processes within the soil profile.

6.6.4 Nitrogen transformations

The dominant formation pathways of N_2O production under contrasting tillage practices remains unclear. Considering zero-tillage significantly increases N_2O production, and N_2O has a global warming potential c. 300 times that of CO_2, mitigating N_2O emissions under zero-tillage is a key challenge. Further research should therefore focus on the dominant production pathways, particularly as denitrification can also result in N_2 production. 15N labelling or the application of the DENIS system allows two potential approaches for assessing nitrogen transformation in soil.

Appendix A

Table A1: Total arable land area in 2008 and 2014 and total zero-tilled (Europe only) and zero & minimum tilled area (Asia, Africa, Oceania, North and South America) in 2008 and 2014. All figures are shown in '000 ha. a = (Kassam et al., 2015); b = (Eurostat, 2008); c = (ECAF, 2014); d = (Eurostat, 2014); e = (Department for Environment Food and Rural Affairs et al., 2015); *na* = data was not available.

Country	**Continent**	**Area in CA 2008[a]**	**Total arable area 2008[b]**	**Area in CA 2014[a,c,d]**	**Total arable area 2014[c,e]**
Ghana	Africa	0	4300	30	4700
Kenya	Africa	15	5300	33	5800
Lesotho	Africa	0.13	325	2	272
Madagascar	Africa	0	3000	6	3500
Malawi	Africa	16	3000	65	3800
Morocco	Africa	4	8065	4	8130
Mozambique	Africa	9	5200	152	5650
Namibia	Africa	0	800	0.34	800
South Africa	Africa	368	12600	368	12500
Sudan	Africa	10	*na*	10	19823
Tanzania	Africa	0	1000	25	13500
Tunisia	Africa	6	2733	8	2900
Zambia	Africa	40	2949	200	3800
Zimbabwe	Africa	139	4000	332	4000
Azerbaijan	Asia	0	1854	1	1926
China	Asia	1330	109085	6670	106298
Dem. Rep. Korea	Asia	0	2350	23	7100
India	Asia	0	158022	1500	156360

Iraq	Asia	0	4950	15	5034
Kazakhstan	Asia	1330	28641	2000	29395
Kyrgyzstan	Asia	0	1280	0.7	1281
Lebanon	Asia	0	134	1	132
Syria	Asia	0	4736	30	4662
Turkey	Asia	0	21979	45	20706
Uzbekistan	Asia	0	4350	2	4400
Austria	Europe	*na*	*na*	28	1232
Belgium	Europe	0	839	0.27	613
Bulgaria	Europe	*na*	*na*	16.5	3197
Croatia	Europe	*na*	*na*	19	832
Cyprus	Europe	*na*	*na*	0.27	62
Czech Republic	Europe	*na*	*na*	41	2373
Denmark	Europe	*na*	*na*	3	2184
Estonia	Europe	*na*	*na*	42	578
Finland	Europe	160	2257	200	1912
France	Europe	200	18286	300	17166
Germany	Europe	354	11877	146	10904
Greece	Europe	0	2567	7	1600
Hungary	Europe	8	4592	5	3560
Ireland	Europe	0.1	1060	0.2	999
Italy	Europe	80	7171	284	6728
Latvia	Europe	*na*	*na*	11	1101
Lithuania	Europe	*na*	*na*	19	2129
Luxembourg	Europe	*na*	*na*	0.44	60
Malts	Europe	*na*	*na*	0	5
Moldova	Europe	0	1822	40	1817
Netherlands	Europe	0	1060	0.5	670
Poland	Europe	*na*	*na*	403	9518

Portugal	Europe	25	1148	32	707
Romania	Europe	*na*	*na*	583	707
Russia	Europe	*na*	*na*	583	7295
Serbia	Europe	*na*	*na*	4500	123122
Slovakia	Europe	10	1377	35	1304
Slovenia	Europe	*na*	*na*	2	165
Spain	Europe	650	12586	795	7998
Sweden	Europe	*na*	*na*	16	2324
Switzerland	Europe	16	407	17	400
Ukraine	Europe	600	32434	700	32531
United Kingdom	Europe	150	6085	362	4376
Canada	North America	13481	44683	18313	46015
Mexico	North America	23	23519	41	22993
United States	North America	26500	161880	35613	154605
Australia	Oceania	12000	44140	17695	46957
New Zealand	Oceania	162	431	162	590
Argentina	South America	19719	36433	29181	39200
Bolivia	South America	706	4171	706	4472
Brazil	South America	25502	68388	31811	80017
Chile	South America	180	1262	180	1289
Colombia	South America	102	1998	127	1675
Paraguay	South America	2400	3640	3000	4800
Uruguay	South America	655	1476	1072	2411
Venezuela	South America	300	2659	300	2700

Data published by Eurostat (2014) provides information on zero-tillage uptake from European countries. Available data for Asia, Africa, Oceania and North and South America are reported by FAO (2014 & 2011) and Kassam et al., (2015) which provide records on adoption of conservation tillage (zero and minimum tillage).

Appendix B

Table B1 Soil penetration resistance (kPa) in zero (ZT) and conventionally tilled (CT) soils down to a depth of 60 cm. The soils had been in zero-tillage for 1-5 years, 6-10 years, and 10-15 years. Means ± one standard error.

Soil depth (cm)	1-5 ZT	1-5 CT	6-10 ZT	6-10 CT	11-15 ZT	11-15 CT
0-5	685 ± 33.4	460 ± 31.5	669 ± 53.3	505 ± 76.8	882 ± 41.6	530 ± 52
6-10	1040 ± 36.5	696 ± 38.7	1163 ± 54.5	753 ± 86.8	1323 ± 28.7	657 ± 84.2
11-15	1203 ± 44.9	749 ± 40.8	1311 ± 77.9	916 ± 94.7	1482 ± 64.7	913 ± 92
16-20	1303 ± 45.3	911 ± 46.8	1312 ± 84.3	941 ± 80.6	1488 ± 59.6	1159 ± 77.3
21-25	1387 ± 49.6	1090 ± 57.1	1269 ± 105	1059 ± 77.7	1506 ± 59.5	1364 ± 64.1
26-30	1505 ± 43.2	1343 ± 51.7	1538 ± 90.7	1350 ± 76.3	1499 ± 92.6	1555 ± 73.6
31-35	1562 ± 55.9	1605 ± 50.2	1607 ± 96	1576 ± 65.6	1588 ± 62.5	1712 ± 48.6
36-40	1618 ± 51.4	1639 ± 54.7	1615 ± 57.9	1606 ± 78.5	1484 ± 92.8	1691 ± 61.4
41-45	1615 ± 40.4	1582 ± 55.8	1572 ± 53.4	1609 ± 80	1409 ± 87.4	1669 ± 68.3
46-50	1586 ± 49.7	1555 ± 44.7	1723 ± 60.4	1704 ± 92.5	1278 ± 110.5	1726 ± 60.1
51-55	1610 ± 45.8	1602 ± 48.2	1667 ± 69.6	1559 ± 72.3	1352 ± 77.7	1813 ± 81
56-60	1644 ± 54.3	1574 ± 42.6	1782 ± 72	1559 ± 73.3	1425 ± 45.5	1745 ± 74.7

Appendix C

Table C1 Mean weight diameter (MWD) in zero (ZT) and conventionally tilled (CT) soils down to a depth of 50 cm. The soils had been in zero-tillage for 2, 15 and 31 years. Means ± one standard error.

Soil depth (cm)	2 CT	2 ZT	15 CT	15 ZT	31 CT	31 ZT
0-10	1.16 ± 0.007	1.44 ± 0.027	0.95 ± 0.010	1.83 ± 0.020	0.96 ± 0.020	2.08 ± 0.021
10-20	0.91 ± 0.007	1.27 ± 0.055	0.87 ± 0.010	1.80 ± 0.035	0.94 ± 0.022	2.12 ± 0.020
20-30	0.97 ± 0.053	1.10 ± 0.044	1.11 ± 0.013	1.64 ± 0.049	1.14 ± 0.020	1.92 ± 0.026
30-40	1.08 ± 0.046	1.12 ± 0.050	1.08 ± 0.020	1.50 ± 0.030	1.21 ± 0.009	1.79 ± 0.021
40-50	1.27 ± 0.045	1.13 ± 0.031	1.18 ± 0.031	1.45 ± 0.039	1.22 ± 0.020	1.79 ± 0.020

References

Abdalla, M., Osborne, B., Lanigan, G., Forristal, D., Williams, M., Smith, P., Jones, M.B., 2013. Conservation tillage systems: A review of its consequences for greenhouse gas emissions. Soil Use and Management 29, 199–209.

Ahmad, S., Li, C., Dai, G., Zhan, M., Wang, J., Pan, S., Cao, C., 2009. Greenhouse gas emission from direct seeding paddy field under different rice tillage systems in central China. Soil and Tillage Research 106, 54–61.

Ahmed, F., Rafii, M.Y., Ismail, M.R., Juraimi, A.S., Rahim, H.A., Asfaliza, R., Latif, M.A., 2013. Waterlogging tolerance of crops: Breeding, mechanism of tolerance, molecular approaches, and future prospects. BioMed Research International 2013, Article ID 963525.

Alaoui, A., Helbling, A., 2006. Evaluation of soil compaction using hydrodynamic water content variation: Comparison between compacted and non-compacted soil. Geoderma 134, 97-108.

Alskaf, K., Sparkes, D.L., Mooney, S.J., Sjögersten, S., Wilson, P., 2019. The uptake of different tillage practices in England. Soil Use and Management 00, 1–18.

Al-Kaisi, M.M., Douelle, A., Kwaw-Mensah, D., 2014. Soil microaggregate and macroaggregate decay over time and soil carbon change as influenced by different tillage systems. Journal of Soil and Water Conservation 69, 574–580.

Alvarez, R., Alvarez, C.R., Lorenzo, G., 2001. Carbon dioxide fluxes following tillage from a mollisol in the Argentine Rolling Pampa. European Journal

of Soil Biology 37, 161-166.

Álvaro-Fuentes, J., López, M. V., Cantero-Martinez, C., Arrúe, J.L., 2008. Tillage Effects on Soil Organic Carbon Fractions in Mediterranean Dryland Agroecosystems. Soil Science Society America Journal 72, 541-547.

Ambani, D., Kapoor, S., 2016. Impact of short-term tillage on soil properties, weeds dynamics and crop yield of wheat. African Journal of Agriculture 3, 169–177.

Anderson, E., 1987. Corn root growth and distribution as influenced by tillage and nitrogen fertilization. Agronomy Journal 79, 544–549.

Andrews, S.S., Karlen, D.L., Mitchell, J.P., 2002. A comparison of soil quality indexing methods for vegetable production systems in Northern California. Agriculture, Ecosystems and Environment 90, 25-45.

Angers, D.A., Caron, J., 1998. Plant-induced changes in soil structure: Processes and feedbacks. Biogeochemistry 42, 55-72.

Angers, D.A., Eriksen-Hamel, N.S., 2008. Full-Inversion Tillage and Organic Carbon Distribution in Soil Profiles: A Meta-Analysis. Soil Science Society of America Journal 72, 1370-1374.

Arshad, M.A., Schnitzer, M., Angers, D.A., Ripmeester, J.A., 1990. Effects of till vs no-till on the quality of soil organic matter. Soil Biology Biochemistry. 22, 595-599.

ASTM, 1970. Special procedures for testing soil and rock for engineering purposes. 5th ed. West Conshocken, PA: ASTM International.

Aziz, I., Mahmood, T., Islam, K.R., 2013. Effect of long term no-till and conventional tillage practices on soil quality. Soil and Tillage Research 131, 28-35.

Bacq-Labreuil, A., Crawford, J., Mooney, S.J., Neal, A.L., Akkari, E., McAuliffe, C., Zhang, X., Redmile-Gordon, M., Ritz, K., 2018. Effects of cropping systems upon the three-dimensional architecture of soil systems are modulated by texture. Geoderma 332, 73-83.

Baggs, E.M., Rees, R.M., Smith, K. a., Vinten, a J. a, 2000. Nitrous oxide emission from soils after incorporating crop residues. Soil Use and Management 16, 82-87.

Baker, J.M., Ochsner, T.E., Venterea, R.T., Griffis, T.J., 2007. Tillage and soil carbon sequestration-What do we really know? Agriculture, Ecosystems and Environment 118, 1-5.

Balota, E.L., Colozzi-Filho, A., Andrade, D.S., Dick, R.P., 2003. Microbial biomass in soils under different tillage and crop rotation systems. Biology and Fertility of Soils 38, 15-20.

Balota, E.L., Kanashiro, M., Colozzi Filho, A., Andrade, D.S., Dick, R.P., 2004. Soil enzyme activities under long-term tillage and crop rotation systems in subtropical agro-ecosystems. Brazilian Journal of Microbiology 35, 300-306.

The World Bank, 2013. No-till: A climate smart agriculture solution for Kazakhstan http://www.worldbank.org/en/results/2013/08/08/no-till-climate-smart-agriculture-solution-for-kazakhstan (Date accessed: 31/10/2019).

Basch, G., Kassam, A., Gonzalez-Sanchez, E.J., Streot, B., 2012. Making sustainable agriculture real in CAP 2020. The European Conservation Agriculture Federation, Brussels, Belgium.

Bateman, E.J., Baggs, E.M., 2005. Contributions of nitrification and

denitrification to N_2O emissions from soils at different water-filled pore space. Biology and Fertility of Soils 41, 379-388.

Batjes, N.H., 2014. Total carbon and nitrogen in the soils of the world. European Journal of Soil Science 47, 151-163.

Bauer, T., Strauss, P., Grims, M., Kamptner, E., Mansberger, R., Spiegel, H., 2015. Long-term agricultural management effects on surface roughness and consolidation of soils. Soil Tillage Research. 151, 28-38.

Bavin, T.K., Griffis, T.J., Baker, J.M., Venterea, R.T., 2009. Impact of reduced tillage and cover cropping on the greenhouse gas budget of a maize/soybean rotation ecosystem. Agriculture, Ecosystems and Environment 134, 234-242.

Beare, M.H., Hendrix, P.F., Cabrera, M.L., Coleman, D.C., 2010. Aggregate-Protected and Unprotected Organic Matter Pools in Conventional- and No-Tillage Soils. Soil Science Society America Journal. 58, 787-795.

Bertrand, M., Barot, S., Blouin, M., Whalen, J., de Oliveira, T., Roger-Estrade, J., 2015. Earthworm services for cropping systems. A review. Agronomy for Sustainable Development 35, 553-567.

Bhogal, A., Nicholson, F.A., Rollett, A., Taylor, M., Litterick, A., Whittingham, M.J., Williams, J.R., 2018. Improvements in the Quality of Agricultural Soils Following Organic Material Additions Depend on Both the Quantity and Quality of the Materials Applied. Frontiers in Sustainable Food Systems 2, 9 (1-13).

Blanco-Canqui, H., 2011. Does no-till farming induce water repellency to soils? Soil Use and Management 27, 2-9.

Blanco-Canqui, H., Lal, R., Owens, L.B., Post, W.M., Izaurralde, R.C., 2005.

Strength properties and organic carbon of soils in the North Appalachian region. Soil Science Society of America Journal 69, 663.

Blaud, A., Menon, M., van der Zaan, B., Lair, G.J., Banwart, S.A., 2017. Effects of Dry and Wet Sieving of Soil on Identification and Interpretation of Microbial Community Composition, in: Advances in Agronomy 142, 119-142.

Blouin, M., Hodson, M.E., Delgado, E.A., Baker, G., Brussaard, L., Butt, K.R., Dai, J., Dendooven, L., Peres, G., Tondoh, J.E., Cluzeau, D., Brun, J.J., 2013. A review of earthworm impact on soil function and ecosystem services. European Journal Soil Science. 64, 161-182.

Boddey, R.M., Jantalia, C.P., Conceiçao, P.C., Zanatta, J.A., Bayer, C., Mielniczuk, J., Dieckow, J., Dos Santos, H.P., Denardin, J.E., Aita, C., Giacomini, S.J., Alves, B.J.R., Urquiaga, S., 2010. Carbon accumulation at depth in Ferralsols under zero-till subtropical agriculture. Global Change Biology 16, 784-795.

Bolliger, A., Magid, J., Amado, J.C.T., Skóra Neto, F., Ribeiro, M. de F. dos S., Calegari, A., Ralisch, R., de Neergaard, A., 2006. Taking Stock of the Brazilian "Zero-Till Revolution": A Review of Landmark Research and Farmers' Practice Advances Agronomy. 91, 47-64.

Bongiorno, G., Bünemann, E.K., Oguejiofor, C.U., Meier, J., Gort, G., Comans, R., Mäder, P., Brussaard, L., de Goede, R., 2019. Sensitivity of labile carbon fractions to tillage and organic matter management and their potential as comprehensive soil quality indicators across pedoclimatic conditions in Europe Ecology Indicators. 99, 38-50.

Born, M., Dorr, H., Levin, I., 1990. Methane consumption in aerated soils of the

temperate zone. Tellus 42B, 2-8.

Briones, M.J.I., Schmidt, O., 2017. Conventional tillage decreases the abundance and biomass of earthworms and alters their community structure in a global meta-analysis. Global Change Biology 23, 4396-4419.

Bronick, C.J., Lal, R., 2011. A review on organic matter in soil aggregates. Acta Pedologica Sinica 48, 412–418.

Bronick, C.J., Lal, R., 2005. Soil structure and management: A review. Geoderma 124, 3-22.

Brookes, P.C., Landman, A., Pruden, G., Jenkinson, D.S., 1985. Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil. Soil Biology and Biochemistry 17, 837–842.

Brown, G., Benito, N., Pasini, A., Sautter, K., Defguimaraes, M., Torres, E., 2003. No-tillage greatly increases earthworm populations in Paraná state, BrazilThe 7th international symposium on earthworm ecology · Cardiff · Wales · 2002. Pedobiologia (Jena).

Brunel, N., Seguel, O., Acevedo, E., 2013. Conservation tillage and water availability for wheat in the dryland of central Chile. Journal of Soil Science and Plany Nutrition 13, 622-637.

Bünemann, E.K., Bongiorno, G., Bai, Z., Creamer, R.E., De Deyn, G., de Goede, R., Fleskens, L., Geissen, V., Kuyper, T.W., Mäder, P., Pulleman, M., Sukkel, W., van Groenigen, J.W., Brussaard, L., 2018. Soil quality – A critical review. Soil Biology and Biochemistry 120, 105-125.

Burr-Hersey, J.E., Mooney, S.J., Bengough, A.G., Mairhofer, S., Ritz, K., 2017. Developmental morphology of cover crop species exhibit contrasting

behaviour to changes in soil bulk density, revealed by X-ray computed tomography. PLoS ONE 12, 1-18.

Busari, M. a., Kukal, S.S., Kaur, A., Bhatt, R., Dulazi, a. a., 2015. Conservation tillage impacts on soil, crop and the environment. International Soil and Water Conservation Research 3, 1–11.

Butterbach-Bahl, K., Baggs, E.M., Dannenmann, M., Kiese, R., Zechmeister-Boltenstern, S., 2013. Nitrous oxide emissions from soils: How well do we understand the processes and their controls? Philosophical Transactions of the Royal Society B: Biological Sciences 368, 1-13.

Capowiez, Y., Cadoux, S., Bouchant, P., Ruy, S., Roger-Estrade, J., Richard, G., Boizard, H., 2009. The effect of tillage type and cropping system on earthworm communities, macroporosity and water infiltration. Soil and Tillage Research 105, 209-216.

Carter, M.R., Holmstrom, D., Sanderson, J.B., Ivany, J., DeHaan, R., 2011. Comparison of conservation with conventional tillage for potato production in Atlantic Canada: crop productivity, soil physical properties and weed control. Canadian Journal of Soil Science 85, 453-461.

Cassel, D.., Whitbeck, J.., 1994. Tomography of Soil-Water-Root Processes. Soil Science Society of Agronomy, Madison.

Castro, G.S.A., Crusciol, C.A.C., Calonego, J.C., Rosolem, C.A., 2015. Management impacts on soil organic matter of tropical soils. Vadose Zo. J.14, 1-8.

Celik, I., Barut, Z.B., Ortas, I., Gok, M., Demirbas, A., Tulun, Y., Akpinar, C., 2011. Impacts of different tillage practices on some soil microbiological properties and crop yield under semi-arid Mediterranean conditions.

International Journal of Plant Production 5, 237-254.

Chatskikh, D., Olesen, J.E., 2007. Soil tillage enhanced CO_2 and N_2O emissions from loamy sand soil under spring barley. Soil and Tillage Research 97, 5-18.

Cheng-Fang, L., Dan-Na, Z., Zhi-Kui, K., Zhi-Sheng, Z., Jin-Ping, W., Ming-Li, C., Cou-Gui, C., 2012. Effects of tillage and nitrogen fertilizers on CH_4 and CO_2 emissions and soil organic carbon in paddy fields of central China. PLoS ONE 7, 1-9.

Coelho, M.B., Mateos, L., Villalobos, F.J., 2000. Influence of a compacted loam subsoil layer on growth and yield of irrigated cotton in Southern Spain. Soil and Tillage Research 57, 129-142.

Coleman, D.C., Crossley, D.A., Hendrix, P.F., 2007. Secondary Production: Activities of Heterotrophic Organisms—The Soil Fauna, in: Fundamentals of Soil Ecology. Elsevier, United Kingdom.

Cooper, H. V., Vane, C.H., Evers, S., Aplin, P., Girkin, N.T., Sjogersten, S., 2019. From peat swamp forest to oil palm plantations: The stability of tropical peatland carbon. Geoderma 342, 09-117.

Copec, K., Filipovic, D., Husnjak, S., Kovacev, I., Kosutic, S., 2015. Effects of tillage systems on soil water content and yield in maize and winter wheat production. Plant, Soil Environment. 61, 213–219.

Creech, E., 2017. Saving Money, Time and Soil: The Economics of No-Till Farming [WWW Document]. Natural Resources Conservation Service in Conservation. URL https://www.usda.gov/media/blog/2017/11/30/saving-money-time-and-soil-economics-no-till-farming (accessed 10.9.19).

Crestana, S., Mascarenhas, S., Pozzi-Mucelli, R.S., 1985. Static and dynamic

three-dimensional studies of water in soil using computed tomographic scanning. Soil Science 140, 1-9.

Czarnes, S., Hallett, P.D., Bengough, A.G., Young, I.M., 2000. Root- and microbial-derived mucilages affect soil structure and water transport. European Journal of Soil Science 51, 435-443.

Da Veiga, M., Reinert, D.J., Reichert, J.M., Kaiser, D.R., 2008. Short and long-term effects of tillage systems and nutrient sources on soil physical properties of a Southern Brazilian Hapludox. Revista Brasileira Ciencia do Solo 32, 1-10.

de Araújo Santos, G.A., Moitinho, M.R., de Oliveira Silva, B., Xavier, C.V., Teixeira, D.D.B., Corá, J.E., Júnior, N.L.S., 2019. Effects of long-term no-tillage systems with different succession cropping strategies on the variation of soil CO_2 emission. Science Total Environment. 686, 413-424.

de Freitas, P.L., Landers, J.N., 2014. The Transformation of Agriculture in Brazil Through Development and Adoption of Zero Tillage Conservation Agriculture. International Soil Water Conservation Research. 2, 35-46.

Davidson, E.A., Janssens, I.A., 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. Nature 440, 165-173.

Dekker, L.W., Ritsema, C.J., 2000. Wetting patterns and moisture variability in water repellent Dutch soils. Journal of Hydrology 231-232, 148-164.

DeLaune, P.B., Sij, J.W., Krutz, L.J., 2013. Impact of soil aeration on runoff characteristics in dual-purpose no-till wheat systems. Journal Soil Water Conservation. 68, 315–324.

Department for Environment Food and Rural Affairs, Department of Agriculture and Rural Development (Northern Ireland), Welsh Assembly Government:

The Department for Rural Affairs and Heritage, The Scottish Government: Rural and Environment Research and Analysis Directorate, 2015. Agriculture in the United Kingdom, 2015, Agriculture in the United Kingdom.

Derpsch, R., 2008. No-tillage and conservation agriculture: a progress report, World Association of Soil and Water Conservation, Bangkok, 7-42.

Derpsch, R., Friedrich, T., Kassam, A., Hongwen, L., 2010. Current status of adoption of no-till farming in the world and some of its main benefits. International Journal of Agricultural and Biological Engineering 3, 1–25.

Devine, S., Markewitz, D., Hendrix, P., Coleman, D., 2014. Soil aggregates and associated organic matter under conventional tillage, no-tillage, and forest succession after three decades. PloS One 9, 1-12.

Dexter, A.R., 1988. Advances in characterization of soil structure. Soil and Tillage Research 11, 199-238.

Díaz-Zorita, M., Grove, J.H., Murdock, L., Herbeck, J., Perfect, E., 2004. Soil structural disturbance effects on crop yields and soil properties in a no-till production system. Agronomy Journal 96, 1651–1659.

Dignac, M.F., Derrien, D., Barré, P., Barot, S., Cécillon, L., Chenu, C., Chevallier, T., Freschet, G.T., Garnier, P., Guenet, B., Hedde, M., Klumpp, K., Lashermes, G., Maron, P.A., Nunan, N., Roumet, C., Basile-Doelsch, I., 2017. Increasing soil carbon storage: mechanisms, effects of agricultural practices and proxies. A review. Agronomy Sustainable Development 23, 193-223.

Ding, X., Zhang, B., Zhang, Xudong, Yang, X., Zhang, Xiaoping, 2011. Effects of tillage and crop rotation on soil microbial residues in a rainfed

agroecosystem of northeast China. Soil and Tillage Research 114, 43-49.

Dinnes, D.L., Karlen, D.L., Jaynes, D.B., Kaspar, T.C., Hatfield, J.L., Colvin, T.S., Cambardella, C.A., 2002. Nitrogen management strategies to reduce nitrate leaching in tile-drained midwestern soils. Agronomy Journal 94, 153-171.

Disnar, J.R., Guillet, B., Keravis, D., Di-Giovanni, C., Sebag, D., 2003. Soil organic matter (SOM) characterization by Rock-Eval pyrolysis: Scope and limitations. Organic Geochemistry. 34, 327–343.

Dominati, E., Patterson, M., Mackay, A., 2010. A framework for classifying and quantifying the natural capital and ecosystem services of soils. Ecological Economics 69, 1858-1868.

Doran, J.W., Jones, A.J., Dick, R.P., Breakwell, D.P., Turco, R.F., 1996. Soil enzyme activities and biodiversity measurements as integrative microbiological indicators. Methods for Assessing Soil Quality, SSSA Special Publication 49, 247-271.

Dorel, M., Lakhia, S., Pététin, C., Bouamer, S., Risède, J.M., 2010. No-till banana planting on crop residue mulch: Effect on soil quality and crop functioning. Fruits 65, 55-68.

Dörner, J., Sandoval, P., Dec, D., 2010. The role of soil structure on the pore functionality of an ultisol. Journal of Soil Science and Plant Nutrition 10, 495-508.

Drakopoulos, D., Scholberg, J.M.S., Lantinga, E.A., Tittonell, P.A., 2016. Influence of reduced tillage and fertilization regime on crop performance and nitrogen utilization of organic potato. Organic Agriculture 6, 75-87.

Drury, C.F., Reynolds, W.D., Tan, C.S., Welacky, T.W., Calder, W.,

McLaughlin, N.B., 2006. Emissions of Nitrous Oxide and Carbon Dioxide: Influence of Tillage Type and Nitrogen Placement Depth. Soil Science Society of America Journal 70, 570-581.

ECAF, Conservation agriculture federation, 2014. Uptake of conservation agriculture in Europe. http://www.ecaf.org/ca-in-europe/uptake-of-ca-in-europe (Date accessed: 9/4/18).

Ellert, B.H., Bettany, J.R., 1995. Calculation of organic matter and nutrients stored in soils under contrasting management regimes. Canadian Journal of Soil Science 75, 529–538.

EPA, 2014. Greenhouse gas emissions. Global Greenhouse Gas Emissions Data.

Eurostat, 2014. Agriculture Survey on Agricultural Production Methods. https://ec.europa.eu/eurostat/statistics-explained/index.php/Agrienvironmental_indicator_-_tillage_practices (Date accessed 9/4/18).

Eurostat, 2013a. Glossary: conservational tillage. https://ec.europa.eu/eurostat/statistics-explained/index.php?title=Glossary:Conservational_tillage (Date accessed 9/4/18).

Eurostat, 2013b. Glossary: zero tillage. https://ec.europa.eu/eurostat/statistics-explained/index.php?title=Glossary:Zero_tillage (Date accessed 9/4/18).

Fabrizzi, K.P., García, F.O., Costa, J.L., Picone, L.I., 2005. Soil water dynamics, physical properties and corn and wheat responses to minimum and no-tillage systems in the southern Pampas of Argentina. Soil and Tillage Research 81, 57-69.

FAO, 2008. An international technical workshop Investing in sustainable crop

intensification: The Case for Improving Soil Health 6, Rome.

FAO, 2019. Top wheat producing countries. http://www.fao.org/home/en (Date accessed 6/27/19).

FAO, 2014. Conservation agriculture. http://www.fao.org/ag/ca/6c.html (Date accessed 9/4/18).

FAO, 2003. Suitalbe methods of tillage for the farm. http://www.fao.org/3/y5146e/y5146e08.htm (Date accessed 7/1/19).

Farooq, M., Flower, K.C., Jabran, K., Wahid, A., Siddique, K.H.M., 2011. Crop yield and weed management in rainfed conservation agriculture. Soil and Tillage Research 117, 172-183.

Feng, W., Plante, A.F., Aufdenkampe, A.K., Six, J., 2014. Soil organic matter stability in organo-mineral complexes as a function of increasing C loading. Soil Biology Biochemistry. 69, 398-405.

Feng, Y., Balkcom, K.S., 2017. Nutrient Cycling and Soil Biology in Row Crop Systems Under Intensive Tillage, in: Soil Health and Intensification of Agroecosystems. Elsevier, Oxford, United Kingdom.

Fernández, R., Quiroga, A., Zorati, C., Noellemeyer, E., 2010. Carbon contents and respiration rates of aggregate size fractions under no-till and conventional tillage. Soil and Tillage Research 109, 103-109.

Ferreras, L.A., Costa, J.L., Garcia, F.O., Pecorari, C., 2000. Effect of no-tillage on some soil physical properties of a structural degraded Petrocalcic Paleudoll of the southern “Pampa” of Argentina. Soil and Tillage Research 54, 31-39.

Finch, H.J.S., Samuel, A.M., Lane, G.P.F., 2014. Lockhart & Wiseman’s Crop Husbandry Including Grassland: Ninth Edition, Lockhart & Wiseman’s

Crop Husbandry Including Grassland: Ninth Edition. Woodhead Publishing, Cambridge, United Kingdom.

Fischer, C., Roscher, C., Jensen, B., Eisenhauer, N., Baade, J., Attinger, S., Scheu, S., Weisser, W.W., Schumacher, J., Hildebrandt, A., 2014. How do earthworms, soil texture and plant composition affect infiltration along an experimental plant diversity gradient in grassland? PLoS ONE 9, 1-12.

Franzluebbers, A.J., Hons, F.M., 1996. Soil-profile distribution of primary and secondary plant-available nutrients under conventional and no tillage. Soil and Tillage Research 39, 229-239.

Galdos, M.V., Pires, L.F., Cooper, H.V., Calonego, J.C., Rosolem, C.A., Mooney, S.J., 2019. Assessing the long-term effects of zero-tillage on the macroporosity of Brazilian soils using X-ray Computed Tomography. Geoderma 337, 1126-1135.

Gantzer, C.J., Anderson, S.H., 2002. Computed tomographic measurement of macroporosity in chisel-disk and no-tillage seedbeds. Soil and Tillage Research 64, 101-111.

Gardner, B.R., Nielsen, D.C., Shock, C.C., 1992. Infrared Thermometry and the Crop Water Stress Index. I. History, Theory, and Baselines. Journal of Production Agriculture 5, 462-466.

Gee, G.W., Or, D., 2002. Methods of Soil Analysis: Part 4 Physical Methods. Methods of Soil Analysis. Soil Science Society of America, Madison, United States of America.

Giller, K.E., Beare, M.H., Lavelle, P., Izac, A.M.N., Swift, M.J., 1997. Agricultural intensification, soil biodiversity and agroecosystem function. Appl. Soil Ecology. 6, 3-16.

Giller, K.E., Witter, E., Corbeels, M., Tittonell, P., 2009. Conservation agriculture and smallholder farming in Africa: The heretics' view. Field Crops Research 114, 23-34.

Girkin, N.T., Vane, C.H., Cooper, H.V., Moss-Hayes, V., Craigon, J., Turner, B.L., Ostle, N., Sjögersten, S., 2019. Spatial variability of organic matter properties determines methane fluxes in a tropical forested peatland. Biogeochemistry 142, 231-245.

Godwin, R.J., 2014. Potential of "No-till" Systems for Arable Farming. The Worshipful Company of Farmers. London.

Govaerts, B., Verhulst, N., Castellanos-Navarrete, A., Sayre, K.D., Dixon, J., Dendooven, L., 2009. Conservation agriculture and soil carbon sequestration: Between myth and farmer reality. Critical Reviews in Plant Sciences 3, 97-122.

Graves, A.R., Morris, J., Deeks, L.K., Rickson, R.J., Kibblewhite, M.G., Harris, J.A., Farewell, T.S., Truckle, I., 2015. The total costs of soil degradation in England and Wales. Ecology Economics. 119, 399-413.

Green, V.S., Stott, D.E., Cruz, J.C., Curi, N., 2007. Tillage impacts on soil biological activity and aggregation in a Brazilian Cerrado Oxisol. Soil and Tillage Research 92, 114–121.

Hainsworth, J.M., Aylmore, L.A.G., 1983. The use of computer-assisted tomography to determine spatial distribution of soil water content. Australian Journal of Soil Research 21, 435-443.

Hallett, P.D., Baumgartl, T., Young, I.M., 2001. Subcritical water repellency of aggregates from a range of soil management practices. Soil Science Society of America Journal 65, 184-190.

Hamza, M.A., Anderson, W.K., 2005. Soil compaction in cropping systems: A review of the nature, causes and possible solutions. Soil and Tillage Research 82, 121-145.

Hangen, E., Buczko, U., Bens, O., Brunotte, J., Hüttl, R.F., 2002. Infiltration patterns into two soils under conventional and conservation tillage: Influence of the spatial distribution of plant root structures and soil animal activity. Soil Tillage Res 63, 181-186.

Harris, G.., Clements, R.., Rose, S.., Parkin, A., Shepherd, M., 2004. Review of impacts of rural land use and management on flood generation. DEFRA.

He, J., Wang, Q., Li, H., Tullberg, J.N., McHugh, A.D., Bai, Y., Zhang, X., McLaughlin, N., Gao, H., 2009. Soil physical properties and infiltration after long-term no-tillage and ploughing on the Chinese Loess Plateau. New Zealand Journal Crop Horticulture. Sci 37, 157-166.

Hermle, S., Anken, T., Leifeld, J., Weisskopf, P., 2008. The effect of the tillage system on soil organic carbon content under moist, cold-temperate conditions. Soil and Tillage Research 98, 94–105.

Hodgson, J.., 1997. Soil Survey field Handbook. Soil Survey Technical Monograph No. 5. Silsoe.

Hou, X., Li, R., 2018. Potato tuber yields in semi-arid environments are increased by tillage and mulching practices. Agronomy Journal 110, 2641-2651.

Huang, Y., Ren, W., Wang, L., Hui, D., Grove, J.H., Yang, X., Tao, B., Goff, B., 2018. Greenhouse gas emissions and crop yield in no-tillage systems: A meta-analysis. Agriculture, Ecosystems and Environment 268, 144-153.

Hütsch, B.W., 2001. Methane oxidation in non-flooded soils as affected by crop

production - Invited paper. European Journal of Agronomy 14, 237-260.

IPCC, 2007. Working Group I Report “The Physical Science Basis.” PCC Fourth Assessment Report. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

IPCC, 2014: Climate Change 2014: Synthesis Report. Contribution of Working Groups I, II, III to the Fifth Assessment Intergovernmental Panel on Climate Change. IPCC, Geneva, Switzerland.

Islam, K.R., Weil, R.R., 2000. Soil quality indicator properties in mid-Atlantic soils as influenced by conservation management. Journal of Soil and Water Conservation 55, 69-78.

Islam, R., Reeder, R., 2014. No-till and conservation agriculture in the United States: An example from the David Brandt farm, Carroll, Ohio. International Soil and Water Conservation Research 2, 97-107.

Jantalia, C.P., Dos Santos, H.P., Urquiaga, S., Boddey, R.M., Alves, B.J.R., 2008. Fluxes of nitrous oxide from soil under different crop rotations and tillage systems in the South of Brazil. Nutrient Cycling Agroecosystems. 82, 161-173.

Jenkinson, D.S., Brookes, P.C., Powlson, D.S., 2004. Measuring soil microbial biomass. Soil Biology and Biochemistry 36, 5-7.

Jordan, D., Stecker, J.A., Cacnio-Hubbard, V.N., Li, F., Gantzer, C.J., Brown, J.R., 1997. Earthworm activity in no-tillage and conventional tillage systems in Missouri soils: A preliminary study. Soil Biology and Biochemistry 29, 489-491.

Kainiemi, V., Arvidsson, J., Kätterer, T., 2013. Short-term organic matter mineralisation following different types of tillage on a Swedish clay soil.

Biology and Fertility of Soils 49, 495-504.

Kandeler, E., Tscherko, D., Spiegel, H., 1999. Long-term monitoring of microbial biomass, N mineralisation and enzyme activities of a chernozem under different tillage management. Biology and Fertility of Soils 28, 343-351.

Karlen, D.L., Mausbach, M.J., Doran, J.W., Cline, R.G., Harris, R.F., Schuman, G.E., 1997. Soil quality: A concept, definition, and framework for evaluation: (A guest editorial). Soil Science Society of America Journal 61, 4-10.

Kassam, A., Friedrich, T., Derpsch, R., Kienzle, J., 2015. Overview of the Worldwide Spread of Conservation Agriculture. Field Actions Science Reports 8, 104-129.

Kassam, A., Friedrich, T., Shaxson, F., Pretty, J., 2009. The spread of conservation agriculture: Justification, sustainability and uptake. International Journal of Agricultural Sustainability 6, 1-13.

Keil, A., D'souza, A., McDonald, A., 2015. Zero-tillage as a pathway for sustainable wheat intensification in the Eastern Indo-Gangetic Plains: does it work in farmers' fields? Food Security 7, 983-1001.

Kladivko, E.J., 2001. Tillage systems and soil ecology. Soil and Tillage Research 61, 61-76.

Knapp, S., van der Heijden, M.G.A., 2018. A global meta-analysis of yield stability in organic and conservation agriculture. Nature Communications 9, 1-9.

Kou, T.J., Zhu, P., Huang, S., Peng, X.X., Song, Z.W., Deng, A.X., Gao, H.J., Peng, C., Zhang, W.J., 2012. Effects of long-term cropping regimes on soil

carbon sequestration and aggregate composition in rainfed farmland of Northeast China. Soil Tillage Research 118, 132-138.

Kumar, R., Rawat, K.S., Jitendra, S., Ashutosh, S., Ashish, R., 2013. Soil aggregation dynamics and carbon sequestration. Journal Applied Natural Science 5, 250–267.

Kutílek, M., 2004. Soil hydraulic properties as related to soil structure. Soil and Tillage Research 79, 175-184.

La Scala, N., Bolonhezi, D., Pereira, G.T., 2006. Short-term soil CO_2 emission after conventional and reduced tillage of a no-till sugar cane area in southern Brazil. Handbook of Environmental Chemistry, Volume 5: Water Pollution 91, 244–248.

Lal, R., Mahboubi, a. a., Fausey, N.R., 1994. Long-Term Tillage and Rotation Effects on Properties of a Central Ohio Soil. Soil Science Society America Journal 58, 517.

Lampurlanés, J., Plaza-Bonilla, D., Álvaro-Fuentes, J., Cantero-Martínez, C., 2016. Long-term analysis of soil water conservation and crop yield under different tillage systems in Mediterranean rainfed conditions. Field Crops Research 189, 59-67.

Larson, W.E., Pierce, F.J., 1994. The dynamics of soil quality as a measure of sustainable management. Defining Soil Quality for a Sustainable Environment. SSSA Special Publication 35, Soil Science Society of America and American Society of Agronomy, 37-51.

Laugier, A., Garai, J., 2007. Derivation of the Ideal Gas Law. Journal of Chemical Education 84, pp 1832.

Laugier, A., Garai, J., 2009. Derivation of the Ideal Gas Law. Journal Chemistry

Educucation 84, pp1832.

Laukkanen, M., Nauges, C., 2011. Environmental and production cost impacts of no-till in Finland: Estimates from observed behavior. Land Economics 87, 508-527.

Li, D., Liu, M., Cheng, Y., Wang, D., Qin, J., Jiao, J., Li, H., Hu, F., 2011. Methane emissions from double-rice cropping system under conventional and no tillage in southeast China. Soil and Tillage Research 113, 77-81.

Liu, X.J., Mosier, A.R., Halvorson, A.D., Zhang, F.S., 2006. The impact of nitrogen placement and tillage on NO, N_2O, CH_4 and CO_2 fluxes from a clay loam soil. Plant Soil. 280, 177-188.

Liu, X., Herbert, S.J., Hashemi, A.M., Zhang, X., Ding, G., 2006. Effects of agricultural management on soil organic matter and carbon transformation - A review. Plant, Soil Environment 52, 531-543.

Lugato, E., Morari, F., Nardi, S., Berti, A., Giardini, L., 2009. Relationship between aggregate pore size distribution and organic-humic carbon in contrasting soils. Soil and Tillage Research 103, 153-157.

Liu, Y., Gao, M., Wu, W., Tanveer, S.K., Wen, X., Liao, Y., 2013. The effects of conservation tillage practices on the soil water-holding capacity of a non-irrigated apple orchard in the Loess Plateau, China. Soil Tillage Research 130, 7-12.

Luo, Z., Wang, E., Sun, O.J., 2010. Can no-tillage stimulate carbon sequestration in agricultural soils? A meta-analysis of paired experiments. Agriculture, Ecosystems and Environment 139, 224–231.

Luo, Z., Wang, E., Sun, O.J., 2010. Soil carbon change and its responses to agricultural practices in Australian agro-ecosystems: A review and

synthesis. Geoderma 155, 211-223.

Lupwayi, N.Z., Clayton, G.W., O'Donovan, J.T., Harker, K.N., Turkington, T.K., Soon, Y.K., 2007. Phosphorus release during decomposition of crop residues under conventional and zero tillage. Soil and Tillage Research 95, 231-239.

Małecka, I., Blecharczyk, A., Sawinska, Z., Dobrzeniecki, T., 2012. The effect of various long-term tillage systems on soil properties and spring barley yield. Turkish Journal of Agriculture and Forestry 36, 217-226.

Man, L., Guilin, H., Qian, Z., 2019. Effects of Soil Aggregate Stability on Soil Organic Carbon and Nitrogen under Land Use Change in an Erodible Region in Southwest China. International Journal of Environmental Research and Public Health 16, 1–14.

Mangalassery, S., Mooney, S.J., Sparkes, D.L., Fraser, W.T., Sjögersten, S., 2015. Impacts of zero tillage on soil enzyme activities, microbial characteristics and organic matter functional chemistry in temperate soils. European Journal of Soil Biology 68, 9-17.

Mangalassery, S., Sjögersten, S., Sparkes, D.L., Mooney, S.J., 2015. Examining the potential for climate change mitigation from zero tillage. Journal of Agricultural Science 153, 1151-1173.

Mangalassery, S., Sjögersten, S., Sparkes, D.L., Sturrock, C.J., Craigon, J., Mooney, S.J., 2015. To what extent can zero tillage lead to a reduction in greenhouse gas emissions from temperate soils? Scientific Reports 4, 4586.

Martínez, E., Fuentes, J.P., Silva, P., Valle, S., Acevedo, E., 2008. Soil physical properties and wheat root growth as affected by no-tillage and conventional tillage systems in a Mediterranean environment of Chile. Soil Tillage

Research 99, 232-244.

Mathew, R.P., Feng, Y., Githinji, L., Ankumah, R., Balkcom, K.S., 2012. Impact of No-tillage and conventional tillage systems on soil microbial communities. Applied and Environmental Soil Science 2012, 1-10.

Matula, S., 2003. The influence of tillage treatments on water infiltration into soil profile. Plant, Soil and Environment 49, 298-306.

McVay, K.A., Budde, J.A., Fabrizzi, K., Mikha, M.M., Rice, C.W., Schlegel, A.J., Peterson, D.E., Sweeney, D.W., Thompson, C., 2006. Management effects on soil physical properties in long-term tillage studies in Kansas. Soil Science Society of America 70, 434-438.

Met Office, 2019. Climate summaries: UK and regional series. URL http://www.metoffice.gov.uk/research/climate/maps-and-data/summaries/index (accessed 25.11.19).

Meyer, N., Welp, G., Amelung, W., 2018. The Temperature Sensitivity (Q10) of Soil Respiration: Controlling Factors and Spatial Prediction at Regional Scale Based on Environmental Soil Classes. Global Biogeochemical Cycles 32, 302-323.

Minasny, B., Malone, B.P., McBratney, A.B., Angers, D.A., Arrouays, D., Chambers, A., Chaplot, V., Chen, Z.S., Cheng, K., Das, B.S., Field, D.J., Gimona, A., Hedley, C.B., Hong, S.Y., Mandal, B., Marchant, B.P., Martin, M., McConkey, B.G., Mulder, V.L., O'Rourke, S., Richer-de-Forges, A.C., Odeh, I., Padarian, J., Paustian, K., Pan, G., Poggio, L., Savin, I., Stolbovoy, V., Stockmann, U., Sulaeman, Y., Tsui, C.C., Vågen, T.G., van Wesemael, B., Winowiecki, L., 2017. Soil carbon 4 per mille. Geoderma 292, 59-86.

Miura, F., Nakamoto, T., Kaneda, S., Okano, S., Nakajima, M., Murakami, T., 2008. Dynamics of soil biota at different depths under two contrasting tillage practices. Soil Biology and Biochemistry 40, 406-414.

Montgomery, D.R., 2007. Soil erosion and agricultural sustainability. Proceedings of the National Academy of Sciences of the United States of America 104, 13268–72.

Mori, Y., Fujihara, A., Yamagishi, K., 2014. Installing artificial macropores in degraded soils to enhance vertical infiltration and increase soil carbon content. Progress in Earth and Planetary Science 1:30.

Munkholm, L.J., Hansen, E.M., Olesen, J.E., 2008. The effect of tillage intensity on soil structure and winter wheat root/shoot growth. Soil Use Management 24, 392-400.

Myhre, G., Shindell, D., Bréon, F.-M., Collins, W., Fuglestvedt, J., Huang, J., Koch, D., Lamarque, J.-F., Lee, D., Mendoza, B., Nakajima, T., Robock, A., Stephens, G., Takemura, T., Zhang, H., 2013. Anthropogenic and Natural Radiative Forcing. Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change 659–740.

Newell, A.J., Vane, C.H., Sorensen, J.P.R., Moss-Hayes, V., Gooddy, D.C., 2016. Long-term Holocene groundwater fluctuations in a chalk catchment: evidence from Rock-Eval pyrolysis of riparian peats. Hydrological Processes 30, 4556-4567.

Nimmo, J.R., 2005. Porosity and pore size distribution, in: Encyclopedia of Soils in the Environment. London, Elsevier.

Nogueirol, R.C., Cerri, C.E.P., Silva, W.T.L. da, Alleoni, L.R.F., 2014. Effect

of no-tillage and amendments on carbon lability in tropical soils. Soil Tillage Research 143, 67-76.

OECD, 2001. Environmental Indicators for Agriculture. Methods and Results. Policy. Agriculture Directorate, OECD, Paris, France.

Ogle, S.M., Breidt, F.J., Paustian, K., 2005. Agricultural management impacts on soil organic carbon storage under moist and dry climatic conditions of temperate and tropical regions. Biogeochemistry 72, 87-121.

Ogle, S.M., Swan, A., Paustian, K., 2012. No-till management impacts on crop productivity, carbon input and soil carbon sequestration. Agriculture, Ecosystems and Environment 149, 37-49.

Oorts, K., Merckx, R., Gréhan, E., Labreuche, J., Nicolardot, B., 2007. Determinants of annual fluxes of CO_2 and N_2O in long-term no-tillage and conventional tillage systems in northern France. Soil and Tillage Research 95, 133-148.

Osunbitan, J.A., Oyedele, D.J., Adekalu, K.O., 2005. Tillage effects on bulk density, hydraulic conductivity and strength of a loamy sand soil in southwestern Nigeria. Soil Tillage Research 82, 57-64.

Pagliai, M., La Marca, M., Lucamante, G., Genovese, L., 1984. Effects of zero and conventional tillage on the length and irregularity of elongated pores in a clay loam soil under viticulture. Soil and Tillage Research 4, 433-444.

Pagliai, M., Vignozzi, N., Pellegrini, S., 2004. Soil structure and the effect of management practices, in: Soil and Tillage Research. pp. 131–143.

Palm, C., Blanco-Canqui, H., DeClerck, F., Gatere, L., Grace, P., 2014. Conservation agriculture and ecosystem services: An overview. Agriculture, Ecosystems and Environment 187, 87-105.

Paustian, K., Collins, H.P., Paul, E.A., 1997. Management Controls on Soil Organic Carbon, in: Soil Organic Matter in Temperate Agroecosystems.CRC Press, Boca Raton, FL.

Peigné, J., Ball, B.C., Roger-Estrade, J., David, C., 2007. Is conservation tillage suitable for organic farming? A review. Soil Use and Management 23, 129-144.

Peigné, J., Cannavaciuolo, M., Gautronneau, Y., Aveline, A., Giteau, J.L., Cluzeau, D., 2009. Earthworm populations under different tillage systems in organic farming. Soil and Tillage Research 104, 207-214.

Pelosi, C., Bertrand, M., Roger-Estrade, J., 2009. Earthworm community in conventional, organic and direct seeding with living mulch cropping systems. Agronomy for Sustainable Development 2, 287-295.

Peng, X., Horn, R., Hallett, P., 2015. Soil structure and its functions in ecosystems: Phase matter & scale matter. Soil and Tillage Research 146, 1-124.

Petrovic, A.M., Siebert, J.E., Rieke, P.E., 1982. Soil bulk density analysis in three dimensions by computed tomographic scanning. Soil Science Society of America Journal 46, 445-450.

Pierret, A., Latchackak, K., Chathanvongsa, P., Sengtaheuanghoung, O., Valentin, C., 2007. Interactions between root growth, slope and soil detachment depending on land use: A case study in a small mountain catchment of Northern Laos. Plant and Soil 301, 51-64.

Pimentel, D., Burgess, M., 2013. Soil Erosion Threatens Food Production. Agriculture 3, 443-463.

Piron, D., Boizard, H., Heddadj, D., Pérès, G., Hallaire, V., Cluzeau, D., 2017.

Indicators of earthworm bioturbation to improve visual assessment of soil structure. Soil Tillage Research 173, 53-63.

Pittelkow, C.M., Linquist, B.A., Lundy, M.E., Liang, X., van Groenigen, K.J., Lee, J., van Gestel, N., Six, J., Venterea, R.T., van Kessel, C., 2015. When does no-till yield more? A global meta-analysis. Field Crops Research 183, 156–168.

Piva, J.T., Dieckow, J., Bayer, C., Zanatta, J.A., de Moraes, A., Pauletti, V., Tomazi, M., Pergher, M., 2012. No-till reduces global warming potential in a subtropical Ferralsol. Plant and Soil 361, 359-373.

Powlson, D.S., Stirling, C.M., Jat, M.L., Gerard, B.G., Palm, C. a., Sanchez, P. a., Cassman, K.G., 2014. Limited potential of no-till agriculture for climate change mitigation. Nature Climate Change 4, 678–683.

Powlson, D.S., Stirling, C.M., Thierfelder, C., White, R.P., Jat, M.L., 2016. Does conservation agriculture deliver climate change mitigation through soil carbon sequestration in tropical agro-ecosystems? Agriculture, Ecosystems and Environment 220, 164–174.

Qin, Z., Dunn, J.B., Kwon, H., Mueller, S., Wander, M.M., 2016. Soil carbon sequestration and land use change associated with biofuel production: Empirical evidence. GCB Bioenergy 8, 66-80.

Ramesh, T., Bolan, N.S., Kirkham, M.B., Wijesekara, H., Kanchikerimath, M., Srinivasa Rao, C., Sandeep, S., Rinklebe, J., Ok, Y.S., Choudhury, B.U., Wang, H., Tang, C., Wang, X., Song, Z., Freeman, O.W., 2019. Soil organic carbon dynamics: Impact of land use changes and management practices: A review. Advances in Agronomy. Academic Press Inc.

Reicosky, D.C., Archer, D.W., 2007. Moldboard plow tillage depth and short-

term carbon dioxide release. Soil and Tillage Research 94, 109-121.

Reth, S., Reichstein, M., Falge, E., 2005. The effect of soil water content, soil temperature, soil pH-value and the root mass on soil CO_2 efflux - A modified model. Plant and Soil 268, 21-33.

Rigon, J.P.G., Calonego, J.C., Rosolem, C.A., Scala Jr., N. La, 2017. Cover crop rotations in no-till system: short-term CO_2 emissions and soybean yield. Scientia Agricola 75, 18-26.

Romaneckas, K., Romaneckiene, R., Šarauskis, E., Pilipavicius, V., Sakalauskas, A., 2009. The effect of conservation primary and zero tillage on soil bulk density , water content , sugar beet growth and weed infestation. Agronomy Research 7, 73-86.

Roper, M.M., Ward, P.R., Keulen, A.F., Hill, J.R., 2013. Under no-tillage and stubble retention, soil water content and crop growth are poorly related to soil water repellency. Soil and Tillage Research 126, 143-150.

Ruiz, S., Or, D., Schymanski, S.J., 2015. Soil penetration by earthworms and plant roots - Mechanical energetics of bioturbation of compacted soils. PLoS ONE 10.

Saenger, A., Cécillon, L., Sebag, D., Brun, J.J., 2013. Soil organic carbon quantity, chemistry and thermal stability in a mountainous landscape: A Rock-Eval pyrolysis survey. Organic Geochemistry 54, 101–114.

Sainepo, B.M., Gachene, C.K., Karuma, A., 2018. Assessment of soil organic carbon fractions and carbon management index under different land use types in Olesharo Catchment, Narok County, Kenya. Carbon Balance Manag. 12;13(1):4.

Sainju, U.M., 2015. Comparison of Net Global Warming Potential and

Greenhouse Gas Intensity Affected by Management Practices in Two Dryland Cropping Sites. Journal of Environmental Protection 6, 1042, 1056.

Salem, H.M., Valero, C., Muñoz, M.Á., Rodríguez, M.G., Silva, L.L., 2015. Short-term effects of four tillage practices on soil physical properties, soil water potential, and maize yield. Geoderma 237, 60-70.

Sapkota, T.B., Mazzoncini, M., Bàrberi, P., Antichi, D., Silvestri, N., 2012. Fifteen years of no till increase soil organic matter, microbial biomass and arthropod diversity in cover crop-based arable cropping systems. Agronomy for Sustainable Development 32, 853-863.

Sauerbeck, D.R., 2001. CO_2 emissions and C sequestration by agriculture - Perspectives and limitations. Nutrient Cycling in Agroecosystems 60, 253-266.

Schneider, C.A., Rasband, W.S., Eliceiri, K.W., 2012. NIH Image to ImageJ: 25 years of image analysis. Natural Methods 9, 671-675.

Sebag, D., Verrecchia, E.P., Cécillon, L., Adatte, T., Albrecht, R., Aubert, M., Bureau, F., Cailleau, G., Copard, Y., Decaens, T., Disnar, J.R., Hetényi, M., Nyilas, T., Trombino, L., 2016. Dynamics of soil organic matter based on new Rock-Eval indices. Geoderma 284, 185-203.

Shen, Y., Tanner, C.B., 2010. Radiative and Conductive Transport of Heat through Flail-Chopped Corn Residue. Soil Science Society of America Journal 54, 653-663.

Shen, Y., McLaughlin, N., Zhang, X., Xu, M., Liang, A., 2018. Effect of tillage and crop residue on soil temperature following planting for a Black soil in Northeast China. Scientific Reports 8.

Shipitalo, M.J., Le Bayon, R.C., 2004. Quantifying the effects of earthworms on soil aggregation and porosity. Earthworm Ecology 10, 183-200.

Singh, J.S., Gupta, S.R., 1977. Plant decomposition and soil respiration in terrestrial ecosystems. The Botanical Review 43, 449-528.

Sisti, C.P.J., Dos Santos, H.P., Kohhann, R., Alves, B.J.R., Urquiaga, S., Boddey, R.M., 2004. Change in carbon and nitrogen stocks in soil under 13 years of conventional or zero tillage in southern Brazil. Soil Tillage Research 76, 39-58.

Six, J., Elliott, E.T., Paustian, K., 1999. Aggregate and Soil Organic Matter Dynamics under Conventional and No-Tillage Systems. Soil Science Society America Journal 63, 1350–1358.

Six, J., Conant, R.T., Paul, E.A., Paustian, K., 2002. Stabilization mechanisms of soil organic matter: Implications for C-saturation of soils. Plant Soil. 241, 155-176.

Six, J., Bossuyt, H., Degryze, S., Denef, K., 2004. A history of research on the link between (micro)aggregates, soil biota, and soil organic matter dynamics. Soil and Tillage Research 79, 7–31.

Six, J., Elliot, E.T., Paustian, K., 2000. Soil microaggregate turnover and microaggregate formation: a mechanism for C organic under no-tillage agriculture. Soil Biology and Biochemistry 32, 2099–2103.

Six, J., Feller, C., Denef, K., Ogle, S.M., de Moraes, J.C., Albrecht, A., 2002. Soil organic matter, biota and aggregation in temperate and tropical soils - Effects of no-tillage. Agronomie 22, 755-775.

Six, J., Ogle, S.M., Breidt, F.J., Conant, R.T., Mosiers, A.R., Paustian, K., 2004. The potential to mitigate global warming with no-tillage management is

only realized when practised in the long term. Global Change Biology 10, 155–160.

Six, J., Paustian, K., 2014. Aggregate-associated soil organic matter as an ecosystem property and a measurement tool. Soil Biology Biochemistry. 68, A4-A9.

Skaalsveen, K., Ingram, J., Clarke, L.E., 2019. The effect of no-till farming on the soil functions of water purification and retention in north-western Europe: A literature review. Soil Tillage Research 189, 98-109.

Smith, P., Powlson, D.S., Glendining, M.J., Smith, J.O.U., 1998. Preliminary estimates of the potential for carbon mitigation in European soils through no-till farming. Global Change Biology 4, 679-685.

Soane, B.D., Ball, B.C., Arvidsson, J., Basch, G., Moreno, F., Roger-Estrade, J., 2012. No-till in northern, western and south-western Europe: A review of problems and opportunities for crop production and the environment. Soil and Tillage Research 118, 66-87.

Somasundaram, J., Reeves, S., Wang, W., Heenan, M., Dalal, R., 2017. Impact of 47 Years of No Tillage and Stubble Retention on Soil Aggregation and Carbon Distribution in a Vertisol. Land Degradation Development 28, 1589-1602.

Song, K., Yang, J., Xue, Y., Lv, W., Zheng, X., Pan, J., 2016. Influence of tillage practices and straw incorporation on soil aggregates, organic carbon, and crop yields in a rice-wheat rotation system. Scientific Reports 6: 36602.

Soucémarianadin, L., Cécillon, L., Chenu, C., Baudin, F., Nicolas, M., Girardin, C., Barré, P., 2018. Is Rock-Eval 6 thermal analysis a good indicator of soil organic carbon lability? – A method-comparison study in forest soils. Soil

Biology Biochem. 117, 108-116.

Staley, T.E., 1999. Soil microbial biomass alterations during the maize silage growing season relative to tillage method. Soil Science Society of America Journal 63, 1845-1847.

Szafranek-Nakonieczna, A., Wolińska, A., Zielenkiewicz, U., Kowalczyk, A., Stępniewska, Z., Błaszczyk, M., 2019. Activity and Identification of Methanotrophic Bacteria in Arable and No-Tillage Soils from Lublin Region (Poland). Microbial Ecology 77, 701-712.

Taina, I.A., Heck, R.J., Elliot, T.R., 2008. Application of X-ray computed tomography to soil science: A literature review. Canadian Journal of Soil Science 88, 1-19.

Tisdall, J.M., Oades, J.M., 1980. The effect of crop rotation on aggregation in a red-brown earth. Australian Journal of Soil Research 18, 423-433.

Tresch, S., Moretti, M., Bayon, R.C. Le, Mäder, P., Zanetta, A., Frey, D., Stehle, B., Kuhn, A., Munyangabe, A., Fliessbach, A., 2018. Urban soil quality assessment-a comprehensive case study dataset of urban garden soils. Frontiers in Environmental Science 6:136.

Triplett, G.B., Dick, W.A., 2008. No-tillage crop production: A revolution in agriculture! Agronomy Journal 3, 153-165.

Trumbore, S.E., 1997. Potential responses of soil organic carbon to global environmental change. Proceedings National Academy Sciences 94, 8284-8291.

Tuzzin de Moraes, M., Debiasi, H., Carlesso, R., Cezar Franchini, J., Rodrigues da Silva, V., Bonini da Luz, F., 2016. Soil physical quality on tillage and cropping systems after two decades in the subtropical region of Brazil. Soil

Tillage Research 155, 351-362.

Upton, A., Vane, C.H., Girkin, N., Turner, B.L., Sjögersten, S., 2018. Does litter input determine carbon storage and peat organic chemistry in tropical peatlands? Geoderma 326, 76-87.

Ussiri, D.A.N., Lal, R., 2009. Long-term tillage effects on soil carbon storage and carbon dioxide emissions in continuous corn cropping system from an alfisol in Ohio. Soil and Tillage Research 104, 39-47.

van Bavel, C.H.M., 1950. Mean Weight-Diameter of Soil Aggregates as a Statistical Index of Aggregation. Soil Science Society of America Journal 14, 20-23.

Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. Microbial biomass measurements in forest soils: Determination of kCvalues and tests of hypotheses to explain the failure of the chloroform fumigation-incubation method in acid soils. Soil Biology and Biochemistry 19, 689–696.

VandenBygaart, A.J., Protz, R., Tomlin, A.D., 2011. Changes in pore structure in a no-till chronosequence of silt loam soils, southern Ontario. Canadian Journal of Soil Science 79, 149-160.

Venables, W.N., Ripley, B.D., 2002. Modern Applied Statistics with S Fourth edition by, World. Springer-Verlag. New York.

Villarreal, R., Soracco, C.G., Lozano, L.A., Melani, E.M., Sarli, G.O., 2017. Temporal variation of soil sorptivity under conventional and no-till systems determined by a simple laboratory method. Soil Tillage Research168, 92-98.

Vogel, H.J., Kretzschmar, A., 1996. Topological characterization of pore space in soil - Sample preparation and digital image-processing. Geoderma 73,

23-38.

Wallis, M.G., Horne, D.J., 1992. Soil Water Repellency. Advances in Soil Science 20, 91-146.

Wang, Y., Zhang, Y., Zhou, S., Wang, Z., 2018. Meta-analysis of no-tillage effect on wheat and maize water use efficiency in China. Science of the Total Environment 635, 1372-1382.

Wang, Z., Liu, L., Chen, Q., Wen, X., Liu, Y., Han, J., Liao, Y., 2017. Conservation tillage enhances the stability of the rhizosphere bacterial community responding to plant growth. Agronomy for Sustainable Development 37:44.

Welkenhuyzen, F., Kiekens, K., Pierlet, M., Dewulf, W., 2010. Industrial computer tomography for dimensional metrology: Overview of influence factors and improvement strategies, in: Optical Measurement Techniques for Structures & Systems. Welkenhuyzen.

Wendt, J.W., Hauser, S., 2013. An equivalent soil mass procedure for monitoring soil organic carbon in multiple soil layers. European Journal of Soil Science 64, 58–65.

West, T.O., Post, W.M., 2002. Soil Organic Carbon Sequestration Rates by Tillage and Crop Rotation. Soil Science Society America Journal 66, 1930-1946.

West, T.O., Six, J., 2007. Considering the influence of sequestration duration and carbon saturation on estimates of soil carbon capacity. Climate Change 80, 25-41.

Whalley, W.R., Ober, E.S., Jenkins, M. Measurement of the matric potential of soil water in the rhizosphere. Journal Experimental Botany 64, 3951-3963.

Yvan, C., Stéphane, S., Stéphane, C., Pierre, B., Guy, R., Hubert, B., 2012. Role of earthworms in regenerating soil structure after compaction in reduced tillage systems. Soil Biology and Biochemistry 55, 93-103.

Zhang, S., Li, Q., Zhang, X., Wei, K., Chen, L., Liang, W., 2012. Effects of conservation tillage on soil aggregation and aggregate binding agents in black soil of Northeast China. Soil Tillage Res. 124, 196-202.

Zheng, H., Liu, W., Zheng, J., Luo, Y., Li, R., Wang, H., Qi, H., 2018. Effect of long-term tillage on soil aggregates and aggregate-associated carbon in black soil of northeast China. PLoS ONE 13.

Zhou, H., Li, B., Lu, Y., 2009. Micromorphological analysis of soil structure under no tillage management in the black soil zone of Northeast China. Journal of Mountain Science 6, 173-180.

Zhou, H., Lu, Y., Li, B., 2009. Advancement in the study on quantification of soil structure. Acta Ecologica Sinica 46, 501–506.