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Summary

This research project tests the hypothesis that (i) increased root growth of selected deeper rooting pasture species, stimulated by biochar (a form of charcoal) application at the base of the normal root zone, will accelerate the accumulation of the root-derived carbon, and (ii) the soil carbon sink will be increased because root-derived carbon will be stabilised in the more C unsaturated subsoil. The research is focussed at the renovation phase of permanent pastures, when older pastures are cultivated and re-sown in order to maintain their productive potential. Cultivation provides the opportunity to amend soils with materials rich in recalcitrant C, such as biochar, with the intention of increasing the soil C sink beyond the C saturation limit of a specific soil.

The study has involved the establishment of lysimeter trials and field plot trials using plant species with contrasting root systems and the addition of biochars designed to relieve plant growth constraints in two contrasting soils, an imperfectly drained Fragiaqualf (Pallic soil) and an infertile, drought prone, Typic Udipsamment (Sandy Recent Soil). The concept involves providing "soil-customised" biochars to improve the lower root zone drainage characteristics in the Pallic soil, whereas in the Sandy Recent Soil the role of the biochar was to increase lower root-zone nutrient availability.

Three phases of the research programme were conducted.

(i) Biochar was manufactured from waste pine for lysimeter and field trials using the Pallic soil and from fresh air-dried biosolids or pelletised biosolids for the lysimeter or field trials, respectively, using the Sandy Recent Soil. A progress report (Hedley et al., July 2012) presented (i) biochar manufacture, (ii) trial establishment, (iii) the results of plant growth rates in all trials, (iv) mineral N leaching from the lysimeters in year 1, and (v) the first root density results from the forage rape grown in the Sandy Recent Soil.

This final report presents the soil and root analysis at the conclusion of the lysimeter trials and at 2 years after establishment of the two field trials.

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Proof of concept Lysimeter Trials

A 2-year lysimeter trial was set-up to compare changes in C stocks of soils under either deep or shallow-rooting pasture species and to investigate whether biochar addition below the top 10 cm could promote root growth at depth. Pipe lysimeters were used to collect soil columns (20 cm diameter and 40 cm deep) from permanent pastures growing on Tokomaru silt loam (Pallic soil) and Motuiti sand (dune phase, Sandy Recent Soil). Soil ploughing at cultivation for pasture establishment was simulated in the two contrasting soils by inverting the 0–10 and 10–20 cm depth soil layers, whilst the 20-40 cm depth remained undisturbed. Biochar made from pine waste was applied to 50% of the lysimeters containing Tokomaru soil, and biochar made from dried biosolids was applied to 50% of the lysimeters containing Motuiti sand. The biochar was mixed at a rate of 10 Mg ha^{-1} in the buried topsoil layer, Three pasture types with contrasting root systems (shallow and deep) were established on the lysimeters and grown for two years (i) perennial ryegrass (*Lolium perenne* L.), ii) red clover (*Trifolium pratense* L.) + cocksfoot (*Dactylis glomerata* L.) mixture, and iii) either chicory (*Chicorium intybus* L.) was planted in the Tokomaru soil (experiment 1), or lucerne (*Medicago sativa* L.) in the Motuiti soil (experiment 2) only, as a more drought resistant species replacing chicory. Cumulative dry matter yields after 2 years were highly dependent on pasture type and ranged between 24 Mg ha^{-1} for chicory and 63 Mg ha^{-1} for the mixture of red clover + cocksfoot in the Tokomaru silt loam, and between 29 Mg ha^{-1} for ryegrass and 42 Mg ha^{-1} for the mixture of red clover + cocksfoot in the Motuiti sandy soil. Biochar amendment did not significantly influence either pasture yield or root growth. After two years the lysimeters were dismantled and the soil profile sectioned and chemically analysed. In the Tokomaru silt loam, soil inversion resulted in a net loss of native organic C in the buried horizon under shallow-rooted

ryegrass, but not under the deeper-rooted red clover + cocksfoot, where there was a net gain in soil carbon. The addition of a C-rich, pine biochar (equivalent to 7.6 Mg C ha^{-1}) to this soil sustained a net C gain (21–40% over the non-biochar treatment, $P < 0.10$) in the buried soil layer under all pasture treatments; this overcame the net loss of native organic C in this horizon under shallow-rooted pastures. In the Motuiti sandy soil all pasture species were able to maintain soil C stocks at 10–20 cm depth over time. In this soil, the exposure of a skeletal and nutrient-depleted soil layer at the surface may have fostered root growth at depth. The addition of a nutrient-rich biochar (equivalent to 3.6 Mg C ha^{-1}) to this soil had no apparent effect on total C stocks.

The depletion of soil C stocks, whilst soils undergo lysimeter experiments has been observed in one previous new Zealand study, so the ability of the deeper rooted red clover and cocksfoot to build soil carbon stocks is exciting and worth further investigation; particularly as new cultivars of more persistent red clovers have been bred.

Field trials in a fine textured, imperfectly drained soil

In this field trial biochar, manufactured from pine waste, was incorporated at ploughing into the subsoil of an imperfectly drained Fragiaqualf (Tokomaru silt loam, Pallic soil) at the stage of pasture renovation to promote pasture root growth. Both the incorporation of the biochar and increased root growth were expected to increase soil carbon in the poorly drained fine textured soil. Pine biochar was applied at a rate of 10 Mg ha^{-1} using a mouldboard plough down to 25 cm and three pasture types with contrasting root systems were grown (rye grass + white clover; cocksfoot + red clover; and chicory + white clover). Cultivation caused a decrease in soil bulk density, to values < 1.3 Mg m^{-3}, independently of the presence of biochar. The growth of ryegrass, cocksfoot and chicory based pastures was not affected by the deeper placement of biochar, a result supported by the earlier lysimeter trials. (Shallow placement of biochar in topsoil has decreased yield of pastures in work reported by others). Soil cores taken two years after deep mouldboard ploughing of the permanent pasture under ryegrass and white clover swards showed that soil C and N stocks to 30 cm had increased by 7–9 % compared to not undertaking the regrassing. The increment in C and N stocks resulted from the inversion of original C rich topsoil to depth and the rebuilding of C in newly established root zone of the resown ryegrass and white clover. Ninety three percent of the stable C incorporated as biochar remained after two years resulting in a 16% increment in soil C to 30 cm, when compared to other non-biochar tillage treatments.

These field trials have provided important information that highlights the potential to build soil C stocks by using ploughs that can place topsoil carbon at depth, and bring C poor subsoils to the surface. In these imperfectly-drained soils, loss of the “buried” topsoils C due to decomposition is slower than the rate at which pasture species can re-build soil C in the C poor sub-soil that has been brought to the surface. In addition the trial clearly showed that biochar C added to these soil is conserved, adding to soil C stocks. Establishing further work on other soils with special ploughs designed to invert topsoils is recommended, as is resampling the existing field trial site 5, and 10 years after establishment.

Field trial in a coarse textured sandy soil

A field-scale experiment was conducted over 2 years to evaluate the agronomic effectiveness of direct-drilled biosolids, and biochar produced from biosolids, with a conventional fertiliser treatment. Old pasture was sprayed with glyphosphate, then 3 weeks later pelletised biosolids (Aa grade, at two rates, Bio-H, Bio-L, 13 and 4.5 Mg ha^{-1} respectively) and biochar (Char,

rate: 5.0 Mg ha^{-1}), produced from the same batch of biosolids at temperatures ranging from 550 to 690 °C, were drilled into the sandy Recent soil (Typic Udipsamment) at 15 cm depth. Basal fertiliser applied by the farmer (Fert) was used as a control. As part of pasture renovation, a forage rape crop was established by direct drilling, grazed in autumn and winter by sheep and then followed by direct drilled annual ryegrass, which was also grazed by sheep. Plant growth and nitrogen (N) and phosphorus (P) uptake were monitored. Soils were sampled after 2 yr to assess total carbon (C) and N stocks. The forage crop and annual ryegrass herbage yields showed a marked response to the amount of available N recovered by plants, especially under Bio-L and Bio-H treatments (up to 74% of N recovered by Bio-H). However, at the end of the 2-yr experiment root mass, soil C and N stocks in all treatments where organic amendments were applied were reduced compared to the Fert treatment. Differences in soil C (*i.e.*, $C_{Amendment} - C_{Fert}$) were large (5–20 Mg C ha^{-1}) despite the addition of exogenous organic C (2.0, 2.7 and 5.9 Mg C ha^{-1} for Bio-L, Char and Bio-H treatments, respectively) at the beginning of the experiment. Carbon losses detected under biosolids amendments could be triggered by the priming effect of the added N on organic matter mineralisation.

The trial has failed to show that deep placement of biochar and biosolids can build the native organic C stocks in these coarse sandy textured soils when compared to the current farmer practice. The loss of soil C from these soils through N enrichment, as in the non-carbonised biosolids treatments, has been demonstrated in other New Zealand research studies using lysimeters, therefore it is recommended that field trials need to be established on sandy recent soils that are currently being developed with centre pivot irrigation and fertiliser application inorder to allow soil C stocks to be monitored..

The prototype biosolids drilling system, which was built for this trial, provided a very successful method of incorporating pelletised biosolids (and biochar) at depth. Since this trial, the farmer commissioned Massey University to build a full scale biosolids injector, which has been used on the farm to place biosolids safely at depth on grazed pasture soils.

Overall Conclusions

The lysimeter studies using imperfectly-drained, fine-textured soils have upheld parts of our hypothesis (i), such as cocksfoot + red clover mixtures, producing greater above ground yields than ryegrass or chicory swards, accelerated the accumulation of the root-derived carbon. The lysimeter studies could not demonstrate hypothesis (ii) that soil C at depth can be stabilised to a greater extent by placing biochar at that depth. These trials however, did show that biochar C placed at depth will offset losses of soil C through decomposition. Biochar treatments in lysimeter and field trials did not stimulate more shoot, or root growth, of selected pasture species than soil alone.

In the field trial on imperfectly drained fine textured soils, inversion of topsoils rich in C followed by reseeding pasture into the C poor subsoil, which had been brought to the surface, appears to be a promising method for increasing soil C stocks. Moreover, the lysimeter study showed that this effect was specially accentuated when deep rooting species can be established .

In the coarse textured sandy soil, inversion of the topsoil and establishment of pastures in the lysimeter studies did not stimulate significant soil carbon change (neither loss nor gain) by the end of two years. In the field trial however, under the undisturbed conditions of direct drilling – and therefore no inversion of the topsoil −, the addition of high N biosolids stimulated forage crop and pasture yields but also caused large reductions in topsoil C. This is consistent with much of the topsoil C in well-drained coarse textured sands being "unstabilised" in the absence of a soil clay fraction. Farming practices that will lead to soil C stabilisation, or loss, in these soils requires further research.

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Research Aim

To increase the permanent C sink in soils of already developed grazed pasture systems through the incorporation of biochar promoting the growth of deeper rooting plant species.

Introduction

A research team from the NZBRC-Massey University (M.J. Hedley, M. Camps-Arbestain, R. Calvelo Pereira, P. Bishop and E. Wisnubroto), Plant and Food Research (S. Green) and Landcare Research's Global Change Processes team (S. Saggar) is undertaking this research that aims to increase the stocks of stabilised soil carbon in pasture soils as a greenhouse gas mitigation strategy. Approximately 50% of soil carbon in New Zealand is under grazed pasture management. If full carbon accounting becomes part of future commitments to international climate change mitigation protocols, then land management techniques that can be applied to increase soil carbon stocks under grazed pastures must be developed, verified and their wider benefits and costs assessed. To achieve increasing soil carbon stocks will require that more of the plant-derived carbon entering soils is transformed into soil carbon sinks of greater permanence than those occupied by the majority of current topsoil carbon. Greater permanence of soil carbon is not achieved by modifying the chemical nature of plant carbon entering the soil. Greater permanence is only achieved when new carbon is stabilised through organo-mineral complexation, or is converted to highly aromatic carbon such as biochar, or, when its decomposition is limited by soil physical or biological conditions. The dynamic equilibrium between biological, chemical and physical stabilisation of soil organic matter (SOM) and its rate of decomposition in NZ pasture soils is such that topsoil carbon stocks are believed to be at or near their maximum capacity. Significant increases in soil carbon stocks can only be achieved by addition of recalcitrant carbon such as biochar or by managing the roots of pasture or forage plants to contribute more carbon to subsoil pools, where stabilisation processes do not appear to be saturated and new carbon has a slower rate of decomposition.

This research project tests the hypothesis that increased root growth stimulated by biochar application to soils will accelerate mineral weathering (in subsoils rich in highly weatherable primary minerals) and promote soil formation in the subsoil. Decomposition of the root-derived carbon will lead to more stabilised C in the subsoil and less soil-derived greenhouse gas (GHG) emissions.

The research is conducted in 4 phases: (i) biochars (a form of charcoal) are made from waste biomass. The biochar properties, determined by the feedstock type and the pyrolysis process, are tailored to overcome root-growth-limiting properties of the receiving soils; (ii) equipment is designed to incorporate biochars at depth in selected soils. Deep placement of biochars has the potential to promote root growth into the subsoil; (iii) plant species with contrasting root systems will be established to take advantage of the biochar-amended soils; and (iv) soil carbon concentrations, root densities will be periodically measured in the soil profiles.
The study involves the establishment of lysimeter trials and field plot trials using plant species with contrasting root systems and the addition of biochars designed to fulfill the soil/plant needs at two different sites (a Pallic Soil and a Sandy Recent Soil).

This research will provide information that will allow land managers and agribusiness to develop farming system economic models around soil carbon sequestration. Moreover, the knowledge acquired will inform decisions to be made on role of soil carbon in the post-2012 Kyoto commitment period and in subsequent variations of the Emissions Trading Scheme (NZETS).

1. Lysimeter Trials

The purpose of the lysimeter trial is to provide a semi-controlled environment in which some spatial variability of soil properties can be removed and the influence of the biochar and sward treatments on soil carbon and nitrogen dynamics can be determined with less "background noise". The lysimeters also allow N leaching in drainage and GHG emissions from the soil surface to be measured. GHG emissions could be influenced by disturbing the soil to introduce the biochar and the subsequent effect of biochar on N dynamics once placed in the soil.

1.1. INTRODUCTION

Enhancing soil carbon (C) sequestration by the conversion of cropland into pastures is recognised as a potential climate change mitigation strategy (Lal *et al.*, 2011). In some areas, large soil C gains have also been observed when pasture replaces forest or scrub as land use (Hedley *et al.*, 2009). This has been attributed to the preferential below-ground C translocation and partitioning by pasture plants (Kuzyakov and Domanski, 2000; Rasse *et al.*, 2005). Over time, a new dynamic equilibrium is achieved in the established pasture where the stabilisation-decomposition mechanisms reach a steady state (Stewart *et al.*, 2007). Several studies suggest that there is a limit to the stabilisation of additional C input to soil – the so called "saturation level" – (Six *et al.*, 2002; Stewart *et al.*, 2007), and that the capacity of a soil to sequester C under specific land use and management will tend to decrease as the soil C stock becomes at or near the upper limit for soil C accumulation.

Assessments on the status of soil C stocks in New Zealand pasture soils indicate either a decline (Schipper *et al.*, 2014), an increase (Mudge *et al.*, 2011) or no change (Tate *et al.*, 2005) in soil C levels during the last decades. Such contrasting trends are partially explained by vertical, lateral and temporal variability in soil properties, and differences in the methodological approach used. As suggested by Dodd *et al.* (2011), it may be difficult to increase C stocks of New Zealand pastoral soils with relatively high C in the 0–10 cm depth zone where soils may be near or close to the upper limit for C accumulation. Recently, Beare *et al.* (2014) estimated the upper C stabilisation limit of New Zealand pastoral soils based on data from measurable properties of the fine mineral fraction and found that most soils have a C deficit, this being greater in subsurface (median = 15 mg C g^{-1}) than in surface horizons (median = 12 mg C g^{-1}). Despite the limitations acknowledged in these studies, their findings suggest that C sequestration strategies focused on the increase of plant-derived C input in subsurface horizons should not be disregarded, as already proposed by Carter and Gregorich (2010) and Rumpel and Kögel-Knabner (2011).

Pastures require periodic renewal (cultivating and re-sowing) in order to maintain their productive potential (Tozer *et al.*, 2013). This often involves a short-term soil C loss until replenished by further pasture growth (Curtin *et al.*, 2010). Strategies to manage pasture and soil so that this temporary loss is prevented, offset, or rapidly refilled should thus be considered. In fact, the use of deep-rooted pastures has been proposed as a management practice to allocate more C at depth (Carter and Gregorich, 2010; Dodd *et al.*, 2011), although this can be hampered by physical (Carter and Gregorich, 2010) and nutrient constraints of subsurface horizons (Dodd *et al.*, 2011).

Periodic cultivation also offers the opportunity to amend soils with materials rich in recalcitrant C, such as biochar, with the intention of increasing the soil C sink beyond the C saturation limit of a specific soil. Biochar has a dominance of aromatic and condensed chemical structures (McBeath *et al.*, 2011), for which many microbes lack adaptation to use as a source of energy (Lehmann *et al.*, 2015). In addition to C sequestration, biochar has the potential to provide a range of benefits under specific conditions (Jeffery *et al.*, 2013; Lehmann *et al.*, 2011) and therefore the choice of a specific biochar able to fulfil particular

soil needs to be considered. While biochars made from woody material tend to have a greater contribution to soil physical properties – although their contribution to soil cation exchange capacity as biochar surface becomes oxidised over time should not be disregarded (Liang *et al.*, 2006)–, those made from ash-rich feedstock have a greater impact on soil chemistry, including nutrient fertility (Camps Arbestain *et al.*, 2015). Moreover, the depth at which biochar is applied should also be considered, as a deep application may i) decrease the risk associated with surface erosion of fine biochar particles (Major *et al.*, 2010), ii) diminish the potential effect of biochar on soil albedo, and iii) contribute to alleviate nutrient limitations or physical constraints for root development at depth, the latter being common in subsurface horizons of pastoral soils under intensive grazing (Houlbrooke *et al.*, 1997).

In this study a lysimeter trial was set-up to evaluate whether the use of deep rooting pasture species, in the presence or absence of biochar, could influence below-ground C translocation and soil C storage two years after pasture establishment. Soil ploughing at cultivation was simulated by inverting the 0–10 and 10–20 cm depth soil layers, and biochar was added (and mixed) to the buried soil layer, where appropriate. Three pasture types with contrasting root systems were grown on two different soils (a silt loam soil and a sandy soil). Distinctive biochars were selected for these two soils so that soil-specific plant growth limitations could be overcome. The hypothesis tested was that the added benefit of combining the use of deep rooting pasture species and biochar application at depth would maintain or increase soil C storage after pasture renovation.

1.2. MATERIAL AND METHODS

1.2.1. Establishment of the lysimeter trial, experimental design and monitoring

Soils under study

Two soils from the Manawatu-Wanganui region (New Zealand) with contrasted chemical and physical properties were used: i) a Tokomaru silt loam soil (a Typic Fragiaqualf) developed from wind-blown loess of Greywacke origin; and ii) a Motuiti brown sand (a Typic Udipsamment) developed from wind-blown beach sand. Relevant properties of the Tokomaru topsoil (0–10 cm) were: pH 5.07, bulk density 0.94 Mg m^{-3}, total C content 35.6 g kg^{-1}, C/N ratio 9.8, and Olsen P 78.1 g kg^{-1} (Table 1.1). Those of the Motuiti topsoil (0–10 cm) were: pH 5.94, bulk density 1.23 Mg m^{-3}, total C content 27.6 g kg^{-1}, C/N ratio 11.7, and Olsen P 16.6 g kg^{-1}. Plant productivity in the silt loam soil is limited by poor physical structure (Scotter *et al.*, 1979), whereas that in the sandy soil is constrained by low nutrient supply and water availability (Cowie and Rijkse, 1977).

Feedstock selection and biochar production

A C-rich, low-ash biochar (PI-350) was produced from pine (*Pinus radiata* D. Don) sawdust to overcome the physical constraints of the Tokomaru soil by improving the drainage at the ploughed layer. The pine sawdust, once air-dried, was pyrolysed in a gas-fired, rotating drum kiln (inner volume of 25 L) at a heating rate of ca. 16 °C min^{-1} reaching a final maximum temperature of 350 °C. PI-350 biochar had a relatively low pH (7.2), high C content (759 g kg^{-1}, 83% as fixed C; atomic H/C_{org}: 0.62), low CEC (0.9 cmol(+) kg^{-1}) and low N content (3.7 g kg^{-1}) (Table 1.2). The fraction of aromatic C out of total C (f_a), as determined by deconvoluting and integrating the NMR spectra, was 0.73.

An ash-rich biochar rich in available phosphorous (P) (BG-550) was produced from a mixture of biosolids and municipal green-waste to improving soil nutrient status and water retention of the Motuiti soil. Biosolids were recovered after primary sedimentation and dewatering at a wastewater treatment plant (Palmerston North, New Zealand). Green waste consisted of a mixture of leaves, hedge pruning, and chipped wood materials. Biosolids and green waste were pre-dried at 70 °C, then mixed at a ratio of 50:50 (wt:wt) and pyrolysed in 200 g batches

in a gas-fired rotating drum kiln (inner volume of 5 L). The feedstock was heated at a heating rate of ca. 16 °C min^{-1} to a final maximum temperature of 550 °C. BG-550 biochar had a pH of 8.2, a high ash content (51.2%) and CEC (11.3 cmol(+) kg^{-1}); C content was low (356 g kg^{-1}, 86% as fixed C; atomic H/C_{org}: 0.74), and N content was relatively high (14 g kg^{-1}) (Table 1.2). The f_a value for this biochar was 0.87.

Table 1.1 Chemical characteristics, per depth, of the two soil types after repacking in the lysimeters. Analytical methods are described in Blakemore *et al.* (1987).

Property	**Soil**	Experiment 1, silt loam soil			Experiment 2, sandy soil		
	Depth, cm[1]	0–10	10–20	20–30	0–10	10–20	20–30
Bulk density	Mg m^{-3}	1.17	0.92	1.38	1.54	1.30	1.80
C	g kg^{-1}	20.0	35.6	12.5	11.3	27.6	3.3
N	g kg^{-1}	2.3	3.6	1.3	1.1	2.4	0.4
C/N	-	9.4	9.8	9.4	10.3	11.7	8.9
Olsen P	mg kg^{-1}	20.8	78.1	6.2	13.7	16.6	7.6
SO_4	mg kg^{-1}	12.5	18.3	9.0	5.0	12.0	5.0
K	cmol kg^{-1}	0.79	1.55	0.45	0.18	0.35	0.17
Ca	cmol kg^{-1}	5.10	6.90	5.50	2.40	4.40	1.20
Mg	cmol kg^{-1}	0.84	1.46	0.84	0.44	1.03	0.19
Na	cmol kg^{-1}	0.09	0.14	0.20	0.04	0.12	0.03
CEC	cmol kg^{-1}	17.0	19.0	15.0	6.0	10.0	3.0

[1] Depth of sampling in the lysimeter at time 0.

Table 1.2 Characteristics of biochar produced from pine and biosolids.

Feedstock Property	Units	Pine PI-350	Biosolids[1] BG-550
pH	-	7.2	8.2
Lime equivalence	kg CaCO3/t	7.4	195
Total C	g kg^{-1}	759	356
Total N	g kg^{-1}	2.7	14
Total H	g kg^{-1}	39	22
Total O	g kg^{-1}	173	51
H/C_{org}	At. Ratio	0.62	0.74
O/C_{org}	At. Ratio	0.17	0.11
Total C/ Total N	Ratio	281	25.4
f_a[2]		0.73	0.87
Ash	%	2.6	51.2
Volatile matter	%	30.2	13.4
Fixed C	%	63.2	30.6
Moisture	%	4.0	4.8
CEC	cmol kg^{-1}	0.88	11.3

pH, total C, H, O and N contents, ash content and volatile matter content (dry basis) and the stable, thermo-resistant fraction or fixed C (dry basis) determined following Calvelo Pereira *et al.* (2011).
Cation exchange capacity (CEC) was measured with $SrCl_2$, as described in Calvelo Pereira *et al.* (2014).
Lime equivalence was determined following Singh *et al.* (2010).
[1] mixed with green waste, 50:50 (wt:wt); [2] f_a, fraction of biochar-C that is aromatic following complementary characterisation by using solid-state cross-polarisation magic-angle spinning CPMAS ^{13}C NMR spectroscopy.

Soil collection and lysimeter establishment

Fifty PVC pipe lysimeters (PVC columns 40 cm depth and 20 cm in diameter) were set up at Palmerston North, New Zealand in December 2010 using the two above-mentioned soils. The trial involved two similar experiments, one using the Tokomaru soil (24 columns + additional 1 column for a destructive sampling at time 0) and one using the Motuiti soil (24 columns + 1 additional column for a destructive sampling at time 0), simulating biochar incorporation into depth when ploughing the soil for seed bed preparation at cultivation. For each experiment, 12 columns included either PI-350 or BG-550 as amendment (biochar-amended soil treatment) and the other 12 columns did not (non-biochar soil treatment).

Experiment 1: Tokomaru soil amended with PI-350 biochar. Soil cores at 0–10, 10–20, and 20–40 cm depth were taken from a pasture site (40°23′S, 175°36′E) nearby Palmerston North. The 0–10 cm and 10–20 cm depth soil layers were sliced and removed for "cultivation", whereas the 20–40 cm layer was taken intact (Photo 1) using the corresponding PVC column. The 0–10 cm layer was hand-mixed with the PI-350 biochar at an application rate of 10 Mg ha^{-1} and also with NPK fertiliser. This layer was then added to the soil column in the PVC container on top of the 20–40 cm layer, at a depth of 10–20 cm, thus inverting the order of layering to simulate mouldboard ploughing at pasture establishment. The original 10–20 cm depth soil was also hand-mixed to simulate the effect of cultivation and added on top of the soil column, at a depth of 0–10 cm, without amendments.

Experiment 2: Motuiti soil amended with BG-550 biochar. Soil cores for the 0–10, 10–20 and 20–40 cm depth were taken from a pasture site (40°9′S, 175°16′E) nearby Palmerston North. The same procedure as for experiment 1 was followed for the Motuiti soil, except that all soil depths had to be repacked into the lysimeter pipe due to the poor structure of this soil.

Once the columns with the two soils were finalised, the lysimeters from experiment 1 and experiment 2 were attached to 1.3 m PVC drainage collection flux meters and lowered into holes bored into the ground such that the surface of the column was 2 cm above ground level. The spaces between columns were covered with a ryegrass lawn mown at 2 cm height (Photo 2). Soil temperature, moisture fluctuations and drainage were monitored regularly using sensors installed in selected lysimeters. The site had a fully equipped weather station to monitor environmental changes.

Soils at time 0 (prior to seeding). An additional PVC pipe lysimeter was prepared for each soil, following exactly the same "cultivation" as described previously and sampled thereafter for physical and chemical characterisation.

Photo 1.1 (Left) Details of how the intact soil cores (200-400 mm depth) were taken at the Tokomaru soil field site.

Photo 1.2 (Right) View of the Lysimeter site showing cores in place, swards established and the lateral irrigator.

Selection of pasture species, experimental design, planting and monitoring

Pasture species were selected i) based on those that successfully grow in both soil types, and ii) to provide a range of rooting patterns that may influence dry matter (DM) production both above- and below-ground. The plant species chosen were: i) perennial ryegrass (*Lolium perenne* L.), ii) red clover (*Trifolium pratense* L.) + cocksfoot (*Dactylis glomerata* L.) mixture, and iii) either chicory (*Chicorium intybus* L.) to be planted in the Tokomaru soil (experiment 1) or lucerne (*Medicago sativa* L.) to be included in the Motuiti soil (experiment 2) only, as a more drought resistant species replacing chicory. Seeds were sown 0.5 cm deep on the 23 December 2010, in a randomised complete block design, where four replicates per each pasture and amendment combination were used.

Sprinkler irrigation was applied to maintain the soil moisture at 70% of field capacity. Swards were lightly trimmed in the early phase of growth and then cut back to 5 cm height to simulate grazing. Harvest occurred when the most prolifically growing sward had reached an approximate cover of 3000–4000 kg DM ha^{-1}. Seventeen harvests were taken during the trial period. Harvested herbage was dried to constant weight in an oven at 65 $^{\circ}$C and yields were expressed as biomass removed per unit surface area (kg DM ha^{-1}). Cow dung and urine were distributed evenly after each harvest. Dry cow dung (< 1mm particle size) was reapplied to replace 34% of the C and approximately 20% of the N removed at harvest, whereas synthetic urine replaced 47% of the N removal (Steele, 1982).

Figure 1.1 summarises the main weather conditions during the 26 months of the study. Values registered for global radiation, air temperature and cumulative rainfall were in the range of those typical for Palmerston North (long-term annual figures: average temperature, 13 °C; mean annual rainfall of 920 mm, and mean annual sunlight hours of 1744).

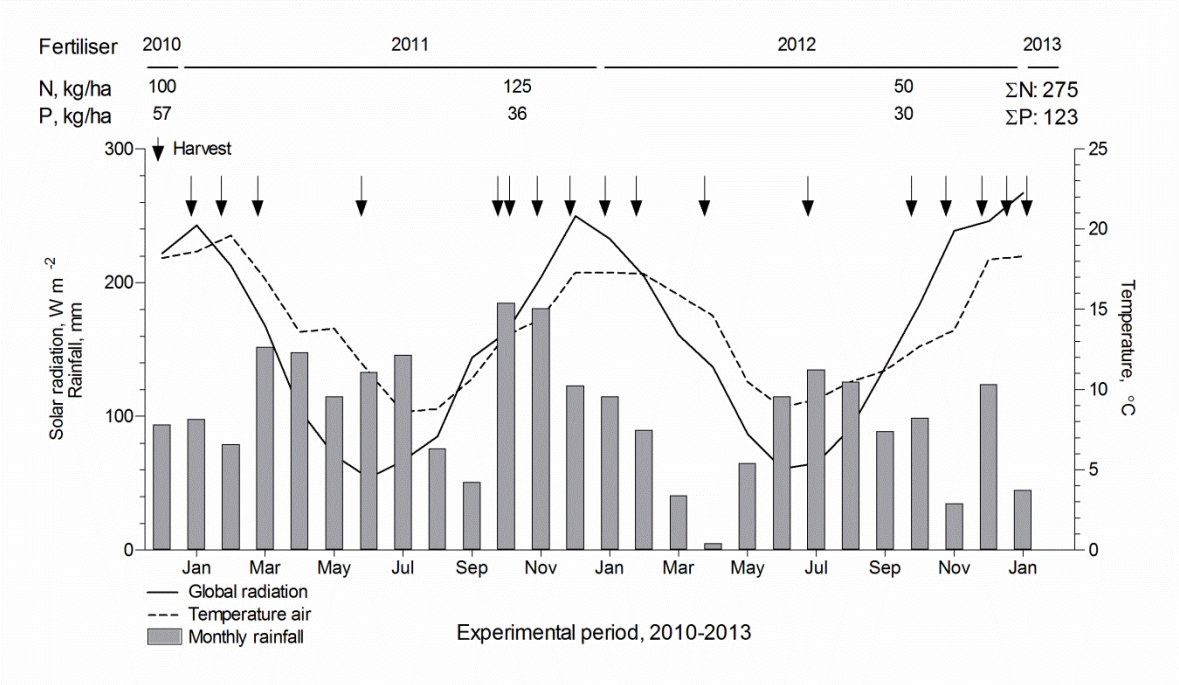

Figure 1.1 Monthly average global radiation, average air temperature and cumulative rainfall from the trial site through the development of the study (2010-2013). A summary of fertiliser application (mainly as N and P) is included; harvest dates are also indicated.

1.2.2. Assessment of soil changes at the end of the trial

Lysimeter sampling

The lysimeter trial ended in January 2013, 777 days (ca. 2 years, T2) after pasture establishment. Final yield was recorded and both experiments dismantled. Lysimeters were lifted out of the flux meters and soil samples obtained by slicing core layers of 20 cm diameter and 2 cm thick at the following depths: 2, 4, 8, 16, 22 and 32 cm. Four 5 cm diameter, 2 cm depth aluminium rings were randomly inserted into the soil surface of each core layer. Each ring was scanned using modified contact probe of ASD FieldSpec 3 Vis-NIR Spectrometer (Kusumo *et al.*, 2009). Vis-NIR spectra were analysed using principal component analysis (PCA) and linear discriminant analysis (LDA) were used to detect soil layers where the wavelength bands due to soil organic matter content had significantly changed. After scanning soil samples were stored at 4 °C until processing.

Chemical characterisation of soil samples

One soil sample per slice for each lysimeter was air-dried, then homogenised and particle size reduced to < 250 μm by gentle grinding. A subsample was used for total soil C and N determinations using a vario MACRO cube CHNS elemental analyser (Elementar Analysensysteme GmbH, Hanau, Germany). Soil samples taken at 14–16 cm depth were additionally treated with $K_2Cr_2O_7$ following Herath *et al.* (2014) to determine the oxidisable organic C (OC_{ox}). Bulk density was calculated for each soil slice considering the volume of

the core used and the weight of the soil oven-dried at 105 °C. Soil pH was measured at a soil:water ratio of 1:2.5 (weight/volume) following Blakemore *et al.* (1987). Samples were stirred and allowed to equilibrate overnight prior to the determination of pH in the supernatant.

Live and dead root density measurement by wet sieving

One soil sample per slice was used to separate a light-biomass fraction (essentially live and dead roots) − except the slice taken at 32 cm depth, in which the live and dead root mass was not measured − following the wet sieving procedure of Kusumo *et al.* (2009). Washed live and dead root material collected on the sieve was oven dried and the live and dead root mass was expressed as mg dry matter per unit of volume and finally the percentage distribution of live and dead root mass in the profile down to 30 cm was calculated.

Assessment of total C and N stocks and net storage changes

Total soil C and N storage was calculated considering the bulk density and thickness of different layers, nominally 0–10, 11–20, 21–30 and 31–40 cm. This allowed us to estimate total C and N stocks for the profile down to 40 cm depth by summing masses for all layers considered. This corresponds to the C (and N) stocks at a fixed depth for each soil. This is the conventional calculation method, which does not fully account for variation in soil mass sampled (Ellert *et al.*, 2001), and hampers any comparison between the treatments due to differences in the mass of soil under consideration. In order to compare soil C (and N) storage between treatments at 0–40 cm depth, the C masses were recalculated based on the equivalent mass for each depth and soil type (*i.e.*, for each experiment).

1.2.3. Statistical analyses

Statistical analyses were conducted with IBM SPSS Statistics version 20 software package (Armonk, New York, USA). Data from each experiment (above-and below-ground biomass, pH, TC and TN concentrations, soil bulk density, TC and TN stocks, and equivalent thickness) were statistically analysed using the GLM procedure of SPSS. The model included the fixed effect of the type of amendment [*i.e.*, biochar-amended soil (biochar made from pine in experiment 1; biochar made from biosolids and greenwaste in experiment 2) and non-biochar soil], the pasture type (*i.e.*, ryegrass, red clover + cocksfoot and chicory, experiment 1; ryegrass, red clover + cocksfoot and lucerne, experiment 2), and the interaction of amendment and pasture type. If a significant ($P < 0.10$) main effect was detected, difference between treatment means was tested using the least significant difference. Relationship between variables was also explored graphically and Pearson's correlation was applied.

1.3. RESULTS

1.3.1. Above- and below-ground biomass

Cumulative average herbage dry matter production after 2 years (Table 1.3) was highly dependent on pasture type and ranged between 24 Mg ha^{-1} for chicory and 63 Mg ha^{-1} for the mixture of red clover + cocksfoot in the Tokomaru silt loam (experiment 1), and between 29 Mg ha^{-1} for ryegrass and 42 Mg ha^{-1} for the mixture of red clover + cocksfoot in the Motuiti sandy soil (experiment 2). Among the pasture species studied, the mixture of red clover + cocksfoot was thus the one that had the highest annual herbage production, and this was irrespective of soil type and amendment (Table 1.3).

Table 1.3 Average (n = 4) and standard error (SEM) for the annual cumulative above-ground herbage production 2 years after "cultivation", biochar addition and growth of different pastures in lysimeters containing Tokomaru silt loam (experiment 1) and in lysimeters containing Motuiti sand (experiment 2).

Experiment 1	Herbage		
Pasture	Biochar	Non-biochar	SEM
Ryegrass	17.25	16.57	0.51
Mixture[1]	29.04	30.03	1.02
Chicory	10.71	11.69	0.27
P-value			
Amendment			*0.606*
Pasture			*< 0.001*
Amendment × Pasture			*0.635*

Experiment 2	Herbage		
Pasture	Biochar	Non-biochar	SEM
Ryegrass	12.87	13.93	0.34
Mixture[a]	23.10	18.53	2.45
Lucerne	13.87	13.91	0.18
P-value			
Amendment			*0.497*
Pasture			*0.003*
Amendment × Pasture			*0.362*

[1] mixture of red clover + cocksfoot

In this study, pasture growth was higher than the annual production in the Manawatu region of New Zealand, which ranges between 7 and 15 Mg DM for the plant species considered (Kemp *et al.*, 2010; Li and Kemp, 2005). It should be noted that leaves of pasture species (*i.e.*, the photosynthetic surface area) tended to extend slightly beyond the surface area of the PVC column and this was more accentuated in the red clover + cocksfoot sward. Therefore, the high herbage production attained by the mixture is the result of expressing yield as a function of the cross-sectional area of the lysimeter.

Total live and dead root mass down to a nominal depth of 30 cm was highly dependent on depth, pasture type, with average values ranging from 1 and 89 mg cm^{-3} in the Tokomaru silt loam (experiment 1) and between 3 and 63 mg cm^{-3}in the Motuiti sandy soil (experiment 2) (Figure 1.2). Similar to the trend in above-ground production, the mixture of red clover + cocksfoot was the one that had the highest live and dead root mass recovered by wet sieving irrespective of soil type and amendment. On average, fifty four percent of the live and dead root mass recovered in the silt loam soil was found in the top 10 cm, while the corresponding value in the sandy soil was 39% (data not shown). In the next 10–20 cm depth, pastures growing on the silt loam accumulated on average ca. 37% of total live and dead root mass, whereas those growing on the sandy soil accumulated on average ca. 49% of the total live and dead root mass recovered by wet sieving at that same depth. Live and dead root mass accumulated at a nominal depth of 20–30 cm depth was always ≤ 12% for all the pasture types considered in this study (data not shown). Biochar only had a significant effect ($P <$ 0.10) on the pasture live and dead root mass distribution pattern when added to the silt loam soil under the mixture of red clover + cocksfoot (experiment 1), with an increase in the average live and dead root mass in the 0–10 cm depth and the corresponding decrease in the

10–20 cm depth, compared to that of the corresponding non-biochar treatment (data not shown).

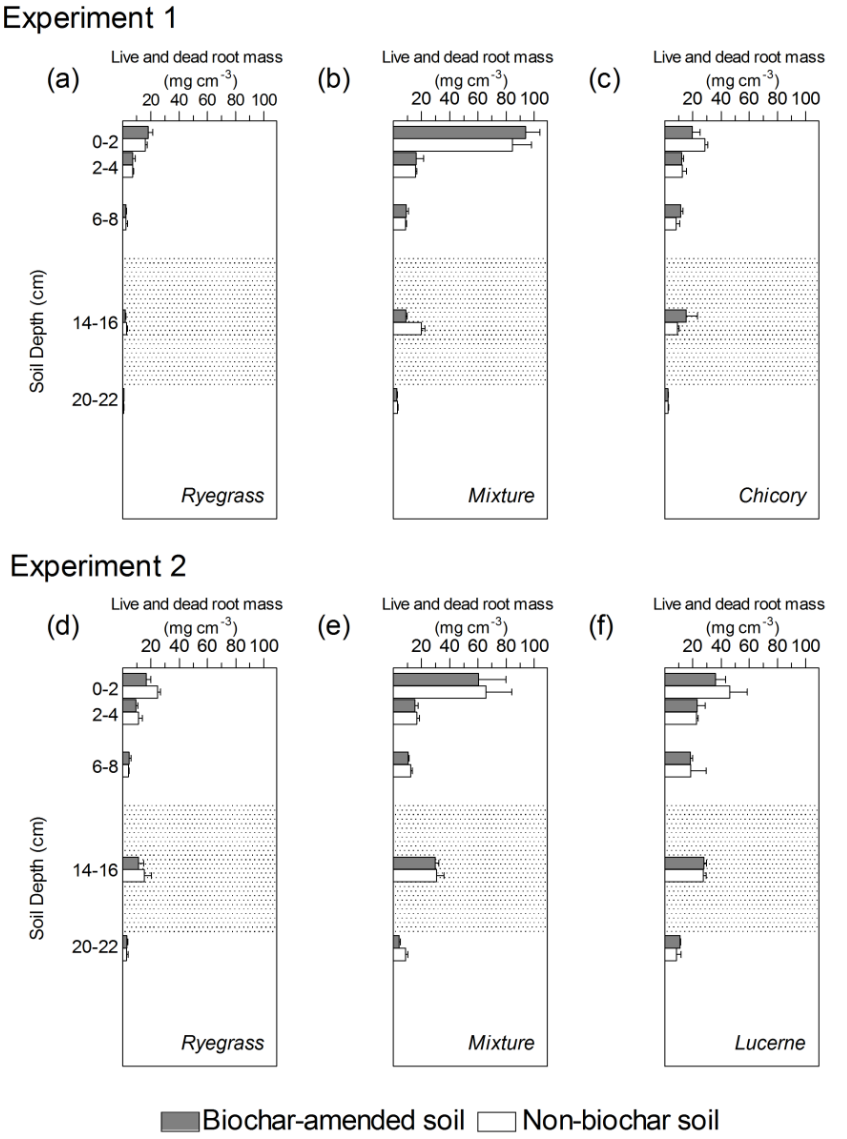

Figure 1.2 Live and dead root mass obtained by wet sieving (up to a nominal depth of 30 cm) 2 years after "cultivation", as influenced by biochar addition and growth of different pastures in lysimeters containing (a) Tokomaru silt loam (experiment 1) and in lysimeters containing (b) Motuiti sand (experiment 2). Data represent the average and SEM, standard error of the mean (n = 4).

These large differences in live and dead root mass between treatments were supported by the spectral reflectance measurements. At the end of the experiment principal component analysis (PCA) of the spectral reflectance of the core slices followed by and linear discriminant analysis (LDA) was able to discriminate differences in Vis-NIR reflectance bands of soil organic matter content, in the Tokomaru silt loam soil, that had changed due to plant species with 75%, 100% and 85% discrimination accuracy for lucerne, red clover + cocksfoot mixture and ryegrass treatments, respectively and due to biochar addition with 100% accuracy. In the Motuiti sandy soil the ability to discriminate was less accurate with 75%, 74% and 60% discrimination accuracy for chicory, cocksfoot mixture and ryegrass treatments, respectively and due to biochar addition with 100% accuracy.

1.3.2. Changes in total C and N distribution and soil bulk density with depth at the end of the trial

Silt loam soil with and without pine biochar amendment under different swards (experiment 1)

At the start of the experiment, immediately after the simulated mouldboard ploughing and the associated inversion of horizons, the repacked soil had 1.8 times higher total soil C (TC) in the 10–20 cm soil layer than that at 0–10 cm (Table 1.1). Soil carbon analyses on the soil sections removed from the lysimeters after 26 months showed that the differences in TC between these two soil layers had narrowed – in the absence of biochar addition − were less with the ratio ranging between 1.1 and 1.3 (Table 1.1; Figure 1.3). Within the first 10 cm depth, TC concentration values were higher in the 0–2 cm topsoil, these ranging between 24 and 49 g kg^{-1} (Figure 1.3), with the maximum values in the soils under the pasture mixture of red clover + cocksfoot. Under all circumstances, TC concentration values in this top10 cm soil layer were greater than the corresponding ones at T0. At the 10–20 cm depth (as measured at 14–16 cm depth) and in the absence of biochar, there was a loss of TC in the ryegrass and chicory treatments compared to T0, but not in the mixture of red clover + cocksfoot. In this layer and at T2, TC values in the treatments with biochar were 7 to 10 g kg^{-1} soil greater ($P < 0.10$) than the treatments without biochar, independent of the pasture type (Figure 1.3). The non-oxidisable fraction of TC in the biochar-amended soils, additional to that in the native soil, ranged between 7 and 9 g kg^{-1} (Figure 1.3), thus closely related to the absolute increase in TC detected in these treatments. Below the top 20 cm depth, TC concentration was always < 13 g kg^{-1}soil (Figure 1.3).

Values of TN at different soil depths paralleled those of TC, except in the presence of biochar (Figure 1.3), as expected, given the low N content of the PI-350 biochar. In the layer where biochar was deployed, values of TC and TN were highly correlated ($r > 0.96$). Bulk density values were inversely correlated with TC concentration values ($r = –0.81$ and –0.70, for the non-biochar and biochar treatments, respectively). The addition of biochar consistently reduced ($P < 0.10$) the bulk density of the Tokomaru soil in the layer it was added (10–20 cm depth), irrespective of the pasture type (Figure 1.3).

Sandy soil with and without biosolids biochar under different swards (experiment 2)

At the start of the experiment, immediately after the simulated mouldboard ploughing and the associated inversion of horizons, the repacked soil had 2.4 times higher TC in the 10–20 cm soil layer than that at 0–10 cm. At the end of the experiment differences between these two soil layers – in the absence of biochar addition − were lower, with this ratio ranging between 1.7 and 2.2 (Table 1.1; Figure 1.4). Pasture type was the factor with the greatest effect ($P < 0.10$) on TC concentration at the depths studied (at 2–4, 6–8 and 30–32 cm depth). Again, TC concentration was higher in the 0–2 cm topsoil, with average values ranging between 14 and 18 g kg^{-1} (Figure 1.4), decreasing in the following 2–10 cm depth with values close or below the TC concentration at T0. At the 10–20 cm depth (as measured at 14–16 cm depth) and in the absence of biochar, TC concentration values remained similar to those at T0, with values ranging from 26 to 27 g kg^{-1} soil. The addition of biochar at this depth did not significantly increase the average TC concentration in that layer (as measured at 14–16 cm depth) − with average values ranging from 24 to 29 g TC kg^{-1} soil −, independently of the pasture type (Figure 1.4). Values of TC in this soil layer were similar in the presence and absence of biochar for the soils under the mixture of red clover + cocksfoot and lucerne; in ryegrass, the average value in the presence of biochar was even smaller than in the absence of it, although differences were not significant at $P = 0.647$. Below the top 20 cm depth, TC concentration was always < 7 g kg^{-1} soil (Figure 1.4).

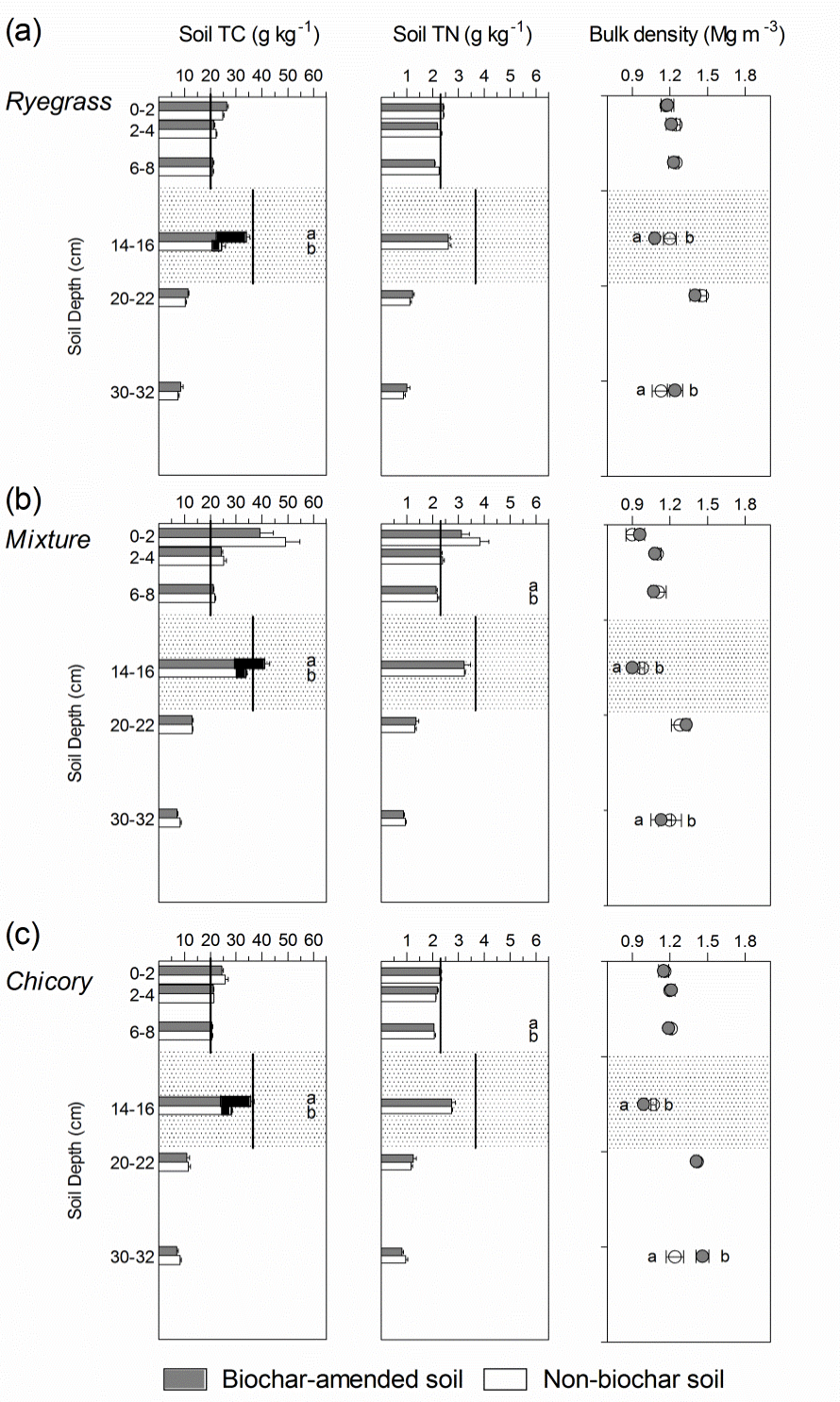

Figure 1.3 Changes in total carbon, total nitrogen and soil bulk density with depth 2 years after "cultivation", biochar addition and growth of different pasture species in lysimeters containing Tokomaru silt loam (experiment 1): (a) ryegrass, (b) mixture (red clover + cocksfoot) and, (c) chicory. Data represent the average and SEM, standard error of the mean (n = 4). For each depth, bars with different letters indicate significant differences between the biochar-amended soil and the corresponding non-biochar soil at *P* < 0.10. At depth 14-16 cm, the difference (TC – OC_{ox}) is indicated (▉) for each treatment. Vertical bars indicate values of TC and TN for the 0–10 and 10–20 cm depths at the beginning of the experiment (T0) for the non-amended soil, as indicated in Table 1.1.

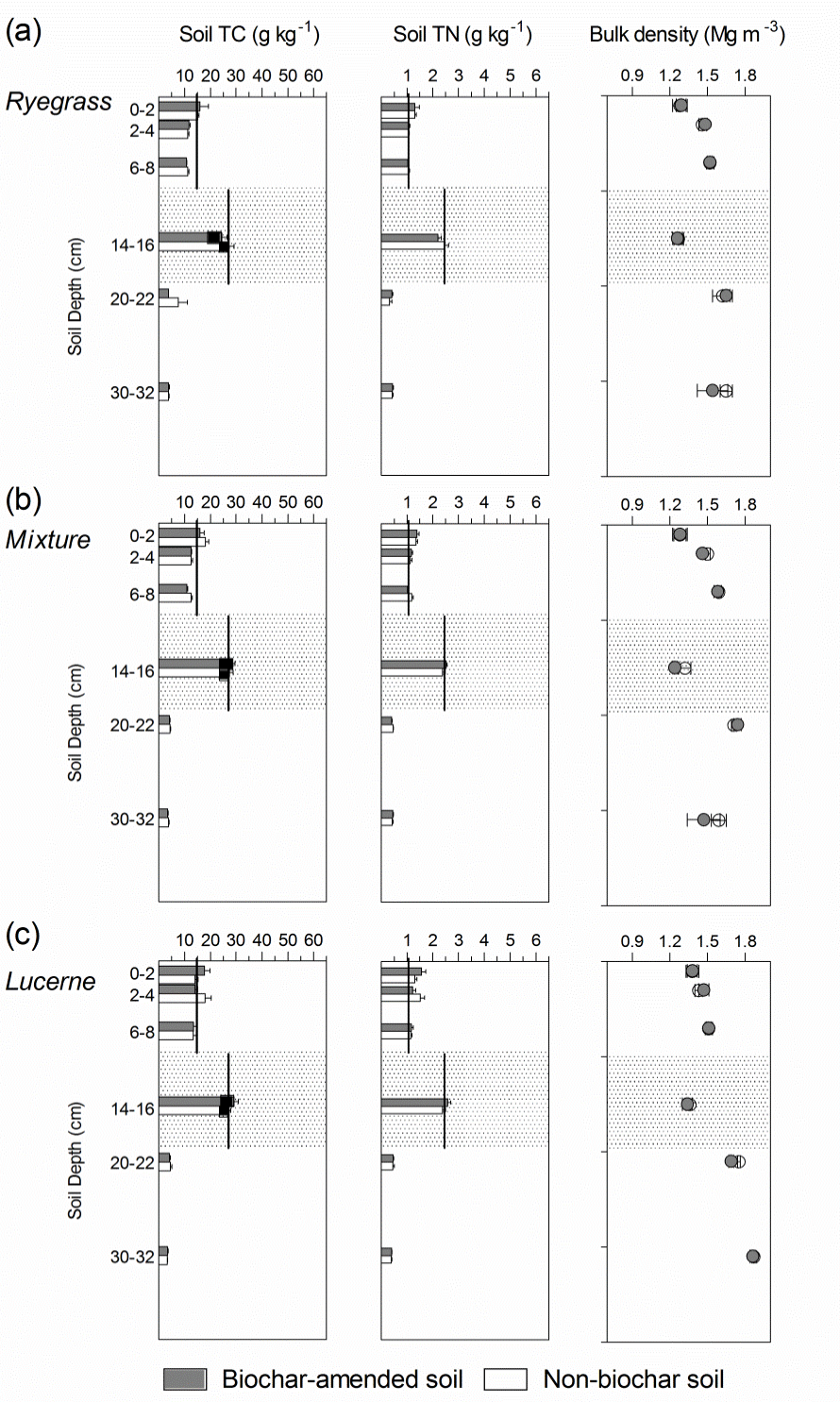

Figure 1.4 Changes in total carbon, total nitrogen and soil bulk density with depth 2 years after "cultivation", biochar addition and growth of different pasture species in lysimeters containing Motuiti sand (experiment 2): (a) ryegrass, (b) mixture (red clover + cocksfoot) and, (c) lucerne. Data represent the average and SEM, standard error of the mean (n = 4). For each depth, bars with different letters indicate significant differences between the biochar-amended soil and the corresponding non-biochar soil at *P* < 0.10. At depth 14-16 cm, the difference (TC – OC_{ox}) is indicated (▉) for each treatment. Vertical bars indicate values of TC and TN for the 0–10 and 10–20 cm depths at the beginning of the experiment (T0) for the non-amended soil, as indicated in Table 1.1.

Again, changes in TN paralleled those of TC, with *r* values > 0.95 in the absence and presence of biochar in the layer where the amendment was deployed. Values of bulk density were negatively correlated with TC (*r* = –0.76 and –0.60, in the absence and presence of

biochar, respectively), and no significant differences were detected between the treatments under study (Figure 1.4).

1.3.3. Changes in TC and TN stocks calculated at a fixed mass

TC and TN stock changes over time (T0, T2)

Silt loam soil with and without pine biochar amendment under different swards (experiment 1)

Values of TC and TN (calculated at an equivalent soil mass corresponding to ca. 0–40 cm soil depth) at T0 and at the end of the trial, T2, are shown in Figure 1.5. In the absence of biochar, TC stocks at T0 were 72.4 Mg C ha^{-1}, with 24, 43 and 33% of it being distributed in the 0–10, 10–20 and 20–40 cm depth, respectively. At T2 and in the absence of biochar, there were losses in TC stocks of 8.1 and 2.0 Mg C ha^{-1}, for the ryegrass and chicory treatments, respectively, which contrasts with the gain of 13.4 Mg C ha^{-1} undergone by the soil under the mixture red clover + cocksfoot. In the presence of biochar, TC stocks at T0 increased to 80.0 Mg C ha^{-1}. At T2, TC stocks in these treatments followed the same pattern as in the absence of this amendment, with a net loss of 4.0 and 6.4 Mg C ha^{-1} for the ryegrass and chicory treatments, respectively, and a net gain of 6.5 Mg C ha^{-1} in the soil under the red clover and cocksfoot mixture.

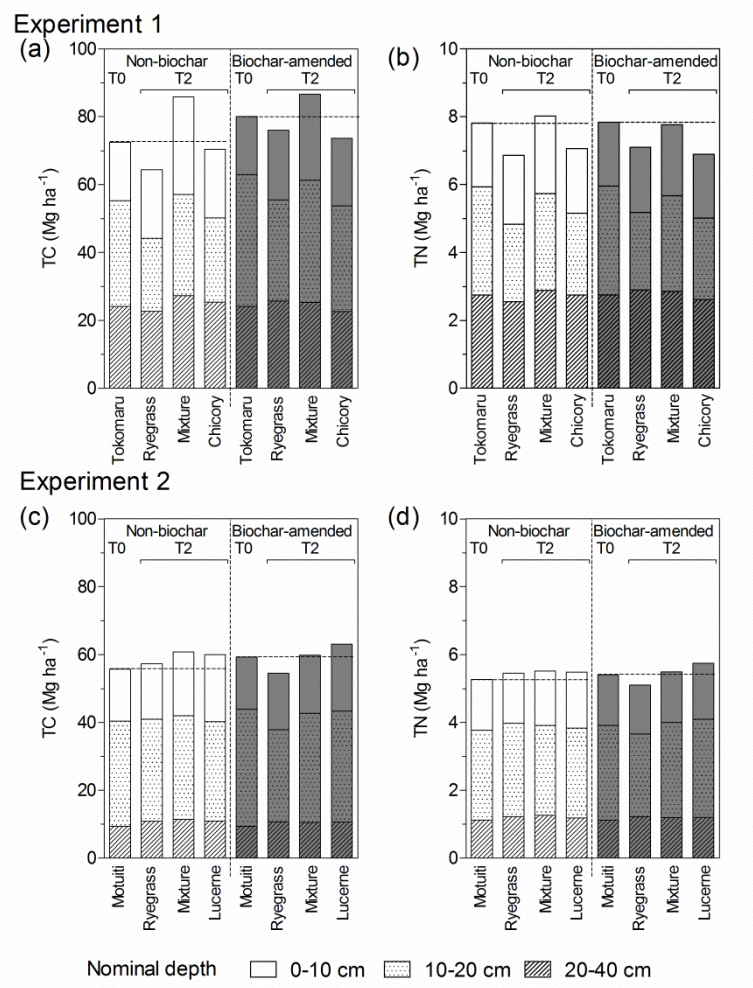

Figure 1.5 Stocks of soil total carbon (TC) and total nitrogen (TN) (nominally, 0–40 cm depth, calculated at an equivalent soil mass) at the beginning of the study (Time 0, T0) and 2 years (T2) after "cultivation", biochar addition and growth of different pasture species in lysimeters containing Tokomaru silt loam (experiment 1): (a), TC, (b), TN; and in lysimeters containing Motuiti sand (experiment 2): (c), TC, (d), TN. Horizontal dashed lines represent stocks at T0 for the corresponding non-biochar soil and biochar-amended soil.

TN stocks followed similar patterns to those of TC stocks, with losses (from 0.9 to 0.8 Mg N ha^{-1}) from soils under ryegrass and chicory type pastures and a minor gain of 0.2 Mg N $ha^{-1}$with the mixture red clover + cocksfoot. As indicated above, this biochar had a low N content and thus barely caused any change in the net TN stocks (Figure 1.5b).

Sandy soil with and without biosolids biochar under different swards (experiment 2)

In the absence of biochar, TC stocks at T0 were 55.7 Mg C ha^{-1}, with 28, 56, and 17% of it being distributed in the 0–10, 10–20 and 20–40 cm depth (Figure 1.5). At T2 none of the treatments without biochar suffered any TC loss, with minor gains ranging from 1.6 to 5.1 Mg C ha^{-1}. In the presence of biochar, TC stocks at T0 increased to 59.3 Mg C ha^{-1}, thus this increase being less accentuated than that in experiment 1, given the lower C content of this ash-rich biochar. At T2 only the soil under the ryegrass treatment showed a TC loss over time (Figure 1.5c). Similar trends were observed in TN stocks (Figure 1.5d).

Effect of biochar on net changes in TC and TN stocks compared to non-biochar treatments at T2

The impact of biochar addition on soil TC and TN stocks with depth was evaluated by comparing the net change after 26 months in the biochar-amended soil compared to the corresponding non-biochar soil (Figure 1.5, Figure 1.6). In the Tokomaru soil (experiment 1, Figure 1.6a,b,c) the addition of biochar resulted in a net gain (1–18% over the non-biochar treatment, $P < 0.10$) under all pastures studied. As expected, the net gain in TC was the greatest in the 10–20 cm layer, to which the biochar was added (equivalent to 7.6 Mg C ha^{-1}). In the Motuiti soil (experiment 2, Figure 1.6d,e,f) the addition of biochar (equivalent to 3.6 Mg C ha^{-1}) resulted in no consistent net TC gain, even in the layer where biochar was amended (*i.e*, at the 10-20 cm depth). Finally, no consistent trend was found for the effect of biochar addition on TN net stock storage change (Figure 1.5, Figure 1.6).

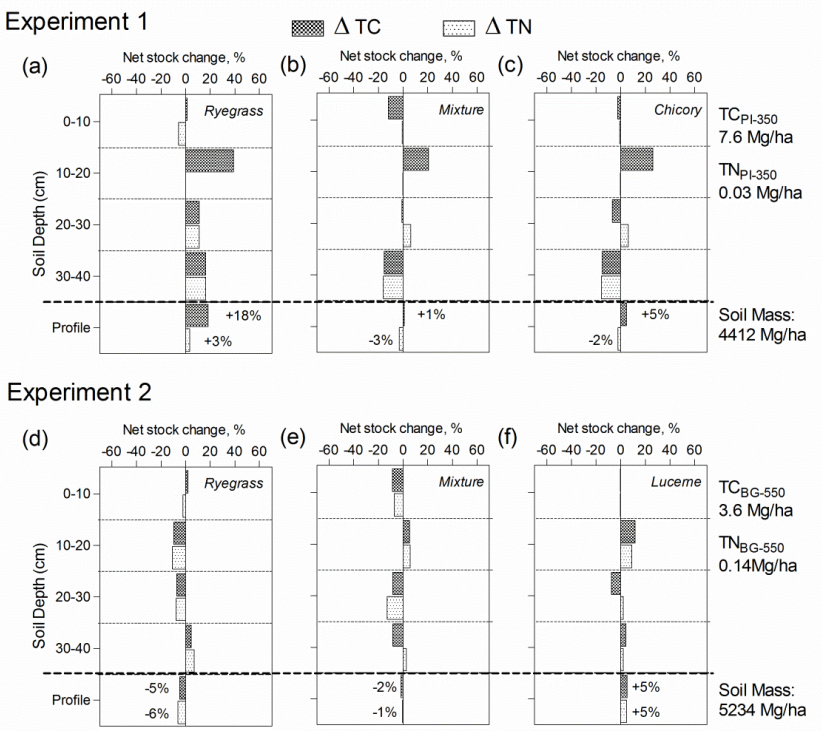

Figure 1.6 Calculated differences in soil total carbon and total nitrogen [ΔTC, ΔTN: ($Stock_{biochar\text{-}amended\ soil}$ – $Stock_{non\text{-}biochar\ soil}$) / $Stock_{non\text{-}biochar\ soil}$, as percentage] with depth 2 years after "cultivation", biochar addition and growth of (a) ryegrass, (b) mixture (red clover + cocksfoot), and (c) chicory, in lysimeters containing Tokomaru silt loam (experiment 1); and growth of (d) ryegrass, (e) mixture, and (f) lucerne, in lysimeters containing Motuiti sand (experiment 2). The actual amounts of TC and TN corresponding to each biochar (PI-350 and BG-550) and soil mass considered in the calculations are also indicated.

1.3.4. Changes in soil pH with depth at the end of the trial

After two years and for all treatments, the first two cm of the Tokomaru soil showed an increase in soil pH compared to T0 (Figure 1.7), whereas the soil pH at deeper depth, except for few exceptions, remained unchanged. In contrast, the Motuiti soil underwent a general acidification process down to 22 cm depth. This effect was to some extent buffered when the BG-550 biochar was added to the Motuiti soil, as pH values of biochar-amended soils were consistently higher ($P < 0.10$) than the corresponding non-biochar soils at 0–10 and 10–20 cm depths at T2 (Figure 1.7e-h).

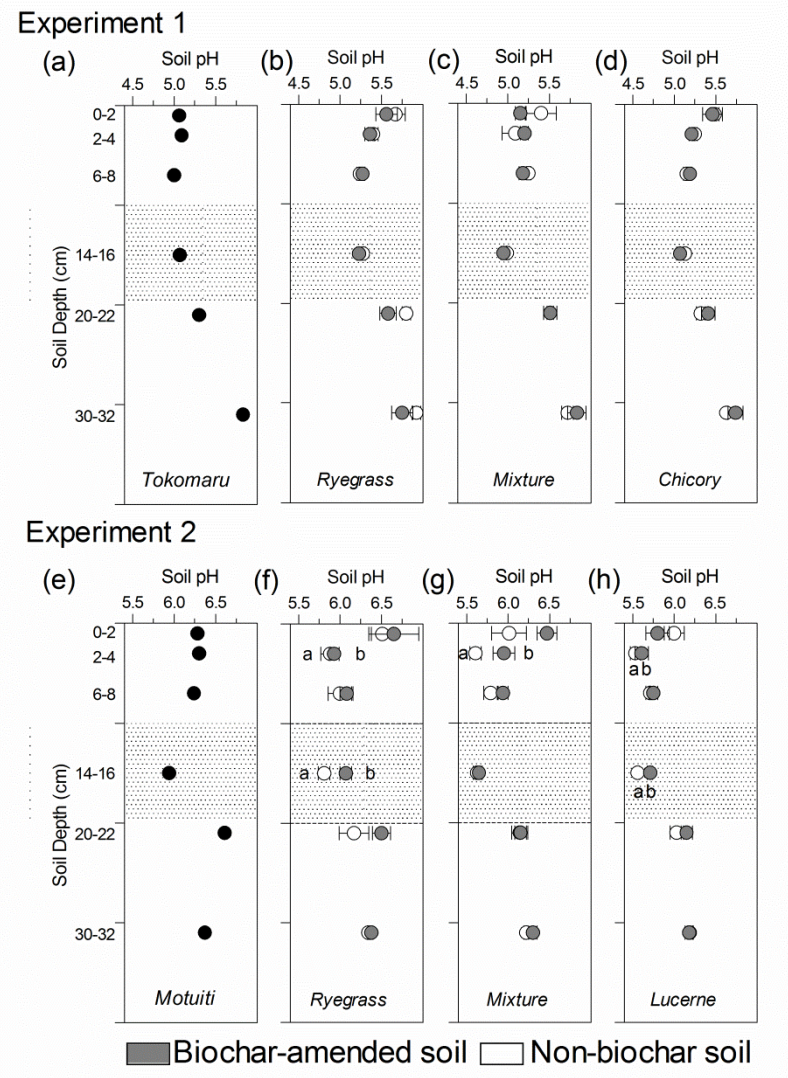

Figure 1.7 Values of soil pH with depth in lysimeters containing: i) Tokomaru silt loam (experiment 1) at (a) time 0, and after 2 years of "cultivation", biochar addition and growth of (b) ryegrass, (c) mixture (red clover + cocksfoot) and (d) chicory; ii) Motuiti sand (experiment 2) at (e) time 0, and after 2 years of "cultivation", biochar addition and growth of (f) ryegrass, (g) mixture (red clover + cocksfoot) and (h) lucerne. Data represent the average and SEM, standard error of the mean (n = 4). For each depth, bars with different letters indicate significant differences between the biochar-amended soil and the corresponding non-biochar soil at *P* < 0.10.

1.4. DISCUSSION

Increasing soil TC stocks by re-establishing pastures on degraded and previous cropland has been well documented (Lal *et al.*, 2011). In regions that have long-established grazed pasture systems and a steady-state has been attained with already high soil TC stocks, it is uncertain whether these stocks can be enhanced and they could even decline under intensive grazing systems (Schipper *et al.*, 2014). As discussed in the *Introduction*, pastures require periodic renewal (cultivating and re-sowing) in order to maintain their productive potential (Tozer *et al.*, 2013). There is concern that, if the cultivation is too frequent, the large amount of pasture residues and soil organic matter (SOM) decomposed after cultivation (Rutledge *et al.*, 2014; Vellinga *et al.*, 2004) will lead to significant soil C loss (Curtin *et al.*, 2010). The aim in this experiment was to investigate whether selection of pasture species (Franzluebbers *et al.*, 2001; Neal *et al.*, 2013) and/or the addition of biochar-C could promote root growth at depth and either maintain or increase soil C stocks.

1.4.1. Change in total C and N stocks – plant species and biochar addition effect

Silt loam soil with and without pine biochar amendment under different swards (experiment 1)

The Tokomaru soil had 15 Mg C ha^{-1} soil (calculated up to a nominal depth of 40 cm) more than the Motuiti soil and yielded a greater above-ground production in the ryegrass and the red clover + cocksfoot treatments, which was as expected given the greater clay content – and additional SOM protection (Six *et al.*, 2002)− and overall soil fertility of the former. However, the Tokomaru soil proved to be less resilient against cultivation than the Motuiti soil, which was *a priori* unexpected and deserves a careful consideration.

The results obtained in this study indicate that the loss of soil organic C undergone by the silt loam soil in the ryegrass and chicory treatments, both in the absence and presence of biochar, occurred mainly in soil layer at the 10–20 cm depth (Figure 1.3). We hypothesise that a considerable fraction of the organic C in this layer – originally present at the 0–10 cm depth – had a fast turnover rate and that, when present at the soil surface, decomposing C is adequately replenished by the C input from growing roots. Once the soil was inverted, these two different swards were not able to replenish the C loss at depth. It was only the mixture treatment, which had a combination of a legume plant with high productivity (Neal *et al.*, 2013) with a long surviving, deep-rooted pasture (Caradus and Evans, 1977), that was able to maintain the C levels in the soil 10–20 cm soil layer. The results also suggest that in all Tokomaru treatments, the repacked top layer, originally from the 10–20 cm depth transferred to the 0-10cm depth, received new C from plant roots − ,which contributed to the increase in TC stocks detected, particulalry in the 0-4 cm layers (Figure1.3) . In fact, in the redclover + cocksfoot treatment it was apparent that it took only 2 years for the new topsoil to reach the organic C levels similar to the original top soil (prior to cultivation). In the ryegrass and the chicory treatments, however, these C gains in the surface horizon were not large enough to balance the TC loss from the whole soil profile. The estimated losses of soil organic C at depth in these two treatments were 31% and 21%, respectively, which is close to the 24% of light particulate soil organic C quantified by Herath *et al.* (2014) in a similar Tokomaru soil under pasture. Nonetheless the contribution of other fractions (e.g., physically-protected SOM) to this loss cannot be disregarded as cultivation affects the stability of macroaggregates (Conant *et al.*, 2007; Rutledge *et al.*, 2014).

The addition of PI-350 biochar to this soil at depth did not contribute to the stabilisation of SOM. Herath *et al.* (2014), also working with a Tokomaru soil but using biochars produced from corn stover at two different temperatures (350 and 550 °C) and growing lucerne, observed the low temperature biochar had a protecting effect on SOM but not the high temperature one. More studies are needed to understand under what circumstances biochar

contributes to SOM protection. Moreover, the PI-350 biochar used in the present study did not contribute to enhance root growth at depth either (Figure 1.2), despite the lowering in soil bulk density attained with this amendment. The incorporation of the PI-350 biochar in the silt loam soil improved aeration in the subsoil, and most probably water discharge (Calvelo Pereira *et al.*, 2015d), but did not directly stimulate root growth at depth, as recently described by other researchers (Prendergast-Miller *et al.*, 2014). In the Tokomaru soil, the main benefit associated with this amendment was thus the addition of a stable C form to the soil, which was able to balance the TC losses associated with pasture establishment in the ryegrass and chicory treatments. However, it should be noted that, based on Vis-NIR reflectance measurements of the initial soil biochar mixtures compared with the soils sampled after 26 months, some of the biochar C was lost by decomposition during these two years (data not shown). This was expected given the low temperature at which this was produced (350 °C) and the considerable fraction of volatile matter (30%) in this biochar.

Sandy soil with and without biosolids biochar under different swards (experiment 2)

The Motuiti soil belongs to a sand dune that was stabilised 500 years ago (Neall, 1977). The TC stock of this soil at the start of the experiment was 56 Mg C ha^{-1} soil (up to a nominal depth of 40 cm), which is a considerable amount for a soil with sand content above 85% (Claridge, 1961; Neall, 1977). The original soil profile had more than 50% of this stock accumulated in the top soil. Once this C-enriched horizon was buried by the simulated cultivation carried out in this experiment, a soil layer with a very low organic C concentration (11 g kg^{-1}; Table 1.1) was exposed at the soil surface in which to establish the new seed bed.

The results obtained indicated that i) in the absence of biochar, no TC losses occurred in the sandy soil under any pasture treatment two years after ploughing, and that ii) differences in TC among pasture treatments were small, although the red clover + cocksfoot mixture was again the one with the greatest gain. In contrast to the Tokomaru soil, the incorporation of C from plant detritus in the Motuiti at the 10–20 cm depth was able to maintain the original TC levels of this soil layer, irrespective of the pasture treatment under consideration. This could be explained as follows: by inverting the soil, a SOM-depleted layer of soil, with a very poor structure and low water retention capacity was placed on top of the soil column, whereas the SOM-rich horizon was placed in the 10–20 cm depth. This horizon had 3 times more C than the newly horizon repacked at the 0–10 cm depth. This resulted in i) a greater retention of water at depth compared to the SOM-poor topsoil, and ii) a greater ability to retain nutrients, which combined probably encouraged the growth of roots at depth. This positive effect associated with the increase in the content of organic C and nutrients in the subsoil has already been reported by others and associated with a positive effect on the subsequent plant growth and the speed of recovery of TC levels (Curtin *et al.*, 2010). The absence of C stock loss during the lysimeter experiment contrasts with the results of Sparling *et al.* (2006), who found that intact cores of a similar Typic Udipsamment, Waitarere sand , lost approximately 23% of the topsoil C during a 4 yr lysimeter experiment. Differences in lysimeter and soil management are obviously important considerations when attempting to compare results been lysimeter experiments, or extrapolate results from lysimeters to the field.

The addition of biochar to the Motuiti soil had a minor effect on TC stocks (compared to non-biochar treatments) (Figure 1.5), which was mainly related to the low amount of C added with this BG-550 biochar (3.6 Mg ha^{-1}). Nonetheless it should be noted that the variability of the data at that depth (Figure 1.4, Figure 1.5; standard deviation for the different treatments ranging from 1.7 to 5.1 Mg ha^{-1}) made difficult the detection of differences. The lack of effect of the P applied with biochar (BG-550) on root growth may be attributed to the fact that fertilizer N and P was applied to replace the net amount of N and P removed that was not put back by the dung and urine applications (Figure 1.1).

Interestingly, the average TC value of the biochar amended soil under ryegrass at the 10–20 cm depth was smaller than that of the corresponding non-biochar treatment (Figure 1.4). A potential explanation for this enhanced loss of C in the presence of this biochar in this specific treatment is given below. The Motuiti soil at the end of the experiment showed, under all pasture treatments, an overall decrease in pH, except in the top 2 cm of soil (Figure 1.2). This acidification was mainly attributed to low pH buffering capacity of this soil against the acidification mainly caused by nitrification of N fertiliser, although legume-based treatments could have further contributed to this acidification (Bolan *et al.*, 1991). The addition of BG-550 biochar buffered to some extent the drop in soil pH, this effect being more apparent in the presence of ryegrass (Figure 1.7). It is possible that the addition of this biochar with liming properties to this SOM-rich soil layer enhanced the decomposition of native organic matter (Whitman *et al.*, 2015). This loss of C in the biochar fertilised layer is discussed again in Part III (Calvelo Pereira et al. 2015c) of this report, with supporting evidence from the field trial. This effect, if occurring in the other two pasture species, was probably mitigated by the important contribution of roots to TC in that soil layer (Figure 1.2). This was not the case of the shallow-rooted ryegrass.

1.4.2. Summary of lysimeter trials main findings

In this study, changes in native organic C of soils after simulated cultivation (with 0–10, C-rich and 10–20 cm, C-poor layers inverted) were strongly related to C allocation by the re-sown pasture root growth. In the silt loam soil, when root development in the 10-20 cm depth was small, as in the case of shallow-rooted species, stocks of native organic C of this buried horizon decreased over time.

In the specific case of the soil under cocksfoot–red clover, native organic C levels of the "new topsoil" reached those of the original top soil layer (prior to cultivation) in less than 2 years.

In the sandy soil no differences between sward types were detected and in all cases soil C stocks at depth were maintained. The exposure of a skeletal and nutrient-depleted soil layer at the surface may have fostered root growth at depth, even that of shallow-rooted species.

The application of biochar at a the rate of 10 Mg ha^{-1} resulted in a net C gain in the silt loam soil where a C-rich biochar was used, whereas no apparent effect was detected in the sandy soil amended with a nutrient-rich biochar. Overall, in either soil, biochar did not have an effect on pasture yield or root growth.

In the sandy soil the inversion of horizons might, *a priori*, represent an attractive option to enhance soil C stocks as long as the low fertility of this newly repacked topsoil is alleviated to ensure yields are not impaired by this management strategy. However, under field conditions the inversion of horizons in sandy soils will be probably hampered by their poor structure and risk of wind erosion during the cultivation and establishment phase. Injection of this biochar to a nutrient-depleted subsurface horizon might then be the only viable option to allocate biochar at depth. In such case, biochar would be added to a low-organic matter and low-nutrient subsurface horizon more prone to respond to biochar addition. More research is needed in this regard, as well as an understanding on the mechanisms through which soil C stocks are preserved at depth.

2. Tokomaru Field Trial

2.1. INTRODUCTION

The wider research goals of this project were: i) to design, manufacture and incorporate customised biochar into pasture subsoils not currently saturated with soil organic C; ii) to promote root growth of a range of pasture species in these subsoils, iii) to accelerate enhanced mineral weathering and soil formation in subsoils in the presence of deeper root systems that will lead to more stabilised C and increase soil C stocks. Calvelo Pereira et al. (2015a ,Part I this report) used simulated cultivation in lysimeters to test the concept that incorporation of C-rich biochar in the lower plough layer (10–20 cm depth) of a heavy textured soil would reduce soil bulk density, enhance root growth of pasture species in the lower profile and result in greater soil C sequestration. The 26 month experiment showed that, soil inversion resulted in a net loss of native organic C in the buried horizon under shallow-rooted species (of 2–7 $Mg/ha_{10\text{-}20\ cm}$), but not under a sward consisting of cocksfoot and red clover, where soil C stocks at that depth were maintained. The addition of a C-rich pine biochar (equivalent to 7.6 Mg C ha^{-1}) resulted in a net C gain (21–40% over the non-biochar treatment) in the buried soil layer under all pastures treatments; this overcame the net loss of native organic C in this horizon under shallow-rooted pastures. However, biochar had no effect on the overall net carbon balance of the soil under cocksfoot/red clover compared to the un-amended treatment. In this section, the results of "proof of concept" lysimeter experiments are tested at the larger scale of field plots to demonstrate to farmers, agricultural industry and policy makers that it is technically feasible to increase permanent C sinks in soils of already developed grazed pasture systems through i) the use of deeper rooting plant species, ii) the incorporation of biochar as a source of stable C, and iii) the potential enhancement of deep root growth in the presence of biochar (e.g., biochar when added in the furrow foot and furrow wall may improve soil drainage in a poorly drained Typic Fragiaqualf by providing connectivity to the macropores of the mole drainage system). This paper reports pasture yield measurements during, and soil carbon and nitrogen stocks measured at the end of, the first two years after seedbed establishment and biochar incorporation.

2.2. MATERIALS AND METHODS

2.2.1. Production of biochar and characterisation

The feedstock was kiln dried pine off-cuts (untreated joinery industry waste), small blocks with maximum thickness 45 mm and width 150 mm. The kiln dried pine waste feedstock (1791 kg at 12.7% moisture) was pyrolysed (in batches of 360 kg) in a static retort (Kilnz Bio-energy; Figure 2.1) to produce 479 kg of biochar over a period of 5 days. The pyrolysis kiln consisted of two 1.0 m^3 stainless steel retorts positioned either side of a central fire box. Exhaust gases from the retorts were piped directly into the central firebox, or vented externally and flared to control the internal retort temperature and prevent incomplete combustion within the fire box. The pyrolysis was conducted with each retort loaded with 180 kg of pine waste. The central fire box fuel consumed 34–36 kg of the dry pined waste as fuel (~10% of feedstock). The maximum pyrolysis temperatures ranged between 350 to 450 °C and were achieved after 3.5 h of heating. After pyrolysis the biochar was stored in 200 L sealed steel drums. The biochar was then roller crushed and screened to pass 15 mm in preparation for application to the soil. The characteristics of the waste pine biochar (KPI-350) are presented in Table 2.1. Briefly, KPI-350 had a relatively low pH (7.4), high C content (851 g kg^{-1}; 75% as fixed C) and low cation exchange capacity (CEC, < 1.2 $cmol_c\ kg^{-1}$).

(a) (b)

Figure 2.1 Kilnz Bio-energy retort filled with 180 kg of pine waste (a) and the charcoal after pyrolyis (b) prior to roller crushing.

Table 2.1 Selected chemical properties of biochar produced from pine pyrolysed at a highest heating temperature of ~350°C.

Variable		Unit	Value
pH			7.4
E.C.		mS/cm	312
Total C		g kg^{-1}	851
Total N		g kg^{-1}	14.2
C/N			60
Ash content		%	2.9
Volatile matter		%	18.6
Fixed C		%	74.6
Moisture		%	3.9
CEC		$cmol_c$ kg^{-1}	1.07
Available cations	Ca^{2+}	$cmol_c$ kg^{-1}	0.49
	K^+		0.46
	Mg^{2+}		0.10
	Na^+		0.06

pH and electrical conductivity (E.C.) determined by the method of Rajkovich *et al.* (2012); Ash content, Volatile matter and Fixed C determined following procedures described in Calvelo Pereira *et al.* (2011); CEC (cation exchange capacity) and available cations determined by extraction with $SrCl_2$ and atomic absorption spectrometry (AAS) (Calvelo Pereira *et al.*, 2014).

2.2.2. Establishment of field trial

The trial site was located at the Massey University, Mogine Sheep Research and Teaching Block (S40°23'22.66", E175°36'39.86") on a Tokomaru silt loam, a Typic Fragiaqualf (Soil Survey Staff, 1999). The trial site was mapped for soil uniformity using an EM38 soil conductivity meter (Geonics Limited, Mississauga, ON, Canada). The initial soil fertility was assessed by sampling four individual points across three transects. The soil conductivity readings were uniform across the site but the soil fertility analysis showed significant spatial variability in Olsen extractable P values from 21 to 67 mg P kg $soil^{-1}$ (Table 2.2). The long-term sheep-grazed pasture was sprayed with glyphosate on 14/3/2011 and mole ploughed at 2.4 m centres to improve drainage to an existing pipe drain. Mole lines were drawn to longitudinally bisect each 2.4 × 10 m plot.

2.2.3. Experimental design, biochar incorporation and pasture establishment

In order to assess the effect of adding biochar at depth to different pastures, a 2 × 3 factorial design was used. Factors included biochar addition (Till, no amendment; and Till-Char, addition of KPI-350 at a rate of ~10 Mg ha^{-1}) and pasture type [three mixed pastures: rye grass + white clover; cocksfoot + red clover; and chicory + white clover]. Treatments were blocked (n = 5) to normalise the effect that phosphate availability could play in the trial, as

indicated by variable Olsen-P values (Table 2.2). The total number of experimental units was 2 × 3 × 5 = 30, with half of the total individual plots including biochar.

Table 2.2 Mean soil test values (n = 4) for the blocks in the trial area. Analytical methods are described in Blakemore *et al.* (1987).

Block	pH	Olsen P (mg P kg^{-1})	SO_4 (mg S kg^{-1})	Exchangeable cations				CEC
				K^+	Ca^{2+}	Mg^{2+}	Na^+	
				($cmol_c$ kg^{-1})				
1	5.3	32	5.0	0.47	4.3	0.86	0.10	14.3
2	5.5	24	4.3	0.26	5.0	0.87	0.11	13.5
3	5.4	27	6.2	0.33	4.7	0.96	0.14	13.0
4	5.4	55	5.3	0.61	4.4	1.08	0.06	13.3
5	5.4	42	7.4	0.53	4.6	1.00	0.14	13.3

The initial cultivation and application of pine waste biochar (KPI-350) was carried out on the 30/3/2011 using a mouldboard plough, which produced furrows 0.25 m deep and 0.40 m wide (Figure 2.2). The biochar was manually placed at the bottom of each furrow prior to burial by the subsequent furrow (Figure 2.1b). At cultivation the single furrow mouldboard plough achieved furrow depths of 0.20–0.24 m. Biochar was applied as uniformly as possible in the furrow bottom over a width of 0.40 m from the furrow wall. This caused biochar to be distributed from depths of 0.24 m at furrow bottom to 0.18 m on the freshly turned furrow. Basal fertiliser of Cropzeal 16N (15.4N:8P:10K:9S) was applied on the 31/3/2011 prior to roto-tilling, rolling and seeding on the 4/4/2011. The plots were drilled with the corresponding mixed pasture species: i) ryegrass (*Lolium perenne* cv Extreme AR1, 20 kg ha^{-1}) and white clover (*Trifolium repens* cv. Bounty, 4 kg ha^{-1}); ii) cocksfoot (*Dactylis glomerata* cv Kara, 4 kg ha^{-1}) and red clover (*Trifolium pratense* cv Sensation R.C., 6 kg ha^{-1}); and iii) chicory (*Cichorium intybus* cv Puna II, 8 kg ha^{-1}) and white Clover (*Trifolium repens* cv. Bounty, 4 kg ha^{-1}).

Figure 2.2 Pine waste biochar being incorporated (10 Mg ha^{-1}) into the Tokomaru trial by hand application to the furrow created by a mouldboard plough, which produced furrows 20 cm deep and 40 cm wide.

2.2.4. Pasture grazing and harvesting

At the end of the first winter period the trial area was rotationally grazed by heavily stocking with sheep. Prior to grazing, pasture production was assessed by mowing a single strip (0.5 m wide × 9.0 m long) in the centre of each plot. The plots were then grazed and plots mown to a uniform 6 cm height to await the next harvest. Each harvest was triggered when pasture cover reached approximately 2500-3000 kg dry matter (DM). Harvest periods on the 10/7/2012 and 12/11/2012 were taken for winter and spring periods, followed by a summer and autumn drought with no growth until winter, allowing harvests on the 24/5/2013 and 19/8/2013. Wet herbage mass was converted to dry matter using subsamples taken from each plot that were weighed wet and then dry, after oven drying at 70°C to constant weight.

Maintenance fertiliser was applied as 400 kg ha^{-1} of 30% potassic superphosphate in June 2011, 200 kg ha^{-1} ammonium sulphate in November 2011, 200 kg ha^{-1} NPKS *Crop 13* in May 2012 and 400 kg ha^{-1} 30% Potassic superphosphate in May 2013.

2.2.5. Soil sampling after year 2

During October 2013 soil cores were taken using a 45 mm diameter percussion corer to a depth of 60 cm from each plot (n = 30). All soil cores were scanned every 1 cm over the range 0.2 – 50 cm, using an ASD fieldspec pro portable reflectance spectrometer using the procedures documented by Kusumo (2009) and Kusumo *et al.* (2010). Subsequently, soil columns were sliced into 5 cm-height sections at the following depths: 0–5, 6–10, 11–15, 16–20, 21–25, and 26–30 cm, and, two 10-cm sections were taken for the 31–40 and 41–50 cm depths. An adjacent uncultivated ryegrass pasture was also sampled at the same time to represent the original uncultivated pasture. Pasture grazing management of the adjacent paddock was similar to that of the experimental plots. Total soil C (and N) concentrations for the soils under ryegrass + white clover (Till and Till-Char treatments), as well as those in the adjacent pasture (referred to as Pasture treatment) were determined as described below. These data were used to calibrate the VISNIR spectra acquired from these samples. The calibration was then used to predict soil C and N concentrations across all plots (Kusumo *et al.*, 2014)

2.2.6. Soil chemical analyses

All soil sections under ryegrass + white clover (Till and Till-Char treatments), as well as those in in comparison with the values obtained from the adjacent pasture (Pasture treatment) were weighed wet and stored at 4 °C until processing. Soil samples were air-dried, then homogenised and particle size reduced to < 250 μm by gentle grinding. A subsample was used for total soil C and N determinations using a vario MACRO cube CHNS elemental analyser (Elementar Analysensysteme GmbH, Hanau, Germany). Further samples were oven dried at 35°C. Bulk density was calculated for each soil sampling considering the volume of the core used and the weight of the soil oven-dried at 105 °C. Soil pH was measured at a soil:water ratio of 1:2.5 (weight/volume) following Blakemore *et al.* (1987). Samples were stirred and allowed to equilibrate overnight prior to the determination of pH in the supernatant.

2.2.7. Calculation of total C and N stocks

There is considerable discussion in the literature (Balesdent *et al.*, 2000; Ellert and Bettany, 1995; Koch and Stockfisch, 2006),on how best to calculate soil C stocks: i) on a volume basis to a fixed sampling depth, ii) on an equivalent mass basis to an approximate fixed sampling depth, or iii) using the weighted mean average volumetric concentration of C in each depth sampled summed to nominal fixed depth.

Firstly, total soil C and N stocks were calculated using the concentration of C and N determined (g C kg soil^{-1}) per sample and the bulk density and thickness of the depth-section sampled. Summing C stocks in each section allowed us to estimate total C and N stocks for the profile up to a fixed depth of 30 cm (the proposed Kyoto Protocol 3.4 sampling depth) and 50 cm (full sampling depth). This corresponds to the C and N stocks at a fixed depth for each soil. The soil mass in the (0–50 cm) fixed depth varied from core to core, partly due to slight errors in soil slice height. Therefore, soil C and N stocks were also calculated for the same equivalent soil mass to 0 to approximately 50 cm depth (Ellert and Bettany, 1995). The same soil mass was achieved in all cores by adjusting the mass of the 41–50 cm slice.

Additionally, in order to accommodate depth sampling artefacts caused by changes in soil bulk density over time, the C masses were recalculated for each soil slice by taking the lowest mass recorded for each slice depth (e.g., 0–5 cm) among all samples. These total C (and N) stocks for each individual slice were used to estimate soil C and N stock changes at each depth and then C and N stocks in each depth were summed to give soil C and N stocks to the full sampling depth.

2.2.8. Statistical analysis

Statistical analyses were conducted with IBM SPSS Statistics version 20 software package (Armonk, New York, USA). An analysis of variance of repeated measurements was used to assess the effect of harvest (time) in the growth rate, as well as the effect of biochar addition and pasture type. Data from soil chemical analyses (bulk density, pH, TC and TN concentrations, TC and TN stocks, and equivalent thickness) were statistically analysed for each soil depth using the ANOVA procedure of SPSS. The model included the fixed effect of the type of amendment [i.e., un-amended soil (Till), soil amended with the biochar made from pine (Till-Char)], and the pasture under normal rotation (Pasture). If a significant ($P < 0.10$) main effect was detected, difference between treatment means was tested using the least significant difference.

2.3. RESULTS

2.3.1. Seasonal changes in pasture growth

Pasture growth rate changed with season, with an overall mean ranging between 8.4 and 57.0 kg DM ha^{-1} d^{-1} (Table 2.3). There was an interaction between the seasonal sward growth rate and the sward type (Table 2.4), with ryegrass + white clover mixture growth rates being faster ($P < 0.05$) for each season except spring late summer (Table 2.3). The addition of pine waste biochar had no significant ($P < 0.05$) positive effect on growth rates (Table 2.3). During the summer of 2012, biochar presence reduced ($P < 0.05$) the ryegrass + white clover growth rates up to 15.4 kg ha^{-1} d^{-1}.

2.3.1. Changes in chemical properties of soils under ryegrass-based pasture after 2 years

Soil bulk density and pH changes

The soil in the adjacent pasture (Pasture treatment; Figure 2.3a) showed a characteristic increase in bulk density with depth (Scotter et al., 1979), from 0.85 Mg m^{-3} in the topsoil to 1.56 Mg m^{-3} at ~45 cm. Cultivated profiles, re-sown to new swards (pooled data as Till and Till-Char) had significantly lower bulk density values than the adjacent pasture at the 16–20 and 21–25 cm depths, with bulk density values < 1.30 Mg m^{-3} (Figure 2.3a). At the scale of soil depth sampling (5 cm thick layers), no significant differences in soil bulk density were

found between un-amended and biochar-amended soil for the ryegrass + white clover sward mixture after 2 years (Figure 2.3a).

Table 2.3 Pasture growth rates (kg ha^{-1} d^{-1}) in the Tokomaru soil for the different pastures and effect of amendment with pine waste biochar produced at ~350°C. Means with different letters denote differences ($P < 0.05$) between pasture types considered. Significant ($P < 0.05$) differences between Nil and Biochar treatments are indicated by *.

Year	2011		2012				2013		
Season	Winter	Spring	Summer	Autumn	Winter	Spring	Summer	Autumn	Winter
Overall mean	13.5	8.4	57.0	38.3	30.7	32.9	ND[1]	31.1	33.6
SEM[2]	0.8	1.3	2.0	2.0	2.3	1.9	ND	1.1	1.1
Pasture effect									
Ryegrass	17a	15.1a	52.8a	38.0a	40.3a	27.8a	ND	36.5a	34.8a
Cocksfoot	11.8b	4.6b	62.7a	34.6a	30.0b	36.3b	ND	31.3b	36.2a
Chicory	11.8b	5.3b	55.6a	42.4a	21.6b	34.6ab	ND	25.6c	29.8ab
Biochar effect (Char – Nil)									
Ryegrass	-4.9	-3.5	-15.4*	6.7	-2.2	-3.4	ND	-1.4	-1.3
Cocksfoot	1.1	1.8*	-6.1	5.5	-5.1	2.2	ND	0.0	3.4
Chicory	-0.1	-0.6	-0.8	1.8	-2.5	-9.3	ND	0.8	0.4

[1] ND, not determined; [2] SEM, standard error of the mean.

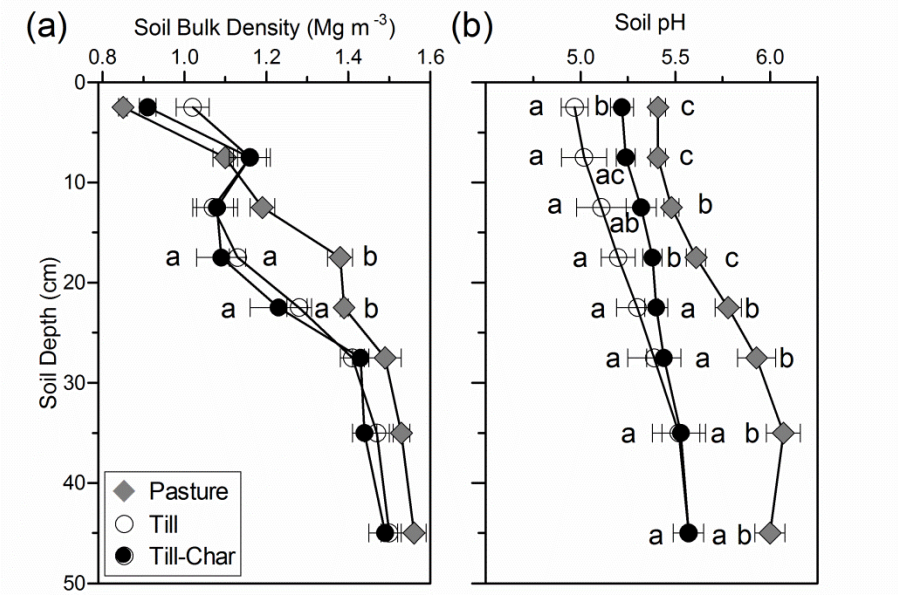

Figure 2.3 Changes in (a) soil bulk density and (b) soil pH over the 0–50 cm soil depth of the Tokomaru silt loam under normal pasture rotation (Pasture), cultivation without amendment (Till) and cultivation with biochar amendment (Till-Char) for ryegrass-based pastures. The bars indicate standard error of the mean. Different letters refer to significant ($P < 0.10$) differences between mean values of bulk density and pH.

The soil under the Pasture treatment (Figure 2.3b) showed a characteristically higher pH than the soils cultivated, either amended or un-amended with biochar (Till and Till-Char). The lowest pH values corresponded always to the Till treatment, whereas biochar addition tended to have intermediate pH values down to around 20 cm depth (Figure 2.3b).

2.3.2. Distribution of total soil C and N in the soil profile

The Pasture treatment showed a distinct stratification of native organic matter, with average values of total soil carbon (TC) and total nitrogen (TN) concentrations decreasing from 44.0 and 4.1 g N kg^{-1} soil in the dense pasture root zone (0–5cm) to 5 and 0.8 g N kg^{-1} soil at the 40–50 cm depth (Figure 2.4). Profiles of cultivated soils, re-sown to new swards of ryegrass and white clover mixture (pooled data as Till and Till-Char) showed higher ($P < 0.05$) TC and TN concentrations than the adjacent permanent pasture at the 11–15, 16–20 and 21–25 cm depths and significantly lower concentrations at the topsoil (0–5 cm depth; Figure 2.4). At 21–25 cm depth, Till-Char showed higher ($P < 0.05$) concentration of C than Till (Figure 11a). Values of TN were similar at this depth (Figure 11b) for both cultivated profiles.

Table 2.4 Repeated measures analysis of variance of pasture growth rate (kg ha^{-1} d^{-1}) data for the different harvests.

Source of variation	Degrees of freedom	Sum of Squares	Mean Square	F-value	P-value
Harvests	4	46786	11133.68	102.73	< 0.001
Harvests × Pasture	8	3885	462.22	4.27	< 0.001
Harvests × Biochar	4	658	156.69	1.45	0.222
Harvests × Pasture × Biochar	8	481	57.27	0.53	0.840
Residual	101	10930	108.38		
Total	126	62740			

Coefficient Greenhouse-Geisser: 0.600

2.3.3. Assessment of net changes in C (and N) storage under ryegrass-based pasture

Figure 2.5 summarises the different approaches chosen to assess changes in TC stocks for the soils studied. A calculation of the soil C mass accumulated to a fixed depth of 30 cm showed that the cultivated soils (pooled data as Till and Till-Char, Figure 2.5a), contained more C (85.0 and 90.0 Mg C $ha^{-1}_{(30\ cm)}$ respectively; $P < 0.05$) than the adjacent uncultivated sward (Pasture; 75.3 Mg C $ha^{-1}_{(30\ cm)}$). The biochar-amended soil (Till-Char) contained 5.0 Mg C $ha^{-1}_{(30\ cm)}$ more than the Till treatment (Figure 2.5a). Therefore, using the equivalent depth calculation, 2 years after the start of the experiment, the net effect of adding biochar on total C was an increase in soil C equivalent to 59% of the added biochar C at time 0 (and to 79% of biochar fixed C at time 0; Figure 2.5). When C masses are accumulated to a fixed depth of 50 cm (Figure 2.5b), a similar result was obtained but the apparent net accumulation of C in the biochar treated soil is only 3.1 Mg C $ha^{-1}_{(50\ cm)}$. When C masses were compared for the soil layer taken at the bottom of the plough layer only (i.e., 21–25 cm, Figure 2.5c) the biochar-amended soil (Till-Char) contained more C mass ($P < 0.05$) than both the un-cultivated adjacent pasture soil (Pasture) and the cultivated soil without biochar (Till). The net carbon gain by adding biochar is an apparent 7.9 Mg C $ha^{-1}_{(21\text{-}25\ cm)}$ (or 93% of C and even above 100% of fixed C originally added with biochar).

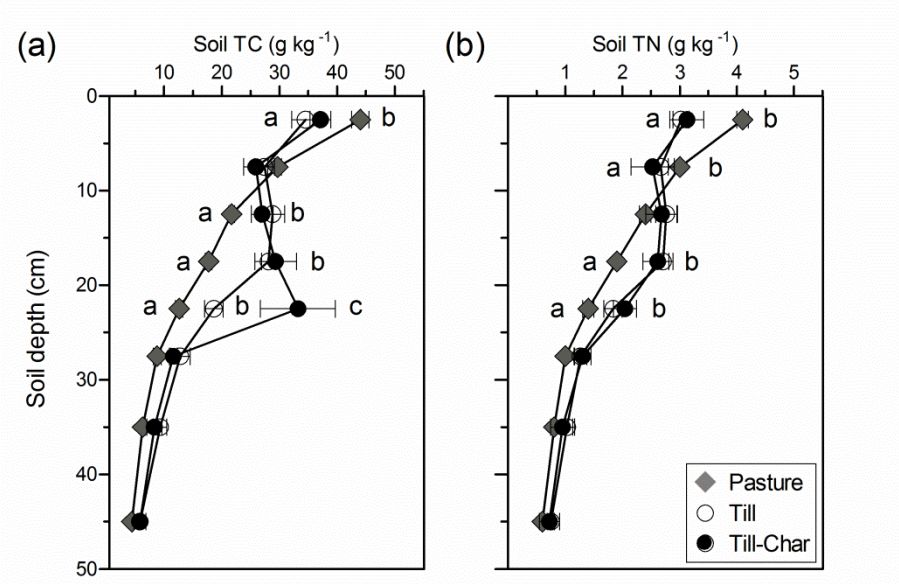

Figure 2.4 Distribution of total C [TC, (a)] and total N [TN, (b)] concentration in the soil profile of the Tokomaru silt loam under normal pasture rotation (Pasture), cultivation without amendment (Till) and cultivation with biochar amendment (Till-Char). The bars indicate standard error of the mean. Different letters refer to significant (*P* < 0.10) differences between mean values of TC and TN.

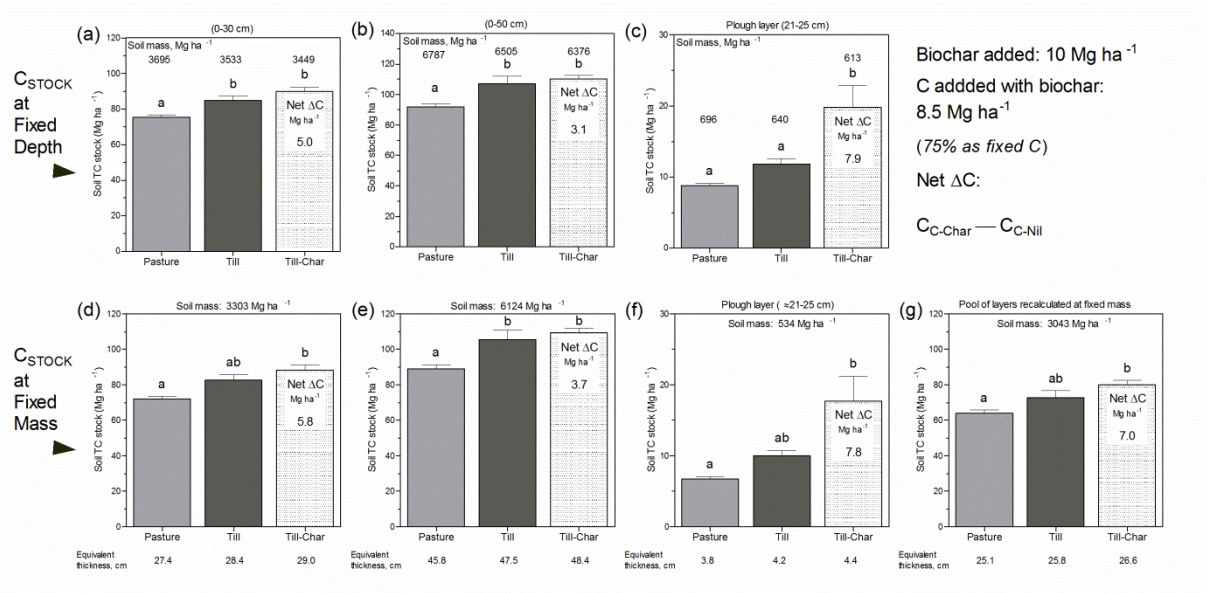

Figure 2.5 Soil total C stocks (TC, Mg ha^{-1}, mean and SEM) for the Tokomaru silt loam under normal pasture rotation (Pasture), cultivation without (Till) and cultivation with biochar amendment (Till-Char) for ryegrass-based pastures. Calculations were done: i) by using a fixed depth approach for the soil profile 0–30 cm (a), for the soil profile 0–50 cm (b), and for the plough layer(c); ii) by using an equivalent or fixed soil mass approach for the soil at ~3300 Mg ha^{-1} of mass (d), for the soil at ~6000 Mg ha^{-1} of mass (e), and for the plough layer (f); and iii) by computing each soil layer individually using an equivalent soil mass and summing up the stocks up to ~3000 Mg ha^{-1} (g). Bars with different letters refer to significant (*P* < 0.05) differences between mean values of TC. Equivalent thickness (cm) of soil layer considered is indicated.

The C masses were recalculated on equivalent soil mass basis, to approximately 30, 50 and 20–25 cm soil depths. The biochar-amended soil (0–30 cm; pooled data Till-Char, Figure 2.5d) contained 88.3 Mg C ha^{-1}$_{(3303\ Mg\ soil)}$ and the uncultivated permanent pasture (NC) soil only 72.0 Mg C ha^{-1}$_{(3303\ Mg\ soil)}$. The biochar-amended soil (Till-Char) contained 5.8 Mg C ha^{-1}$_{(3303\ Mg\ soil)}$ more than the corresponding Till treatment. The net difference between Till-Char and Till treatments was equivalent to 68% of the added biochar C at time 0. When C mass

expressed per equivalent soil mass 6,124 Mg soil accumulated to 50 cm depth were compared (Figure 2.5e), both cultivated treatments (Till and Till-Char) contained more TC (105.3 and 109.1 Mg C $ha^{-1}_{(6124\ Mg\ soil)}$, respectively; $P < 0.05$) than the uncultivated pasture (NC: 88.9 Mg C $ha^{-1}_{(6124\ Mg\ soil)}$). The apparent net accumulation of C in the biochar treated soil is only 3.7 Mg C $ha^{-1}_{(6124\ Mg\ soil)}$. When C masses are compared for the equivalent soil mass at the bottom of the plough layer only (21–25 cm, Figure 2.5f), only the biochar-amended soil (Till-Char) contained more C mass than the un-cultivated soil (Pasture): 17.7 vs. 6.7 Mg C $ha^{-1}_{(534\ Mg\ soil)}$, respectively. The apparent net C gain by adding biochar was 7.8 Mg C $ha^{-1}_{(534\ Mg\ soil)}$ (the amount of biochar fixed C added at time 0 was 7.5 Mg C ha^{-1}).

Finally, the C masses were recalculated on equivalent soil mass basis for each 5 cm depth down to a theoretical 30 cm depth (Figure 2.5g; Table 2.5). The Till-Char soil contained more TC ($P < 0.05$) than the Pasture soil (80.0 Mg C $ha^{-1}_{(3043\ Mg\ soil)}$ vs. 64.1 Mg C $ha^{-1}_{(3043\ Mg\ soil)}$, respectively). However, by following this procedure we found that the Till-Char soil contained 7.0 Mg TC $ha^{-1}_{(3043\ Mg\ soil)}$ more than the Till treatment, a net difference equivalent to 82% of the biochar C (> 100% of the and biochar fixed C) added at time 0 (Figure 2.5g). Regarding net changes in N stocks, the same calculations were done and trends found were similar (Table 2.5).

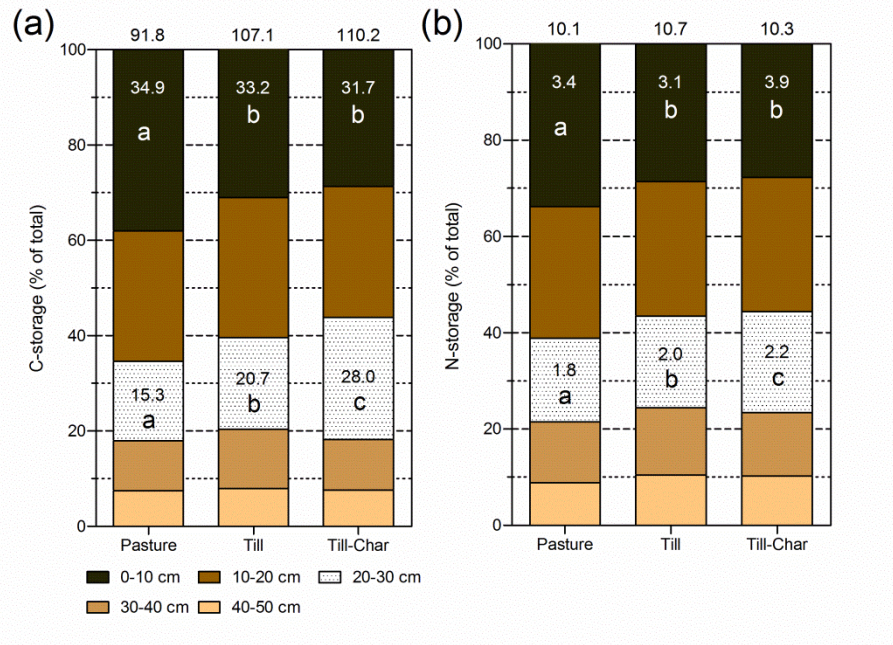

Figure 2.6 Vertical distribution of mean total C [TC, (a)] and total N [TN, (b)] stocks in the soil profile of the Tokomaru silt loam under normal pasture rotation (Pasture), cultivation without (Till) and cultivation with biochar amendment (Till-Char) for ryegrass-based pastures. The numbers in and above the columns give mean C and N-stocks in Mg ha^{-1} calculated at a fixed depth. Bars with different letters refer to significant ($P < 0.10$) differences between mean values for TC and TN percentage distribution.

Table 2.5 Comparison of the thickness of an equivalent soil mass and quantities of TC and TN for each treatment considered: cultivated soil amended with biochar (Till-Char), cultivated soil un-amended (Till) and no cultivated soil under normal rotation (Pasture). Different letters indicate significance at $P < 0.10$ between treatments.

Soil layer (Mg soil ha^{-1})	Treatment	Layer thickness (cm)	TC (Mg ha^{-1})	TN (Mg ha^{-1})
0–413	Till-Char	4.55a	15.32a	1.29a
	Till	4.06a	14.25a	1.25a
	Pasture	4.88b	18.18b	1.69b
413–920	Till-Char	4.39a	13.13a	1.28a
	Till	4.41a	13.90a	1.35a
	Pasture	4.62a	15.05a	1.54b
920–1386	Till-Char	4.35a	12.58a	1.25a
	Till	4.38a	13.43a	1.29a
	Pasture	3.92a	10.13b	1.11a
1386–1841	Till-Char	4.21a	13.32a	1.19a
	Till	4.04a	12.77a	1.23a
	Pasture	3.31b	8.06b	0.88b
1841–2375	Till-Char	4.40a	17.74a	1.09a
	Till	4.19b	9.94b	0.98a
	Pasture	3.84b	6.75b	0.77b
2375–3043	Till-Char	4.69a	7.77a	0.87a
	Till	4.76a	8.53a	0.85a
	Pasture	4.51a	5.89a	0.68b

2.4. DISCUSSION

2.4.1. Pasture growth

Pasture growth was influenced by general weather conditions during the 2-year period, which affected especially plant establishment. Higher growth rates corresponded to moist summer conditions in 2012 and the break of the summer-autumn drought in winter 2013 (Table 2.3). This implied that both cocksfoot and chicory showed lower growth rates than expected due to a slower to establishment when compared with ryegrass. This contrasted with results from a lysimeter experiment using a similar biochar (pine-derived) amending the same silt loam under 10 cm depth under pastures including ryegrass, cocksfoot and chicory Calvelo Pereira et al. (2015a ,Part I this report), where cocksfoot showed a remarkable annual herbage production (with seasonal peaks above 100 kg DM ha^{-1} d^{-1}). However, this study confirms, that, as found by Calvelo Pereira et al. (2015a Part I this report), the inversion of topsoil including biochar amendment has no relevant effect on pastures' growth.

2.4.2. Soil bulk density

The un-cultivated pasture soil showed the characteristic increase in bulk density with depth up to a maximum at the top of the fragipan layer at 45 cm [B_g1 horizon; Scotter *et al.* (1979)] for the Tokomaru silt loam soil type. The reduction in soil bulk density found in the range of 16-25 cm depth was consistent with the mouldboard plough inverting the permanent pasture sod. Biochar had no apparent effect on soil bulk density at the 21-25 cm depth. Previous laboratory- and lysimeter-based studies on the same Tokomaru silt loam soil and using similar biochar (Calvelo Pereira *et al.*, 2015a; Herath *et al.*, 2013) did report significant reduction in soil bulk density with biochar application. In this field experiment it should be noted that the within treatment replicate variability was higher than in the previous studies and is probably

responsible for the lack of significance in bulk density of the biochar treated 16–20 and 20–25 cm depths (Figure 2.3), along with the scale at which soil sampling (0-5 cm soil layers) was carried out. The scale of samples taken and the inherent variability under field conditions are factors that must be considered when extrapolating data from laboratory experiments to field situations.

2.4.3. Calculation of TC and TN stocks

An objective of the present study was to assess changes in soil C stocks attributable to i) cultivation, ii) biochar addition to cultivated soils, and iii) the influence of different pasture species. Protocols for the calculation of soil TC and TN stocks include: i) a sampling depth extended to the subsoil, where organic matter concentrations may remain similar over time and are unlikely to be influenced significantly by treatment; ii) reliable determinations of bulk density profiles; and iii) measuring equivalent soil masses as the basis of assessing short-term net changes between different management options (Balesdent *et al.*, 2000; Ellert and Bettany, 1995; Koch and Stockfisch, 2006). All three preconditions were met in this study. By pooling the C and N masses recalculated on equivalent soil mass basis for each 5 cm depth up to a theoretical 30 cm depth we fully accommodated depth sampling artefacts caused by biochar addition into the subsoil when the paddock was cultivated. Overall, by using the calculation of TC and TN stocks at a fixed soil mass we were able to discount errors caused by the spatial and treatment-induced variance in soil bulk density.

After several years of no cultivation, ploughing (at time 0) caused substantial changes to the distribution of organic C concentrations in the different soil layers (Figure 2.4) which are consistent with the high topsoil C concentrations in the permanent pasture being redistributed to the 11–15, 16–20 and 21–25 cm depths (Figure 2.4). The relatively high values of TC (and TN) stocks found here, higher than the uncultivated soil (Figure 2.5; Table 2.5), indicated that accumulation of C in the period studied (short-term: 2 years) was promoted by soil inversion. We found that the inclusion of biochar (Till-Char) further increased the TC concentration at the 21–25 cm soil depth, consistent with the depth of the furrow bottom and furrow wall. Changes in TN were negligible given that biochar added a very low amount of N (concentration < 15.0 g kg^{-1}; Table 2.1).

In the present study, the addition of pine waste biochar compared to normal cultivation management increased stocks of soil TC over 2 years by 5.0, 5.8 and 7.0 Mg C ha^{-1} for the different assessment approaches used (fixed depth, fixed mass and pooling of fixed masses, respectively; ~0–30 cm depth). Thus, about 16% of the apparent TC increased when calculating following TC by the fixed mass method was due to soil bulk density changes in the profile. When such changes were carefully considered by pooling fixed masses, increases in the apparent TC difference were up to 40%. Accordingly, we support the assessment of C stocks by pooling fixed masses if an accurate accounting of apparent TC (and TN) changes is to be obtained. Despite the lack of general soil bulk density changes between Till and Till-Char treatments, the results suggest that subtle changes in the bulk density of the entire profile (Figure 2.3a) − and not only in the layer of biochar deployment − could influence the assessment of C and N density changes.

2.4.4. Soil inversion and incorporation of biochar C

Despite the fact that measurements considered here did not distinguish aromatic C from other C forms, most of the apparent net gain in C in the Till-Char treatment, even after calculating with different methods, can be attributable to the biochar C, as up to 75% of the biochar C was fixed C with a high degree of aromaticity and hence stability against degradation (Calvelo Pereira *et al.*, 2011; Wang *et al.*, 2013).

In New Zealand pasture soils, most of the C is allocated in the topsoil (Tate *et al.*, 2005). In this study, we showed that this layer could be inverted by deep mould board ploughing and

this not only raised the C and N stocks in the lower profile (Figure 2.4, c.f. 15- 25 cm depths pasture vs Till) but, during the 2-yr post ploughing period , ryegrass + white clover mixture growth began to raise the C and N storage (as % of TC and TN) in the subsoil brought to the surface (Figure 2.4, c.f. 10–15 cm depth *Pasture* vs 0-5 cm depth *Till*) resulting in more accumulation of C and N in the whole profile (Figures 2.4 and 2.5).

Returning to monitoring the TC and TN stocks at this site after longer periods of time (5–10 years) will help to improve the true assessment of net changes of C and N dynamics from deep plowing and biochar addition to temperate pasture soils. It must keep in mind that the probable future outcome of ploughing alone, without biochar management, will be a net C loss (Conant *et al.*, 2007).

2.5. CONCLUSIONS

Biochar was easily incorporated by plough into the furrow foot and furrow wall at the cultivation phase of pasture renovation. The technique can be easily automated and "contained" to avoid handling risks from fine charcoal dust. Major reductions in the bulk density of soil in the lower root zone resulted from tillage compared to leaving the soil in permanent pasture. Incorporation of biochar caused further small reductions in bulk density but these were not significant at the scale of soil sampling (5 cm thick layers). Incorporation of biochar, into the fine textured, poorly drained Typic Fragiaqualf did not significantly increase the growth of ryegrass, cocksfoot and chicory based pastures, a result supported by the earlier lysimeter trials. Soil cores taken two years after deep mouldboard ploughing of the permanent pasture showed that soil C and N stocks to 30 cm had increased by 7-9 % compared to not undertaking the regrassing. The increment in C and N stocks resulted from the inversion of original C rich topsoil to depth and the rebuilding of C in newly established root zone of the resown ryegrass and white clover. Ninety three percent of the stable C added incorporated as biochar remained after two years resulting in a 16% increment in soil C to 30 cm, when compared to other non-biochar tillage treatments.

3. Tangimoana Field Trial

3.1. INTRODUCTION

Sandy soils are characterised by having more than 68% sand and less than 18% clay in the first 100 cm of the soil profile (WRB, 2006). The coarse texture of these soils accounts for their high water infiltration and low water storage capacity. A lack of reactive surfaces gives these soils a low cation exchange capacity (CEC) and poor structure, which partly explains their low capacity to protect organic matter (OM) from microbial decomposition (Baldock and Skjemstad, 2000). Sandy soils dominate the landscape of the west coast of the lower half of the North island in New Zealand (the sand country), covering circa 100,000 ha (Molloy, 1998), mainly dedicated to forestry, but also to intensive pastoral farming where irrigation can be implemented. Land use in these sandy soils is challenged due to wind erosion, drought-prone conditions and low fertility (Cowie *et al.*, 1967).

Application of exogenous OM such as mulches, composts, manures and biosolids is commonly used to ameliorate the soil fertility and physical properties of low-fertility soils of variable texture (da Fonseca *et al.*, 2011; Ryals *et al.*, 2014). Reports on the effects of the application of treated sewage effluent to sandy soils in New Zealand, however, reveal contrasting results on topsoil C in pasture lands with either an increase (Vogeler, 2009) or a decrease (Sparling *et al.*, 2006) in their values several years after treatment. The use of biosolids as a manure for pastures is permitted for dry stock provided that i) there is a 6-month interval between surface application and grazing, and ii) they comply with the thresholds established in the Guidelines for the safe application of biosolids to land in New Zealand (NZWWA, 2003). Moreover, the pelletisation of biosolids (Oleszkiewicz and Mavinic, 2002) offer the potential of obtaining low-cost nutrient sources to lift biomass productivity in these soils where direct local application of sewage sludge is difficult (*i.e.*, farms remote from urban centres). Dried, pelletised biosolids are now produced at least two major centres in New Zealand, being highly transportable and currently compliant with the highest ('Aa') quality grade (NZWWA, 2003). When heavy metal content is low and the presence of pathogens limited, the load of nitrogen becomes the rate limiting factor of the addition of biosolids to land (Gibbs, 2003; Knowles *et al.*, 2011).

The production of biochar from biosolids further decreases the risk of pathogens and that of organic pollutants (Cantrell *et al.*, 2007) and reduces heavy metal availability (Inguanzo *et al.*, 2002) thus providing a safer route for application to grazed pasture and, most importantly, increases C stability, thus decreasing the greenhouse gases emissions associated with these wastes (Lehmann *et al.*, 2006).The incorporation of biochar into soils can potentially modify the physical properties of sandy soils, particularly water retention (Novak *et al.*, 2009; Tammeorg *et al.*, 2014) due to fine pore structure, and also improving cationic nutrients retention over time (Hina *et al.*, 2010; Uchimiya *et al.*, 2012) due to the development of surface negative charge. Proof of concept laboratory and glasshouse trials have demonstrated that nutrient-rich, high-ash biochar produced from biosolids can increase the provision of phosphorus to plants (Wang *et al.*, 2012a). However, some studies have also reported no or even negative effects of biochar on plant growth (Jeffery *et al.*, 2011). In fact, proof of concept trials in lysimeters (Calvelo Pereira *et al.*, 2015a) incorporating in depth a nutrient-rich biochar (made from biosolids) during a simulated ploughing of a sandy soil showed that biochar amendment had little influence on yield or root growth of three different pasture species resulting in no apparent effect on total soil C stocks after 2 years.

This paper evaluates field scale practical solutions for the incorporation of nutrient rich biochar (made from biosolids) into a sandy soil (Typic Udipsamment) with the aim of improving soil C and N stocks. The trial was designed to follow farmer's cultivation

practices, including a pasture renovation using a forage rape crop as the break crop between old and newly sown pasture. This trial compares the agronomic effectiveness of direct application of pelletised biosolids with biochar made from the same biosolids. Net changes in soil carbon and nitrogen storage 2 years after trial initiation were measured and changes in other properties as bulk density are reported here.

3.2. MATERIALS AND METHODS

3.2.1. Production of biochar from granulated biosolids

Biosolids (Grade Aa) recovered after primary sedimentation and then dewatering and pelletising the sludge in a gas-fired rotary granulator, were provided by Hutt Valley Water Services, New Zealand). The granulated biosolids (5–6 mm diameter, 5.1% moisture) were pyrolysed in batches of 200 to 388 kg in a Kilnz Bio-energy static retort (Calvelo Pereira *et al.*, 2015b) to produce 300 kg of biosolids biochar over a period of 3 days.

Table 3.1 Selected chemical properties of biosolids and biochar produced from biosolids at a highest heating temperature of ~650°C.

Element	Unit	Biosolids	Biochar
Total C	g kg^{-1}	450	549
Total N	g kg^{-1}	43.0	52.8
H/C_{org}	At. ratio	-[1]	0.55
C/N	ratio	10.5	10.4
Moisture	%	-	3.2
Ash	%	-	28.1
Volatile matter	%	-	24.7
Fixed C	%	-	44.0
P	g kg^{-1}	10.3	17.6
Available-P	g kg^{-1}	0.95	0.36
S	g kg^{-1}	< 0.01	4.30
Ca	g kg^{-1}	10.0	16.3
Mg	g kg^{-1}	1.65	2.60
Na	g kg^{-1}	0.74	1.20
K	g kg^{-1}	1.28	2.43
Cd	mg kg^{-1}	0.84	2.25
Cr	mg kg^{-1}	23.0	123
Cu	mg kg^{-1}	240	445
Pb	mg kg^{-1}	41.0	76.5
Zn	mg kg^{-1}	550	685

[1] not determined.
Total C, H and N contents were determined using an elemental analyser.
Moisture, ash content, volatile matter content (dry basis) and the stable, thermo resistant fraction or fixed C were determined following (Calvelo Pereira *et al.*, 2011).
Determination of macronutrients (S, Ca, Mg, Na, and K) and micronutrients (Cd, Cr, Cu, Pb, and Zn) by ICP-MS after nitric/hydrochloric acid digestion (EPA, 1999).
Available P was determined by using the 2% formic acid extraction procedure (Wang *et al.*, 2012a) on subsamples not ground.

Each retort was loaded with 142 to 200 kg of granulated biosolids and co-pyrolysed with kiln dried pine wood (27 to 57 kg per each retort) which provided internal venting to the pile of biosolids. The carbonised pine wood was removed after pyrolysis was complete. The central

fire box fuel consumed 51 to 53 kg of the dry pine wood waste as fuel. The maximum heating temperature was achieved after 4 h, ranging between 550–690 °C. The average yield of pyrolysis was 38%. After cooling, the biosolids biochar was stored in 200-L sealed steel drums. The biochar retained the granulated shape of the feedstock and was directly applied to the soil in this form. Main chemical properties of biosolids and biochar used here are reported in Table 3.1. Briefly, the biochar had a moderate ash (28.1 %) and C (549 g kg^{-1}) contents – with an atomic H/C_{org} of 0.55 −, as well a high N content (52.8 g kg^{-1}; Table 3.1). On average, nutrient contents (cations, P) became 1.7-fold concentrated (Table 3.1).

3.2.2. Establishment of field trial and incorporation of biosolids and biochar

The trial site was located at Landcorp Tangimoana, (Harry's Paddock) (S40°18'17.32", E175°16'10.05") on the Hokio–Waitarere association of soils (Cowie *et al.*, 1967); the most common soil type in the area is a Typic Udipsamment (Soil Survey Staff, 1999). The initial soil fertility was assessed by sampling across two transects; the soil analysis showed no significant spatial variability (Table 3.2). The long-term sheep-grazed pasture was sprayed with glyphosate. All plots received the farmer's rate of establishment N-P-K (12.6%-14%-15%) fertiliser at 150 kg ha^{-1}.

The risk of wind erosion of the sandy topsoil at this site prevented the use of full cultivation to establish a forage rape crop. Instead, direct drilling was used; a tine applicator was designed and built to uniformly meter the biosolids and biochar into the trial plots at 15 cm depth at 21 cm centres between zones of application (Figure 3.1; Figure 3.2). During late November 2011 (3 weeks after spraying) the treatments with granulated biosolids were drilled into the plots at two rates of application, 13 Mg ha^{-1} (Bio-H) and 4.5 Mg ha^{-1} (Bio-L) using the custom built tine applicator (Figure 3.2a). The biochar produced from biosolids (Char) was similarly drilled at a rate of 5.0 Mg ha^{-1}. The tine applicator also passed through the control plots (Fert) without addition of materials (Figure 3.2b). The application rates represented total nutrient contents [kg ha^{-1} for nitrogen (N), phosphorus (P) and potassium (K)] of: i) 559N, 130P, 16K; ii) 193N, 45P, 6K; and iii) 265N, 89P, 12K for Bio-H; Bio-L and Char, respectively. The P application rates represent a typical tri-annual pasture maintenance rate for Bio-H and Char, while Bio-L was applied at an annual rate of P. We considered the fact that pyrolysis of 13 Mg biosolids yields approximately 5.0 Mg of biosolids-biochar (*i.e.*, 38%; see above). Appropriate resource consents were obtained by Landcorp farming to permit the biosolids application.

Table 3.2 Main soil chemical characteristics (mean and standard error of the mean – SE; n = 4) for the experimental site.

Property[1]	Units	Mean	*SE*
Ph		5.94	*0.05*
Bulk density	Mg m^{-3}	1.13	*0.02*
Olsen P	mg kg^{-1}	39.2	*3.50*
SO_4-S	mg kg^{-1}	7.89	*0.51*
K	cmol kg^{-1}	0.34	*0.07*
Ca	cmol kg^{-1}	7.14	*0.54*
Mg	cmol kg^{-1}	1.26	*0.15*
Na	cmol kg^{-1}	0.14	*0.01*
CEC	cmol kg^{-1}	11.4	*0.87*
Total Base Saturation	%	77.4	*3.83*

[1] Analytical methods in Blakemore *et al.* (1987).

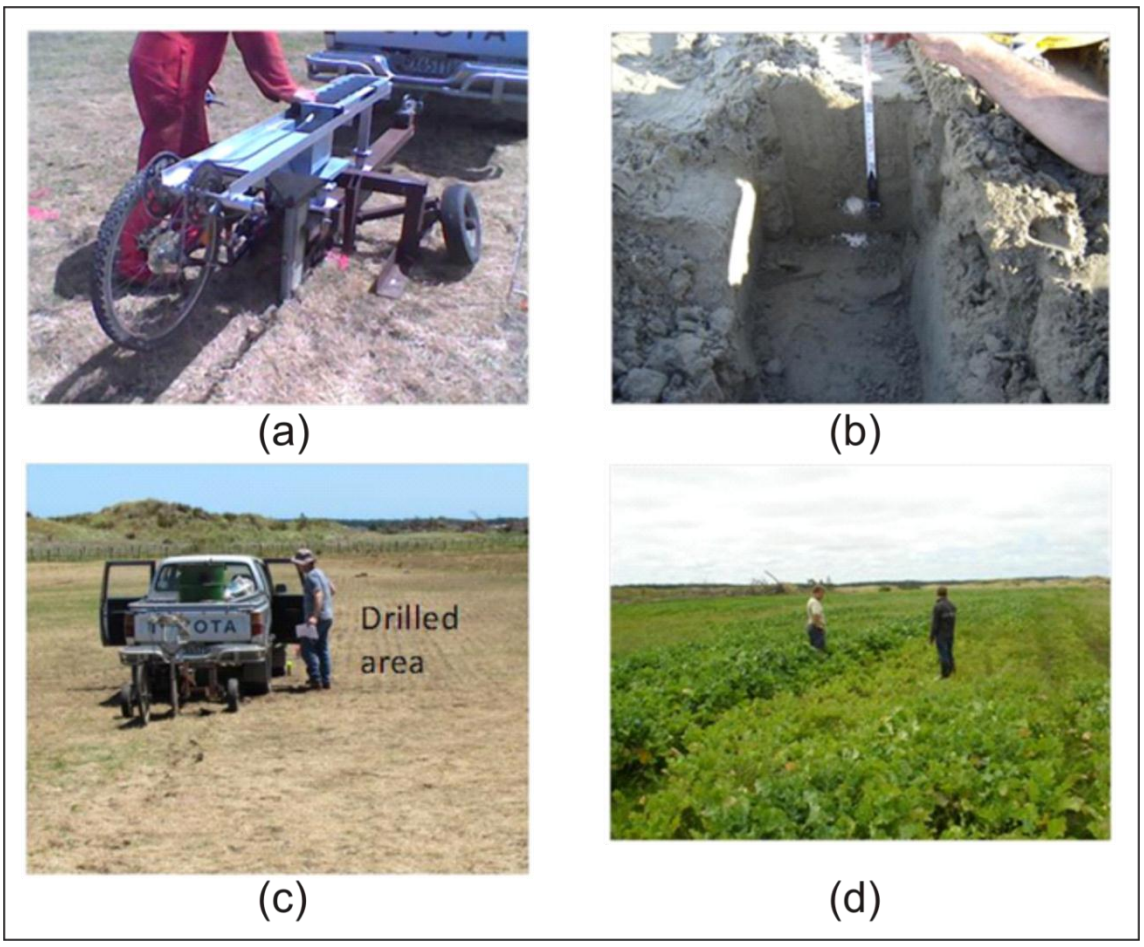

Figure 3.1 a) Tine applicator with ground wheel-driven, sliding tray metering for precise rate application, b) test application of lime chip at 20 cm depth in a recent river sand, c) drilling Hokio sand trial area at 21 cm centres, 15 cm depth, d) Forage rape (Brassica Goliath) Bio-H plot (left person) and Fert only plot (right person) growing in Hokio sand on 20/1/2012.

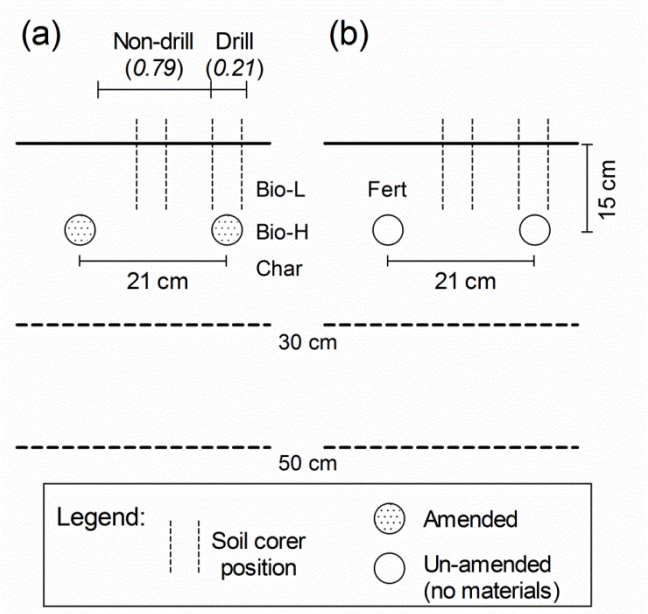

Figure 3.2 Diagram of the soil drilling for (a) amended plots, where either biosolids (Bio-L, Bio-H) or biochar (Char) were incorporated in the subsoil and (b) un-amended plots, where no materials where added (i.e., Fert or control). Approximate location of soil coring is indicated to remark that soil coring was done including the line where the treatments where incorporated. Relative area (ha) for both non-drill and drill zones are also indicated.

3.2.3. Pasture renovation with forage rape: Year 1

The pasture renovation crop was forage rape (*Brassica napus* L. var. Hunter). Rape was over sown by direct drill on December 2011 and yields measured on the March 2012. The trial was grazed with sheep during the second week of April 2012. Wet herbage mass obtained from each harvest was converted to dry matter (DM) using subsamples taken from each plot that were weighed wet and then dry, after oven drying at 70 ^{o}C to constant weight. All herbage samples were analysed for total N and P content (Twine and Williams, 1971). Soil samples were taken down to 30 cm to separate a light-biomass fraction (essentially live and dead roots) following the wet sieving procedure of Kusumo *et al.* (2009).

3.2.4. Pasture seeding: Year 2

The trial area was subsequently sprayed with glyphosate in October 2012 and mowed to remove residual stems from the forage rape crop prior to replanting with the three trial pastures in November 2012. This pasture renovation − that included three different pasture mixtures (ryegrass/white clover, cocksfoot/red clover, and lucerne/white clover) − was unsuccessful due to severe drought conditions during summer 2012-13. All plots were then re-sown with annual ryegrass (*Lolium perenne* L., var Moata) in April 2013 following spraying with glyphosate and broadcast application of DAP fertiliser (100 kg ha^{-1}, including: 20 kg P and 18 kg N ha^{-1}). The ryegrass was harvested following the same procedures as indicated above in September and November to determine DM production and N and P uptake on the November harvest.

3.2.5. Soil sampling after Year 2

In November 2013, all treatments were sampled (n = 4) for the assessment of net changes in total soil C and N. A small trench was excavated in the middle of each plot to locate the drill lines of the biosolids and biochar treatments (Figure 3.2). One set of soil cores were obtained by coring directly through the drill line (soil including the direct drilled biosolids or biochar at 15 cm depth) and a second set by coring between drill lines (non-drill line). A 45-mm diameter percussion corer was used to extract cores to a depth of 60 cm. Areas of high water table (damp soil < 25 cm depth) were avoided. The soil cores were scanned every 1 cm interval to 50 cm depth using an ASD fieldspec pro portable reflectance spectrometer and the method documented by Kusumo (2009) and Kusumo *et al.* (2010). Afterwards, soil cores were sliced in 5 cm-height sections at the following depths: 0–5, 6–10, 11–15, 16–20, 21–25, and 26–30 cm. Additionally, two 10 cm-height sections were taken to include 31-40 and 41-50 cm depth.

Chemical analyses

All soil sections were weighed wet and stored at 4 °C until processing. Soil samples obtained from the drill line were air-dried, then homogenised and particle size reduced to < 250 μm by gentle grinding. Bulk density was calculated for each soil sampling considering the volume of the core used and the weight of the soil oven-dried at 105 °C. Subsamples from the drill line cores were used for total soil C and N determinations using a vario MACRO cube CHNS elemental analyser (Elementar Analysensysteme GmbH, Hanau, Germany). The measured C and N values from the drill line cores were used to calibrate the spectral reflectance from the drill line cores using the spectral processing and partial least squares regression methods described by Kusumo *et al.* (2010). The validation equation was: Vis-NIR predicted (g C kg^{-1}) = Measured (g C kg^{-1}) × 0.95; (r^2 =0.86). This calibration was used to predict soil C and N concentrations in the non-drill line soil core samples. Soil pH was measured at a soil:water

ratio of 1:2.5 (weight/volume) following Blakemore et al. (1987). Samples were stirred and allowed to equilibrate overnight prior to the determination of pH in the supernatant.

3.2.6. Calculation of total C and N stocks

The soil core taken from the drill line (drill lines 21 cm apart) and the core taken from the non-drill line represented 21.4% and 78.6% of the paddock area, respectively (Figure 3.2). These percentages were used to calculate "area weighted" C and N stocks per $ha_{(30\ cm)}$ from the C and N analyses conducted on the drill line and non-drill line core samples. Total soil C and N stocks were calculated three ways after considering the bulk density and thickness of each of the different 5 cm layers to 30 cm depth. Firstly, total C and N stocks for the profile down to 30 cm were calculated by summing masses for all layers sampled. This corresponds to the C (and N) stocks at fixed depth for each soil. Variation in bulk densities around the site caused the weight of soil sampled to range from 4083 to 4854 Mg $ha^{-1}_{(30\ cm)}$. Secondly, the soil C stocks were also calculated for a mean mass of 4400 Mg $ha^{-1}_{(30\ cm)}$ by computationally lengthening, or shortening, the core by altering only the mass in 26–30 cm soil depth, in which the C concentration was the lowest. This produced soil samples from depths ranging from 28.06 to 32.8 cm. Finally, the total C (and N) stocks were calculated by assuming a common mean bulk density at each 5 cm depth sampled from 0–30 cm.

3.2.7. Statistical analysis

Statistical analyses were conducted with IBM SPSS Statistics version 20 software package (Armonk, New York, USA). Data from soil chemical analyses (bulk density, TC and TN concentrations, TC and TN stocks, and equivalent thickness) were statistically analysed, for each soil depth considered individually, using the ANOVA procedure of SPSS. The model included the fixed effect of the type of amendment [i.e., un-amended soil (Fert), soil amended with a low dose of biosolids (Bio-L), soil amended with a high dose of biosolids (Bio-H) and soil amended with biochar made from the same biosolids (Char)]. If a significant ($P < 0.10$) main effect was detected, differences between treatment means were tested using the least significant difference.

3.3. RESULTS

3.3.1. Summary of forage crop and annual ryegrass yields, nitrogen and phosphorus uptake

Forage rape yield from plots treated with fresh biosolids at either application rate were significantly higher ($P < 0.05$) compared to that under Fert treatment (Table 3.3), whereas there were no significant differences ($P < 0.05$) between the Fert and Char treatments, despite the second having a higher mean value (8.8 vs. 6.6 Mg ha^{-1}; Table 3.3). The highest forage rape yield (16 Mg DM ha^{-1}) corresponded to the Bio-H treatment (Table 3.3). The live and dead root masses in the top 30 cm − measured at the final grazing of the rape − ranged between 20 and 48 Mg DM ha^{-1} and were strongly negatively related (r^2 =0.84) to the forage rape yield and plant N and P uptake (N uptake shown in Figure 3.3).

The highest accumulated yield from the two spring harvests of the annual ryegrass grew in the Bio-H (3.7 Mg DM ha^{-1}) and Bio-L (3.6 Mg DM ha^{-1}) plots (Table 3.3); the Char plot yield (3.2 Mg DM ha^{-1}) being lower and not significantly different from the farmer applied fertiliser (Fert, 3.1 Mg DM ha^{-1}). The combined crop growth removed 160, 192, 290 and 572 kg N ha^{-1} (31, 41, 61 and 71 kg P ha^{-1}) from the Fert, Char, Bio-L and Bio-H plots, respectively. This represented an apparent N (P) recovery from the total N and P applied as Char, Bio-L and Bio-H treatments of 12, 68 and 74 %, (10, 65 and 30 %) respectively.

Table 3.3 Yields and live and dead root mass at harvest of forage rape (Stage 1 of pasture renovation) and the sum of spring and early summer yields of annual ryegrass. Their respective N and P uptake is then expressed as an apparent percentage up take of N and P applied as biosolids and biochar (mean and standard error of the mean – SEM; n ≥ 8).

	Fert		Char		Bio-L		Bio-H	
	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
Forage Rape								
Yield (Mg/ha)	6.6	1.3	8.8	2.0	14.3	1.3	16.2	1.5
N uptake (kg/ha)	94	8	130	23	213	40	501	63
P uptake (kg/ha)	19.4	4.6	27.3	8.5	45.4	4.8	54.4	6.8
Live and Dead root biomass (Mg/ha)	47.9	6.7	33.8	3.5	22.7	8.3	20.2	2
Annual Ryegrass								
Sum yield (Mg/ha)	3.1	0.2	3.2	0.1	3.6	0.2	3.7	0.1
N uptake (kg/ha)	65.2	4.3	61.9	4.3	77.2	4.4	70.7	3.1
P uptake (kg/ha)	12.1	0.5	13.4	0.9	15.5	1.0	16.1	1.0
Apparent recovery of N (% added)			12		68		74	
Apparent recovery of P (% added)			10		65		30	

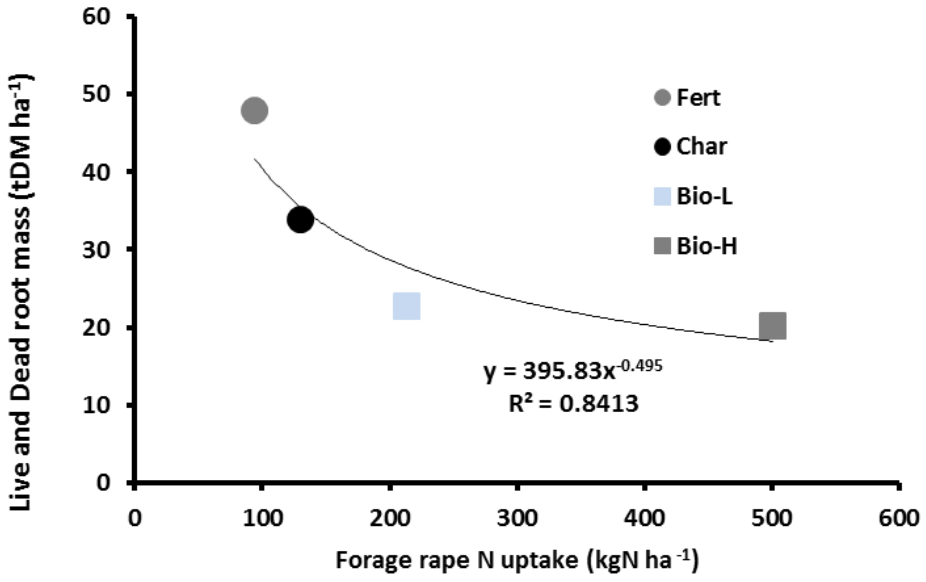

Figure 3.3 The relationship between forage rape N uptake and live and dead root biomas at harvest before the final grazing.

3.3.2. Soil bulk density and pH after 2 years

The soil under the farmer's fertiliser application programme (Fert) showed a characteristic high bulk density (Figure 3.4a) that increased continuously with depth, from 1.11 Mg m^{-3} in the topsoil to a maximum of 1.72 Mg m^{-3} at ~22.5 cm; bulk density was slightly reduced down to 1.58 Mg m^{-3} at ~45 cm. The different treatments showed similar profiles for soil bulk density (Figure 3.4a). However, at the drilling depth (~16–20 cm), both biosolids (at the low

dose only) and biochar treatments had lower ($P < 0.05$) soil bulk density (< 1.55 Mg m^{-3}; Figure 3.4a) compared to the Bio-H and Fert treatments, there being no significant differences between the Bio-L and Char treatments.

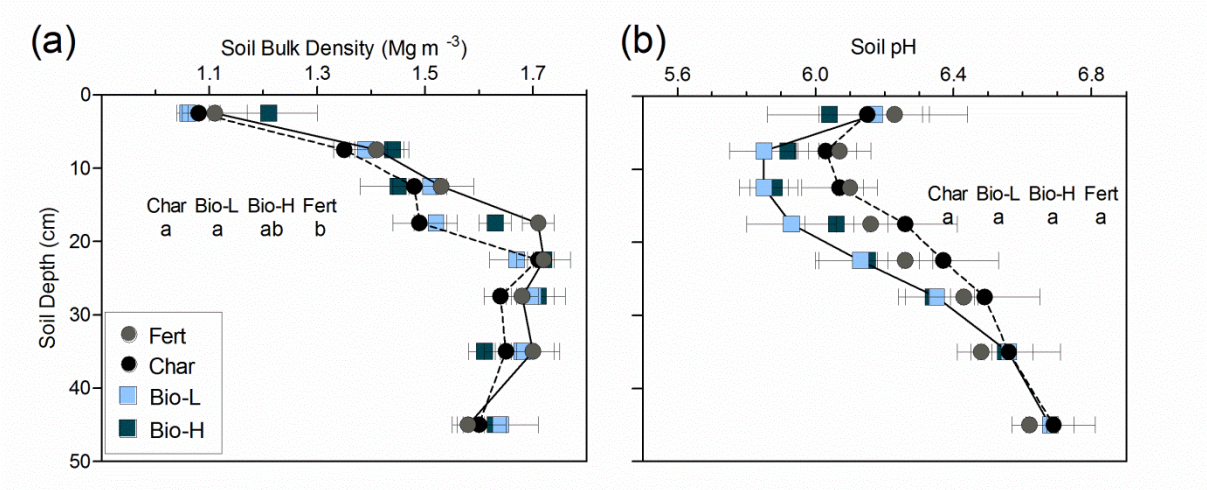

Figure 3.4 Change in (a) bulk density and (b) pH of soil sampled from the drill lines in Hokio sand under ryegrass. Cores were taken two years after the application of biosolids at two rates (Bio-L, 4.5 Mg ha^{-1}: Bio-H, 13 Mg ha^{-1}), biochar produced from the same biosolids at ~650°C (Char, rate of 5.0 Mg ha^{-1}) and basal fertilisation only (Fert or Control). The bars indicate standard error of the mean (SEM). Different letters refer to significant ($P < 0.05$) differences between mean values of soil bulk density and soil pH at the layer where treatments were drilled.

Two years after the start of the experiment, no major differences in soil pH at the 0-10cm depth were found between any of the treatments for the soil under the newly sown annual ryegrass sward (Figure 3.4b). The fertiliser only and biochar amended soil had the highest pH values (always > pH 6.0; Figure 3.4b). Biosolids addition reduced soil pH below 6.0 at 6–10, 11–15 and 15–20 cm depths.

3.3.3. Total soil C and N distribution; changes in C/N ratio

In the soil under normal fertiliser application (Fert) average values of total soil carbon (TC) and total nitrogen (TN) concentrations decreased with depth from 47.3 g C kg^{-1} soil and 4.5 g N kg^{-1} soil in the dense pasture root zone (0–5cm) to 4.5 g C kg^{-1} soil and 0.47 g N kg^{-1} soil at the 40–50 cm depth (Figure 3.5a). At 16–20 cm depth, addition of biosolids-biochar (Char) and biosolids at the low dose (Bio-L; Figure 3.5) increased ($P < 0.10$) soil TC and TN concentrations. At 16–20 cm depth, the soil of the Char treatment showed higher ($P < 0.10$) C/N ratio (average value of 9.3) than the rest of the treatments (data not shown).

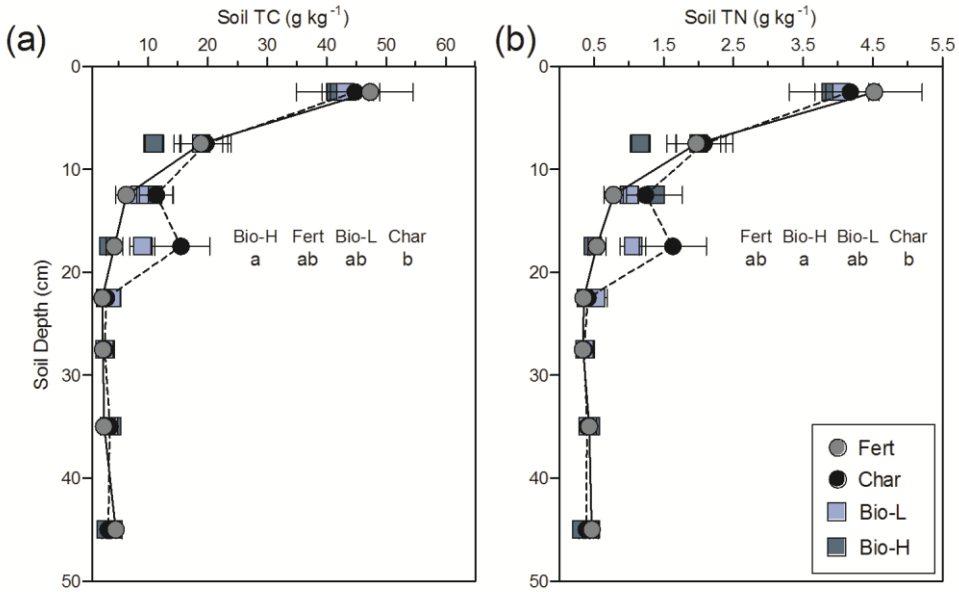

Figure 3.5 Distribution of total C [TC, (a)] and total N [TN, (b)] concentration of soil sampled from the drill lines in Hokio sand under ryegrass. Cores were taken two years after the application of biosolids at two rates (Bio-L, 4.5 Mg ha^{-1}: Bio-H, 13 Mg ha^{-1}), biochar produced from the same biosolids at ~650°C (Char, rate of 5.0 Mg ha^{-1}) and basal fertilisation only (Fert or Control). The bars indicate standard error of the mean (SEM). Different letters refer to significant ($P < 0.10$) differences between mean values of soil bulk density at the layer where treatments were drilled.

3.3.4. Assessment of net changes in C and N storage

TC stocks

The soil carbon mass accumulated to a fixed depth of 30 cm (incorporating the representative areas of drill line and non-drill line sample cores) were 51.5, 64.4, 61 and 69.6 Mg C ha^{-1}$_{(30\ cm)}$ for Bio-H, Bio-L, Char and Fert, respectively (Table 3.4). Amounts calculated by the common soil mass to 30 cm gave similar amounts (< 5% variance) and similar trends between treatments, whereas those calculated using the common bulk density per slice method gave considerable higher values for the Char treatment (67.1 Mg C ha^{-1}; Table 3.4).The Bio-H treatment had significantly ($P < 0.05$) lower TC at all depths than the Bio-L, Char and Fert treatments, which all had similar TC stocks. The Bio-H treatment had lower C contents at all depths down to 20 cm, those at the 10–15 and 15–20 cm depths being markedly lower than other treatments (Figure 3.6).

Table 3.4 Total carbon and nitrogen stocks (mean and standard error of the mean – SE; n = 4) to 30 cm depth 2 years after treatment application and immediately following a rotation of fodder rape and annual ryegrass.

	Fert		Char		Bio-L		Bio-H	
Method	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
Soil Carbon Stocks				Mg C ha^{-1}				
Volume basis (0-30cm)	69.6	6.8	61.0	2.8	64.4	2.0	51.5	3.2
Common wt. to depth (4400 Mg soil/ha)	69.5	7.1	61.1	2.8	64.2	1.9	51.0	3.5
Common bulk density/slice	72.9	8.7	67.1	1.3	63.8	2.0	52.9	6.1
Soil Nitrogen Stocks				Mg N ha^{-1}				
Volume basis (0-30cm)	7.1	0.6	6.2	0.3	6.5	0.2	5.6	0.3

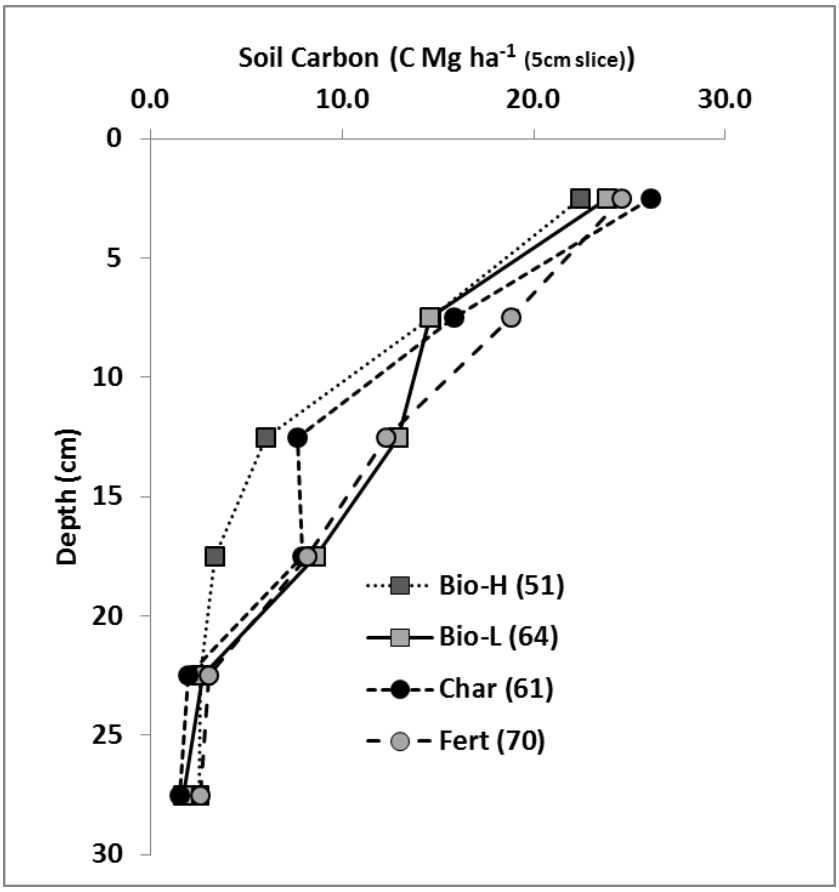

Figure 3.6 Soil total C stocks (TC, Mg ha^{-1},) for the Hokio sand under ryegrass. Cores were taken two years after the application of biosolids at two rates (Bio-L, 4.5 Mg ha^{-1}: Bio-H, 13 Mg ha^{-1}), biochar produced from the same biosolids at ~650°C (Char, rate of 5.0 Mg ha^{-1}) and basal fertilisation only (Fert or Control). Each depth value is calculated from the bulk density and the VIS/NIR prediction of the C concentration in each 5cm soil slice taken from four replicate drill line and a non-drill line cores, with the drill and and non drill line cores representing 0.214 and 0.786 ha, respectively.

TN stocks

The soil N stocks accumulated to a fixed depth of 30 cm (incorporating the representative areas of drill line and non-drill line sample cores) were 5.6, 6.5, 6.2, and 7.1 Mg N $ha^{-1}_{(50\ cm)}$ for Bio-H, Bio-L, Char and Fert, respectively (Table 3.4). Variation between replicates was sufficient to make these differences non-significant ($P > 0.05$).The Bio-H treatment had lower TN at the 15–20 cm depth than the Bio-L, Char and Fert treatments, which all had similar TN stocks (Figure 3.7).

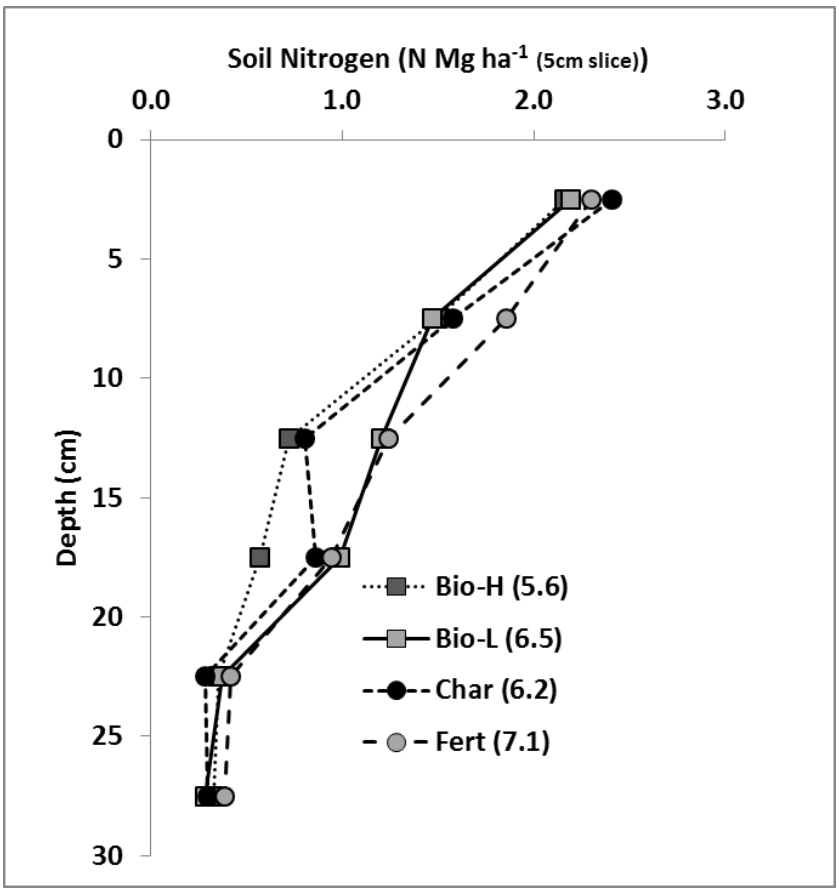

Figure 3.7 Soil total N stocks (TN, Mg ha^{-1}) in the Hokio sand under ryegrass. Cores were taken two years after the application of biosolids at two rates (Bio-L, 4.5 Mg ha^{-1}: Bio-H, 13 Mg ha^{-1}), biochar produced from the same biosolids at ~650°C (Char, rate of 5.0 Mg ha^{-1}) and basal fertilisation only (Fert). Each depth value is calculated from the bulk density and the VIS/NIR prediction of the C concentration in each 5cm soil slice taken from four replicate drill line and a non-drill line cores, with the drill and and non-drill line cores representing 0.214 and 0.786 ha, respectively.

3.4. DISCUSSION

Small differences in elevation through the paddock caused the topsoil to change from poorly (with higher organic matter content, *e.g.* 65 g C kg^{-1}) to excessively drained (with lower organic matter content, *e.g.* 34 g C kg^{-1}) creating the large variation in total C (at 0–5 cm, Figure 3.5a). Whilst these differences were accommodated in the trial design by blocking replicate treatments, the differences in drainage status had a major impact on success, or failure, of establishing the forage rape and subsequent annual ryegrass. Previous studies in this area on similar soils (Wallis *et al.*, 1993) reported the existence of a considerable spatial variability in water repellence associated with quality and quantity of topsoil organic matter content.

The forage crop and annual ryegrass yields (Table 3.3) were positively related to the amount of available N recovered by both crops. Biosolids, at high and low application rates were a source of readily-available N, which was relatively efficiently recovered by the crop and pasture (68% and 74% for Bio-L and Bio-H treatments, respectively; as shown in Table 3.3 and supported by other research (Kowaljow *et al.*, 2010). Barton *et al.* (2005) had also shown a high level of N recovery from pelletised biosolids used to fertilise irrigated turf grass in Western Australia grown on a Typic Xeropsamment (mean annual rainfall of 859 mm and annual temperature of 18.7 °C and pH 4.8 $_{CaCl_2}$). This is in contrast to work in Canada, where Kelty *et al.* (2004) found that only 26% of the N in the pelletized biosolids was mineralised in

the first year after application to a forest soil (Typic Dystrochrept, with mean annual rainfall of 1140 mm annual temperature 10 °C and pH 4.). In contrast, most of the N in the biochar manufactured from the same biosolids was less available in the short term for plant growth (Char, Table 3.3), which is consistent with the N being locked within the condensed aromatic structures of the biochar as shown by Wang *et al.* (2012b).

Unlike the crop and pasture yields, the amounts of live and dead root mass measured after grazing the forage rape were inversely related to plant available N (Figure 3.3). This reduction in root mass with increased soil fertility is consistent with results of pasture root measurements reviewed by Dodd *et al.* (2011). Comfort *et al.* (1988) and Wang *et al.* (2014) have reported that higher rates of N fertilisation leads to lower root densities in cereals. For recently resown pastures that were 5 months old, however, Moir *et al.* (2013) have shown that root growth of 13 different pasture species was directly correlated to shoot growth and N uptake from urine treated soil. The response of root growth to nutrient availability and environmental variables is obviously complex and extrapolating experimental results requires caution. In our field experiment the root mass, which included live and dead material, decreased as plant available N and P increased with higher additions of biosolids. This decrease took place in the first 7 months of the experiment and was particularly marked in the Bio-H treatment (Figure 3.3).

The decrease in live and dead root mass detected 7 months after the start of the experiment in the treatments fertilised with higher N rates (Bio-H and Bio-L) compared with the conventionally fertilised treatment is consistent with the corresponding lower soil C stocks measured after 2 years (Table 3.4). This positive relationship between the live and dead root mass (measured 7 months after treatment application) and the soil carbon stocks to 30 cm (measured after two years) is shown in Figure 7 when considering all treatments. The mass of the C stock to 30 cm, however, is negatively related to the accumulated plant N uptake (a measure of N availability during the experiment; Figure 7). Decreases in soil C attributed to markedly increased N availability have been noted both in studies with inorganic fertilisers, as reviewed by Lu *et al.* (2011), and with sewage effluent (da Fonseca *et al.*, 2011; Egiarte *et al.*, 2005). After two years of sewage effluent application to 4 contrasting soils in lysimeters, Barton *et al.* (2005) reported increased mineralisation of soil organic matter with considerable release of N from a Typic Udipsamment, which they attributed to a "priming effect " caused by the application of the N rich sewage effluent. After 4 years, at the conclusion of the experiment, Sparling *et al.* (2006) reported a large (47%) reduction in organic C in the A horizon of the Typic Udipsamment caused by effluent irrigation. In this experiment we observed a 26% reduction in $TC_{(0\text{-}30cm)}$ between the soil treated with fertiliser alone (Fert) and the soil treated with high biosolids (Bio-H, Table 3.4) and a narrowing of the C:N ratio of soil organic matter from 9.7–9.9 in the Fert, Biochar and Bio-L treated soils to 9.1 in the Bio-H treated soil. The increased mineralisation of soil organic matter N plus the added treatment N also is consistent with the lower soil pH measured in the Bio-H soil profile (Figure 3.4) presumably caused by nitrification followed by nitrate leaching as reported in other studies (Egiarte *et al.*, 2005; Egiarte *et al.*, 2006). As with the experimental work of da Fonseca *et al.* (2011), in which the application of the highest effluent N treatment of 520 kg N ha^{-1} y^{-1} caused a loss of soil C to occur very rapidly within 6-7 months, the decrease in live and dead root mass compared with the treatment under conventional fertilisation was evident in our trial at the final harvest of the forage rape, which was 7 months after the biosolids were drilled in. A consistent finding from our study and the studies reviewed by Lu *et al.* (2011) and those involving sewage effluent (da Fonseca *et al.*, 2011) is the addition of more available N (within limits) leads to the highest above ground biomass, often without an increase in root mass. Biomass richer in N does not increase the pool of slowly decomposing residues and therefore as found by Barton *et al.* (2005) and Sparling *et al.* (2006) leads to a reduction in soil N and C stocks.

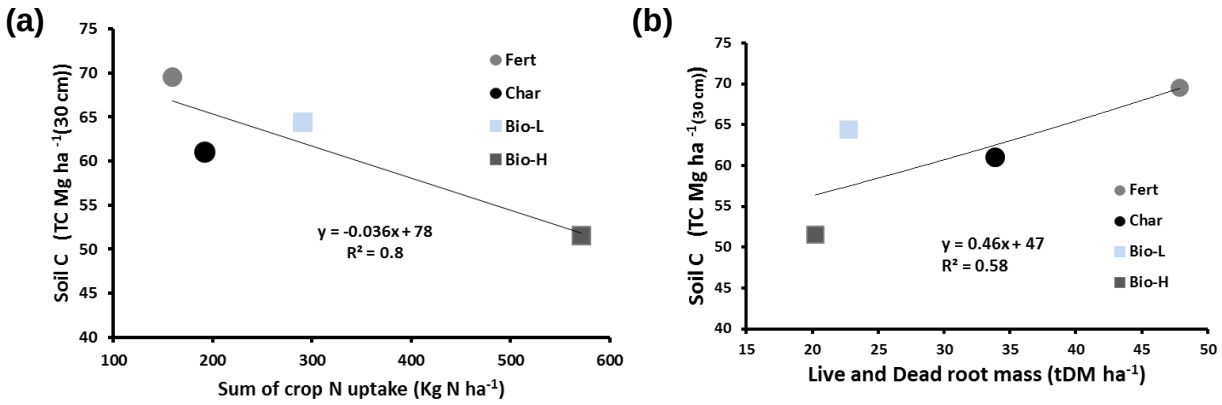

Figure 7 The relationship between (a) the accumulated uptake of N by the forage rape and annual ryegrass and (b) the live and dead root mass 7 months after treatment application and the soil carbon stocks to 30 cm measured two years after the application of biosolids at two rates (Bio-L, 4.5 Mg ha^{-1}: Bio-H, 13 Mg ha^{-1}), biochar produced from the same biosolids at ~650°C (Char, rate of 5.0 Mg ha^{-1}) and basal fertilisation only (Fert).

There were large net losses of C (ranging from 5–20 Mg C ha^{-1}) in the organic amendment treatments, compared to the conventional fertilised treatment, despite the application (at time 0) of exogenous C at rates of 2.0, 2.7 and 5.9 Mg C ha^{-1} with the Bio-L, Char and Bio-H treatments, respectively. Whereas the C loss detected in the Bio-H and Bio-L treatments could be triggered by the priming effect of the added N, this was not the case for the biochar treatment, as most of the N was non-available. As discussed in the lysimeter experiment (Part I of this report, Calvelo Pereira et al.,2015a) the biochar treatment (Char) will have raised soil pH and P availability, which may have stimulated organic C mineralisation. Finally, it should be noted that, based on TC values at 16–20 cm depth in the drill line (area weighted data), an amount equivalent to 58% and 25% of the C applied as Bio-H and Bio-L treatments was lost after this two-year experiment, while an estimated 13% of C was gained in the Char treatment. These results thus emphasise the low stability of organic C in biosolids material – as compared to that of carbonised form − as well as the poor capacity of this sandy soil to stabilise this exogenous source of organic C.

3.5. CONCLUSIONS

A field-scale experiment consisting of the deep application of either biosolids or biochar produced from biosolids into a sandy soil (Typic Udipsamment) through direct drilling was carried out with the aim of comparing their agronomic effectiveness against a conventional fertilised treatment in the short term (2 years). Farmer's cultivation practices were followed, including a pasture renovation using a forage rape crop between old and newly sown pasture (annual ryegrass). Both the forage crop and the annual ryegrass herbage yields were positively related to the amount of available N recovered by plants, especially when the biosolids were applied. This was directly related with the amount of (available) N present in the biosolids. Concomitantly root mass and hence soil C (and N) stocks after 2 years were lower in treatments where organic amendments were applied compared to that under basal fertiliser application. While C losses detected in the Bio-H and Bio-L treatments could be triggered by the priming effect of the added N, mostly in available form, that is not clear in the case of the biochar, as it contains mostly recalcitrant C and N. More research is needed to clarify the

circumstances under which the addition of biochar triggered native organic matter mineralisation.

4. Acknowledgements

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