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Biochar and mycorrhizal fungi influence on nutrient uptake by two pasture species in New Zealand

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Abstract

The symbiotic relationship between plants and mycorrhizal fungi can play an important role in enhancing nutrient uptake from the soil. Many New Zealand farm soils have elevated levels of phosphorus because of regular, long term, application of phosphate fertiliser. Much of this phosphate is not available to plants. This study aimed to investigate the effect of biochar on mycorrhizal growth and subsequent phosphorus uptake by white clover and ryegrass. The experiment was conducted in a 3×2×2 factorial design, including two biochar produced from pine at different final pyrolysis temperature (350 and 550°C), mycorrhizal inoculation (*Glomus sp*. arbuscular mycorrhiza) and phosphate rock, respectively. The soil sample was provided from the 10cm surface horizon of an Egmont black loam. The pots were filled with the soil sample mixed with biochar applied at 10 tha^{-1}.Phosphate rock was applied at a rate of 70 kg ha^{-1}. The seeds were inoculated by *Glomus sp*. arbuscular mycorrhizae before sowing. The ryegrass also contained the endophyte, AR37. Dry weight, phosphorus and nitrogen concentrations of plant tissue were measured68 days after sowing. There was an increase in both phosphorus uptake and dry weight in plants grown with the high temperature biochar (B550). There was significant increase in phosphorus uptake and herbage yield in clover plants if inoculated by mycorrhizal fungi. The results demonstrated that biochar have the potential to affect the nutrients in soil and microbial activity.

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Introduction

Phosphorous is a necessary macronutrient and a very important part of pasture farming in New Zealand. Intensive dairy systems in New Zealand rely on elevated pasture production based on optimal grazing conditions through the year (MacLeod and Moller 2006). Perennial ryegrass (*Lolium perenne* L.) and white clover (*Trifolium repens* L.) are forage species widely used in pastoral livestock systems in New Zealand (Kemp *et al.*, 2010). Usually, phosphorus (P) fertilisation is essential in New Zealand farming in order to maintain high herbage production and provide this key macronutrient for plant growth, especially considering that soils generally show low P saturation (Monaghan *et al.*, 2007). P fertilisation has been applied in New Zealand farms for 100 years, causing an increase in total P content of the topsoil (Moir *et al.,* 1997), mainly as insoluble-, non-available-P.

Mycorrhizal fungi can increase the availability of phosphate and help plants to take up phosphate and increase available-P reserves (Trolove *et al.,* 2003). Generally, the nutrient uptake from soil solution, and water acquisition, increased when plants were inoculated with mycorrhizal fungi (Oseni *et al.,* 2010).

Biochar is a carbon-rich substrate produced by pyrolysis (IBI 2012; Lehmann *et al.,* 2011) used as soil amendment amenable to induce changes on soil structure, water-holding capacity, soil fertility and micro-organism abundance and/or functionality, including mycorrhizal fungi (Kloss *et al.,* 2013; Sun and Lu 2013; Warnock *et al.,* 2007). The particle size distribution of biochar is also an important property of biochar (Cetin *et al.,* 2004), as it can have a greater effect on soil than type of feedstock or conditions of pyrolysis (Zimmerman, 2010; Angers and Recous, 1997). Physical properties of biochar, such as porosity, depend upon its particle size distribution. Biochar with different pores can affect differently physical and chemical characteristics of soils. The macro–pores of biochar increase the water-holding capacity of soils and adsorption of water-soluble organics due to a high specific surface area. The micro–pores of biochar affect the population of soil micro-organism significantly by to providing protection from soil faunal predators and a food source in the form of adsorbed organic compounds (Briggs *et al.,* 2012). The application of biochar as a soil amendment can be beneficial to plant hosts due to increased mycorrhizal fungi abundance and/or functionality in soil (Warnock *et al.,* 2010; Rillig and Mummey, 2006) by enhancing plant–fungus symbiosis. It has been argued that biochar application increased soil nutrient availability and enhanced mycorrhizal root colonisation by the improvement of soil characteristics (Ishii and Kadoya, 1994). Biochar can directly improve plant productivity (Schulz and Glaser, 2012) by increasing soil nutrient content and the availability of the soluble forms of nutrients, even as a potential P source with high-agronomic efficiency (Wang *et al.,* 2012 A). The improvement of nutrient retention can explain the indirect effect of the biochar addition in soil (Lehmann *et al.,* 2003). Therefore biochar application and mycorrhizal fungi can both improve plant productivity, however the specific mechanism(s) of biochar on mycorrhizal fungi remains largely unknown(Lehmann *et al.,* 2011; Warnock *et al.,* 2007).

For investigating the role of mycorrhizal fungi when biochar is added to soil in detail, our study pursued the subsequent objectives: i) to assess the effect of biochar amendment on herbage production and nutrient uptake in absence of chemical fertilisers; ii) to compare the effect of pyrolysis temperature; iii) to study influence of fungi when soil P retention is high and a P-fertiliser of low availability is added. A sward composed by ryegrass and clover, very productive in New Zealand, was grown under greenhouse conditions, mixed with biochar produced from pine at two different temperatures, 350 and 550°C.

Materials and method

Biochar characteristics and soil properties

Pine (*Pinus radiata* D. Don) logging slash residues were used as feedstock. Two biochar substrates were produced by slow pyrolysis in a 5 L gas-fired rotating

drum kiln, as described by Calvelo Pereira *et al.* (2011), using two different highest heating temperatures (HHT), 350°C, and 550°C (B350 and B550, respectively). Full control of the heating rate was not possible, but temperature changes and mean heating rates were monitored. After cooling, the biochar was homogenised and sub-samples were taken for: i) to determine the particle size distribution by using dry-sieving method; and ii) chemical characterisation and thermal analysis (finely ground biochar; < 500 µm).

Biochar samples (100 g, n=3) were sieved by weighting aliquots that were passed through 8 sieves: 2, 1, 0.5, 0.25, 0.15, and 0.053 mm. Data were calculated as the proportion of the initial aliquot mass in the different size fractions defined: > 2, 2 – 1, 1 – 0.5, 0.5 – 0.25, 0.25 – 0.15, 0.15 – 0.053, < 0.053 mm and finally cumulative distribution was calculated (Figure 1).

Biochar pH and electric conductivity (EC) were measured (n = 2) using ground charcoal in deionized water (1:20 w:v) after 1.5 hours shaking (Rajkovich *et al.,* 2012). Total C, hydrogen (H) and nitrogen (N) contents were determined with a vario MACRO cube CHNS elemental analyser (Elementar Analysensysteme GmbH, Hanau, Germany). The ash content was determined by thermal analysis with a thermo-gravimetry analyser (SDT Q600, TA Instruments, Melbourne, Australia) (Calvelo Pereira *et al.,* 2011). Oxygen content was estimated as follows: O = 100 – (C + H + N + ash) (all expressed as weight %). Thermo-gravimetric (TG) and derivative (DTG) curves were obtained with the SDT Q600 instrument described earlier and the volatile matter content (dry matter basis) and the stable, thermo-resistant fraction or fixed C (dry matter basis) determined (n = 3) by following the method described by Calvelo Pereira *et al.* (2011). Biochar available N and P were measured following procedures described by Wang *et al.* (2012 A and B).

Soil from the A horizon was collected from an Egmont black loam, a Typic Orthic Allophanic Soil, from Mokoia in South Taranaki, New Zealand. The site supports highly productive dairy farming and has been farmed continuously since European settlement of the area over 100 years ago (Moir *et al.*, 1997). The soil hasallophanic clays that react with phosphorus fertiliser to significantly limit its availability and has high P retention. Chemical characteristics of the soil: 33.3 mgP kg^{-1}(Olsen-P), 1.11 mgN $kg^{-1}$and a pH value of 6.2.

Greenhouse experience setup

A greenhouse experiment was conducted at the Plant Growth Unit, Massey University, Palmerston North, New Zealand. PVC pipes (15 × 20 cm^2) used as pots, were divided into three compartments of equal size. The plant roots were confined to the top compartment by a 30 µm film that mycorrhizal hyphae could grow through to access nutrients in the second compartment. The hyphae were prevented from growing into the third compartment by a filter film with a pore size of 0.45 µm.

A factorial design (3×2×2) was used by a completely randomized design (CRD) with 4 replications. The three factors used here were: i) biochar amendment, including 3 levels [no amendment, addition of B350, addition of B550; dose of biochar was always 10 t ha^{-1} (biochar was deployed in the first compartment)]; ii) fungi presence, including 2 levels (M_0, no mycorrhizal inoculum added, M_1,mycorrhizal inoculum added); and iii) phosphate-rock addition, including to levels (P_0, no phosphate rock addition, P_1, addition of 70 kg phosphate-rock ha^{-1}). G*lomus sp.* arbuscular fungi were used as mycorrhizal treatment. As phosphate rock has low water solubility, it will not diffuse through the film and it was assumed that P could only be made available through interactions with soil micro-biota. The control treatment was the one with soil alone.

Common pasture plants were grown in the PVC pipes filled by the corresponding amendments. Six seeds each of Tetraploid ryegrass Base containing the endophyte AR37 (*Lolium multiflorum* Lam) and conquest white clover (*Trifolium repens*) were sown

in each pot. The seeds of white clover were inoculated with *Glomus sp*. arbuscular mycorrhizal fungi. These were thinned to 4 plants of each per pot after 7 days sowing. The soil moisture of pots was maintained at field capacity. No additional N or P fertilisation was provided. Pots were left in a greenhouse and maintained at ambient temperature for ~10 weeks.

The plants were harvested after 68 days of growth. Fresh weight recorded and biomass dried at 65°C for at least 24 hours prior to analysis. The foliage was analyzed for total nitrogen and total phosphorus content by Kjeldahl method.

Statistical analyses

Collected data was performed using analysis of variance (ANOVA) using SAS program (SAS Institute, Cary, NC, USA, 2003) and differences between individual means were determined using Tukey's Studentized test at the 5 and 1% level of probability.

Results and discussion

The particle size distribution of B350 and B550 was similar. The effective size (D10) was recorded at 0.35 mm of diameter. The particle size between 1 and 2mm was the most common (60 – 70%) in the biochar used in this study (Figure 1). The main chemical characteristics of biochar substrates used in this study are summarised in Table 1. Both biochar substrates had a basic pH, with values of 8.4, and 9.8 for B350 and B550, respectively; EC values were below 0.5 mS m^{-1} (Table 1). Carbon content was moderately high (B350: 756 g kg^{-1}; B550: 892 g kg^{-1}) and N contents were always <4 g kg^{-1}. The atomic H:C_{org} ratio was lower for B550 (0.31) than in B350 (0.58); their respective O:C_{org} ratios were 0.05 and 0.19 (Table 1). Ash content was always < 20 g kg^{-1}. Moisture content was always very low (≤ 2.5%). B350 had more volatile matter content (35.5%) than B550 (11.1%). The relative contribution of fixed C to the C_{org} content was 79 and 98% in B350 and B550, respectively. B550 has a high content available N (369 g kg^{-1}) and a low content of available P (54 g kg^{-1}) (Table 1).

Table 1. Relevant chemical properties of biochar sproduced from pine feedstock used in this study. HHT: highest heating temperature; EC, electrical conductivity; Fix.C: fixed C; VM: volatile matter.

Biochar	B350	B550
HHT / °C	350	550
Property		
pH (H_2O)	8.4	9.8
EC / mS m^{-1}	0.09	0.47
C_{org} / g kg^{-1}	756.0	892.0
N / g kg^{-1}	3.2	3.7
H / g kg^{-1}	37.0	23.0
O / g kg^{-1}	196.1	62.8
Ash / g kg^{-1}	7.7	18.5
Fix.C / g kg^{-1}	597.1	870.9
VM / %	35.5	11.1
Moisture / %	2.5	1.0
Atomic ratio		
H:C_{org}	0.58	0.31
O:C_{org}	0.19	0.05
Available N / mg kg^{-1}	90.0	369.0
Available P / mg kg^{-1}	72.0	54.0

The dry weight (DW) of ryegrass and white clover plants was differently affected by treatments; ryegrass biomass constituted on average 90% of DW of the herbage (Figure 2). As shown in Figure 2a-b, it appears that white clover responded more positively to the main effect of treatments than ryegrass. The highest dry weight of clover plant was recorded when the plant was treated by mycorrhizal fungi, phosphate rock and biochar produced at 550°C (B550) together. However, the dry weight of ryegrass plant increased

by the addition of biochar made at 350°C (B350) conjunction with mycorrhizal fungi and the phosphate rock application.

There was a decrease in the phosphorus content of white clover tissue as compared to control when the phosphate rock was applied with the biochar made at high temperature (B550). The interaction effect of biochar and mycorrhizal inoculation showed no significant difference between treatments in both treated plants (Table 2). As shown in Table 2 and Figure 2c, the significant simple and interaction effects of biochar amendment and mycorrhizal inoculation on phosphorus content in ryegrass tissues were not observed.

Table 2. Statistical summary of ANOVA analysis. DM, dry matter; Rye, ryegrass; WC, white clover; n.s., not significant.

Variable	DM accumulation		P concentration		N concentration		Uptake	
Source	Rye	WC	Rye	WC	Rye	WC	P	N
A*	n.s.	n.s.	n.s.	0.083	0.014	n.s.	0.004	n.s.
M*	n.s.	0.080	n.s.	0.006	n.s.	n.s.	n.s.	n.s.
P*	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
A × M	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
A × P	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
M × P	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
A × M × P	n.s.	n.s.	n.s.	0.002	n.s.	n.s.	n.s.	n.s.

* A: Amendment, M: Mycorrhiza, P: P addition.

However, the amendment with biochar produced at high temperature (B550) increased white clover phosphorus content when it was added with mycorrhizal fungi and phosphate rock (Figure 2d). As shown in Table 2 and Figure 2c, the significant simple and interaction effects of biochar amendment and mycorrhizal inoculation on phosphorus content in ryegrass tissues were not observed. Therefore it can be concluded that the plants tissues phosphorus content was depended on the kind of plants.

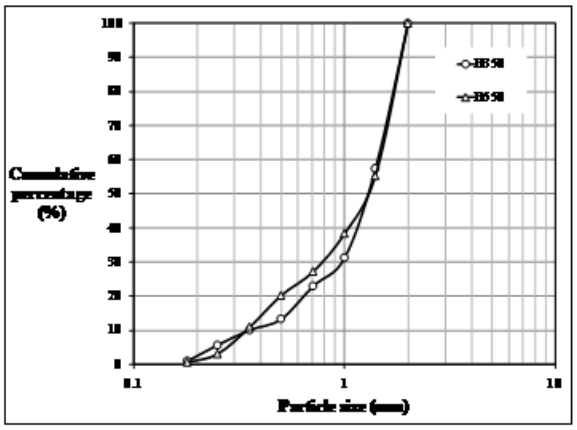

Fig. 1. The particle size distribution of biochar samples.

Regarding N uptake, each plant showed a specific pattern. The nitrogen content of ryegrass herbage decreased with biochar addition, whereas the opposite was true for white clover (Figure 2e-f). The nitrogen concentration of white clover herbage always tended to increase when the biochar produced at high temperature was present (Figure 2f).

Considering pasture mix response, addition of biochar significantly ($P \leq 0.05$) affected the phosphorus uptake by the sward (Figure 3). The biochar produced at 550°C (B550) increased the tissue phosphorus content compared to control and the low temperature biochar (B350) (Figure 3; Table 2). The mycorrhizal inoculation and phosphate application tended to increase the phosphorus content (Figure 3a). This was plant-dependent, as commented above. Considering the total herbage, no clear effect on N uptake was detected (Figure 3b).

The results demonstrated that there was an increase in both phosphorus uptake and dry weight in plants grown with the biochar made at a highest heating temperature of 550 °C (B550). There was a significantly more phosphorus and a higher dry weight in clover plants that were inoculated with mycorrhizal fungi (Figures 2). There was a

corresponding decrease in N content of the plant tissue. This was probably that the biochar addition caused to amend the soil characteristics and microorganism activity, particularly mycorrhizal fungi. Biochar as a soil amendment can play an important role to enhance plant–microorganism symbiosis. It can be observed in the nitrogen content of white clover tissue because this plant can fix nitrogen by a symbiotic life (Figure 2e-f). It was reported that biochar by amending soil physical and chemical properties can affect soil nutrient availability (Gundale and DeLuca, 2006; Matsubara *et al.,* 2002). It was also noted that increasing in soil nutrient availability may cause to enhance host plant response and raise tissue nutrient concentration in addition to higher colonization rates of the host plant roots by mycorrhizal fungi (Rillig and Mummey, 2006; Warnock *et al.,* 2010). Of course, there are a number of complexities within the growth patterns and interactions of plants grown in the different treatments that could be explored further. Therefore it can be the next challenge in that area.

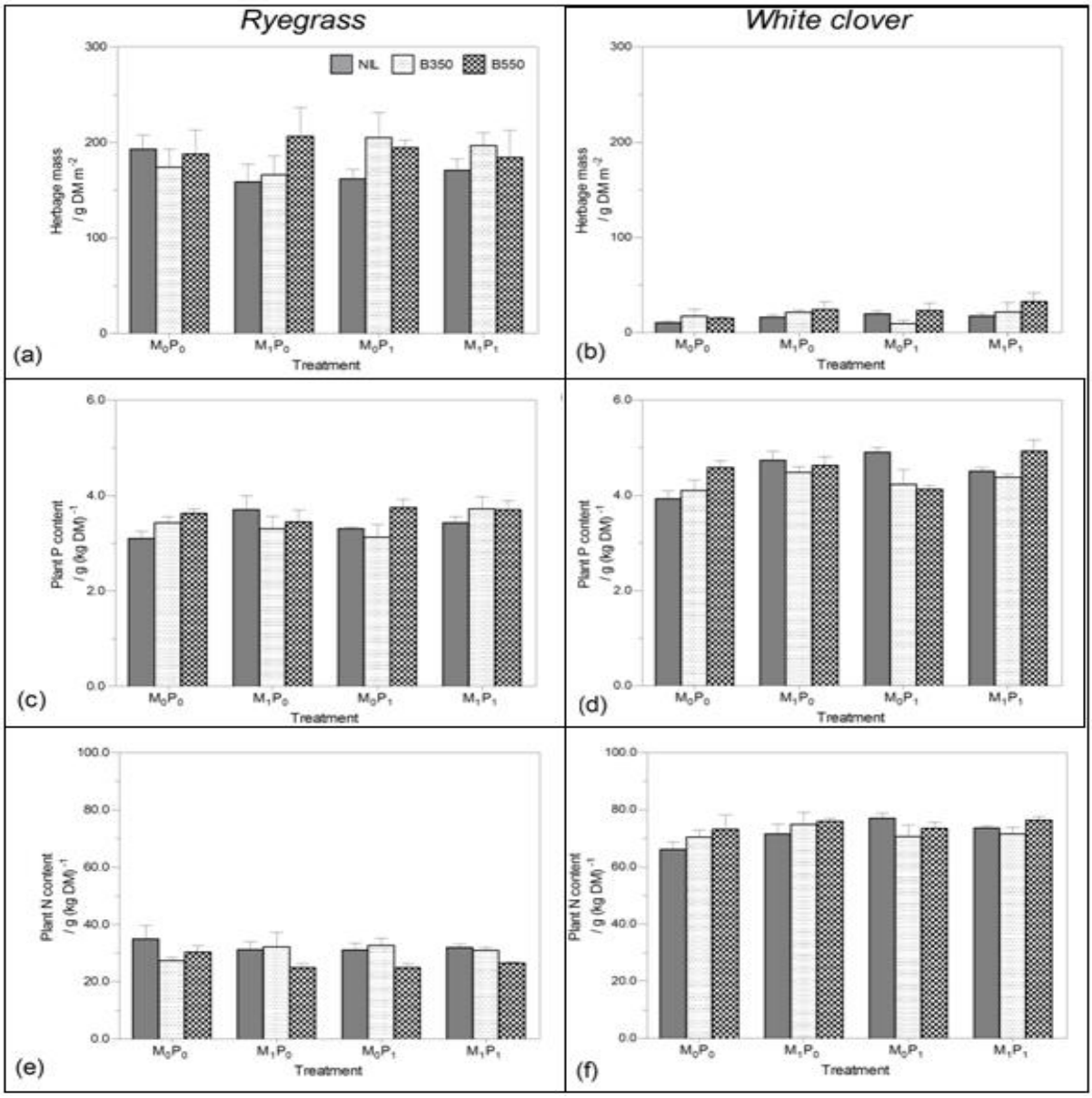

Fig. 2. Effect of biochar amendment (NIL, B350 and B550), mycorrhizae inoculation (M_0 and M_1) and phosphate rock addition (P_0 and P_1) on: i) dry weight (DW) of ryegrass (a) and white clover (b); ii) plant P content of ryegrass (c) and white clover (d); and iii) plant N content of ryegrass (e) and white clover (f). Average values (n = 4) are shown; vertical bars represent +SEM.

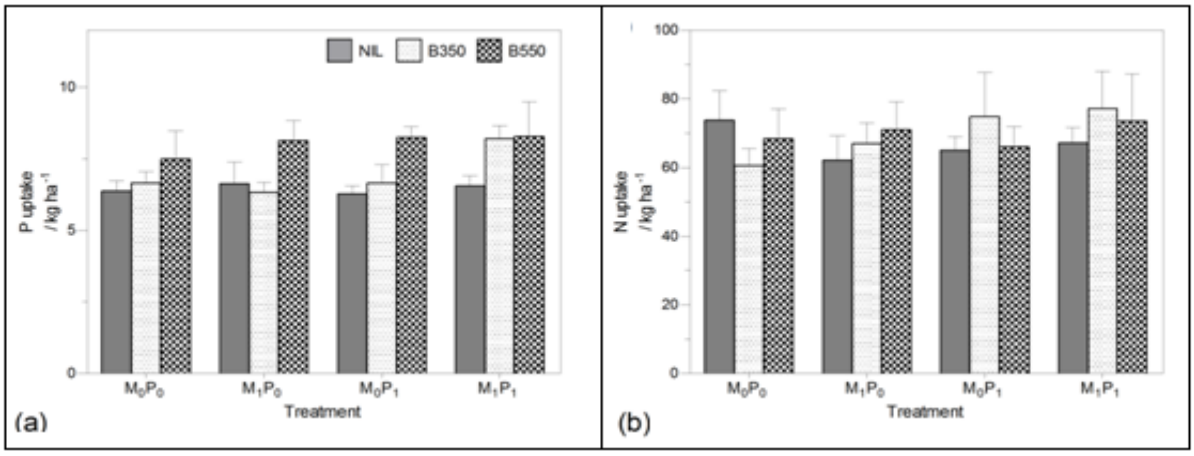

Fig. 3. Average values (n = 4) for (a) P and (b) N uptake (kg ha–1) for the pasture sward studied, considering biochar amendment (NIL, B350 and B550), mycorrhizae inoculation (M0 and M1) and phosphate rock addition (P0 and P1). Vertical bars represent +SEM.

Conclusions

The hypothesis set for this study was that ryegrass and white clover differ in their amount of nutrient uptake including phosphorus and nitrogen when they were treated by biochar and mycorrhizal fungi. The results demonstrated that biochar have the potential to significantly affect the nutrients in soil. That was significantly supported by the experimental evidence from this study however there was different effect between treatments. The treatments affected the phosphorus content and dry weight of white clover more than of ryegrass. In addition, biochar can improve soil characteristics and microbial activity. It needs that the application of the results from this study can be explored under field condition.

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References

Angers DA, Recous S. 1997. Decomposition of wheat straw and rye residues as affected by particle size. Plant and Soil **189(2),** 197-203. http://dx.doi.org/10.1023/A:1004207219678

Briggs C, Breiner JM, Graham RC. 2012. Physical and Chemical Properties of Pinus ponderosa Charcoal: Implications for Soil Modification. Soil Science **177,** 263-268.

Calvelo Pereira R, Kaal J, Camps-Arbestain M, Pardo Lorenzo R, Aitkenhead W, Hedley M, Macías F, Hindmarsh J, Maciá-Agulló JA. 2011. Contribution to characterisation of biochar to estimate the labile fraction of carbon. Organic Geochemistry **42(11),** 1331-1342. http://dx.doi.org/10.1016/j.orggeochem.2011.09.002

Cetin E, Moghtaderi B, Gupta R, Wall TF. 2004. Influence of pyrolysis conditions on the structure and gasification reactivity of biomass chars. Fuel **83,** 2139-2150. http://dx.doi.org/10.1016/j.fuel.2004.05.008.

Gundale MJ, DeLuca TH. 2006. Temperature and source material influence ecological attributes of ponderosa pine and Douglas-fir charcoal. Forest Ecology and Management **231,** 86-93. http://dx.doi.org/10.1016/j.foreco.2006.05.004

IBI. 2012. Standardized Product Definition and Product Testing Guidelines for Biochar That Is Used in Soil. IBI, International Biochar Initiative **47.** Accesed on June 2012. http://www.biocharinternational.org/sites/default/files/Guidelines_for_Biochar_That_Is_Used_in_Soil_Final.pdf.

Ishii T, Kadoya K. 1994. Effects of charcoal as a soil conditioner on citrus growth and vesicular-arbuscular mycorrhizal development. Journal of the Japanese Society for Horticultural Science **63,** 529-535. http://dx.doi.org/10.2503/jjshs.63.529

Kemp PD, Kenyon PR, Morris ST. 2010. The use of legume and herb forage species to create high performance pastures for sheep and cattle grazing systems. Revista Brasileira de Zootecnia **39,** 169-173.

Kloss S, Zehetner F, Wimmer B, Buecker J, Rempt F, Soja G. 2013. Biochar application to temperate soils: Effects on soil fertility and crop growth under greenhouse conditions. Journal of Plant Nutrition and Soil Science, http://dx.doi.org/10.1002/jpln.201200282.

Lehmann J, Pereira da Silva J, Steiner C, Nehls T, Zech W, Glaser B. 2003. Nutrient availability and leaching in an archaeological Anthrosol and a Ferralsol of the Central Amazon basin: fertilizer, manure and charcoal amendments. Plant and Soil **249,** 343-357.

Lehmann J, Rillig MC, Thies J, Masiello CA, Hockaday WC, Crowley D. 2011. Biochar effects on soil biota – A review. Soil Biology and Biochemistry **43,** 1812-1820.

MacLeod CJ, Moller H. 2006. Intensification and diversification of New Zealand agriculture since 1960: An evaluation of current indicators of land use change. Agriculture, Ecosystems & Environment **115,** 201-210.

Matsubara Y, Hasegawa N, Fukui H. 2002. Incidence of Fusarium root rot in asparagus seedlings infected with arbuscular mycorrhizal fungus as affected by several soil amendments. Journal of the Japanese Society for Horticultural Science **71,** 370-374. http://dx.doi.org/10.2503/jjshs.71.370

Monaghan RM, Hedley MJ, Di HJ, McDowell RW, Cameron KC, Ledgard SF. 2007. Nutrient management in New Zealand pastures— recent developments and future issues. New Zealand Journal of Agricultural Research **50,** 181-189.

Moir JL, Hedley MJ, Mackay AD, Tillman RW. 1997. The effect of fertilizer history on nutrient accumulation and plant available nutrient supply in legume based pasture soils. In: 18th International Grasslands Congress, Saskatoon, Canada, University of Saskatchewan **18,** 68–69.

Oseni T, Shongwe N, Masarirambi M. 2010. Effect of arbuscular mycorrhiza (AM) inoculation on the performance of tomato nursery seedlings in vermiculite. International Journal of Agriculture & Biology **12,** 789-790.

Rajkovich S, Enders A, Hanley K, Hyland C, Zimmerman A, Lehmann J. 2012. Corn growth and nitrogen nutrition after additions of biochars with varying properties to a temperate soil. Biology and Fertility of Soils **48,** 271-278. http://dx.doi.org/10.1007/s00374-011-0624-7

Rillig MC, Mummey DL. 2006. Mycorrhizas and soil structure. New Phytologist **171,** 41-53. http://dx.doi.org/10.1111/j.14698137.2006.01750.x

Schulz H, Glaser B. 2012. Effects of biochar compared to organic and inorganic fertilizers on soil quality and plant growth in a greenhouse experiment. Journal of Plant Nutrition and Soil Science **175,** 410.

Sun F, Lu S. 2013. Biochars improve aggregate stability, water retention, and pore-space properties

of clayey soil. Journal of Plant Nutrition and Soil Science.
http://dx.doi.org/10.1002/jpln.201200639.

Trolove SN, Hedley MJ, Kirk GJD, Bolan NS, Loganathan P. 2003. Progress in selected areas of rhizosphere research on P acquisition. Soil Research **41,** 471-499.
http://dx.doi.org/10.107/SR02130

Wang T, Camps-Arbestain M, Hedley M, Bishop P. 2012 A. Predicting phosphorus bioavailability from high-ash biochars. Plant and Soil **357,** 173-179.

Wang T, Camps-Arbestain M, Hedley M, Bishop P. 2012 B. Chemical and bioassay characterisation of nitrogen availability in biochar produced from dairy manure and biosolids. Organic Geochemistry **51,** 45-48.
http://dx.doi.org/10.1016/j.orggeochem.2012.07.009

Warnock D, Lehmann J, Kuyper T, Rillig M. 2007. Mycorrhizal responses to biochar in soil – concepts and mechanisms. Plant and Soil **300,** 9-20.
http://dx.doi.org/10.1007/s11104-007-9391-5

Warnock DD, Mummey DL, McBride B, Major J, Lehmann J, Rillig MC. 2010. Influences of non-herbaceous biochar on arbuscular mycorrhizal fungal abundances in roots and soils: Results from growth-chamber and field experiments. Applied Soil Ecology **46,** 450-456.
http://dx.doi.org/10.1016/j.apsoil.2010.09.002

Zimmerman AR. 2010. Abiotic and Microbial Oxidation of Laboratory-Produced Black Carbon (Biochar). Environmental Science & Technology **44,** 1295-1301.
http://dx.doi.org/10.1021/es903140c