**Soil Response to Perennial Herbaceous Biofeedstocks under Rainfed Conditions in the
Northern Great Plains, USA**

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Abstract

Perennial herbaceous biofeedstocks (PHB) have been proposed to confer multiple ecosystem services to agricultural lands. However, the role of PHBs to affect change in soil condition is not well documented, particularly for treatments with multiple species. The objective of this study was to quantify potential changes to soil properties resulting from PHB treatments in central and western North Dakota over a 5-yr period. Treatments with multiple perennial plant species were hypothesized to induce greater improvements in soil condition compared to monocultures. Soil properties were evaluated in seven PHB treatments (four monocultures, three mixtures) at five sites with sampling occurring immediately prior to treatment establishment in 2006 and again in 2011 across a 0 to 1.2 m depth. Perennial herbaceous biofeedstocks had minor and inconsistent effects on soil bulk density, electrical conductivity, and soil pH, and no effect on available P and soil organic C (SOC) in 2011. Contrasts between monoculture and mixtures in 2011 yielded no significant differences for any soil property at any site. However, PHB treatments did induce significant changes in soil properties between 2006 and 2011, with substantial declines in available P ($>10 \text{ kg P ha}^{-1} \text{ yr}^{-1}$) at sites with high initial P and modest increases in SOC ($0.9\text{-}5.7 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$) at sites with low initial SOC. Electrical conductivity decreased at two sites, though changes were minor (-0.08 to -0.18 dS m^{-1}). Soil pH did not change over the 5-yr study. Results from this study underscore the value of PHBs to remediate nutrient-laden and/or degraded soils, while concurrently resisting increased salinity and fertilizer-induced acidification.

Keywords: *Ecosystem services; Northern Great Plains; Soil pH; Switchgrass*

47 **Abbreviations**

48 CRP, Conservation Reserve Program

49 ESM, Equivalent soil mass

50 GHG, Greenhouse gas

51 P, Phosphorus

52 PHB, Perennial herbaceous biofeedstock

53 SOC, Soil organic carbon

1. Introduction

As the portfolio of biofuel production options expand globally, it is essential to understand how different biofeedstocks affect soil ecosystem services across spatial and temporal scales. Currently, production of first-generation biofuel crops such as corn (*Zea mays* L.) have significant negative environmental consequences, including increased net greenhouse gas (GHG) emissions (Adler et al., 2007; Scharlemann and Laurance, 2008), compromised water quality (David et al., 2010), and decreased wildlife habitat (Wright and Wimberly, 2013). Utilization of second-generation perennial herbaceous crops as biofuel sources has been suggested to reduce these negative consequences (McLaughlin et al., 2002), due mainly to their lower requirements of agricultural inputs and their ability to be grown on marginal land (Hill et al., 2006). Many ecosystem service benefits associated with perennial herbaceous biofeedstocks (PHB) have been found to be derived from changes to soil properties (Blanco-Canqui, 2010). Limited soil disturbance, coupled with increased organic matter inputs and water uptake in comparison to annual crops, contribute to changes in soil properties that can affect climate and water regulation, nutrient cycling, and salinity mitigation (Franzluebbers, 2015; Stewart et al., 2015).

In the northern Great Plains of North America, most assessments of PHB have focused on switchgrass (*Panicum virgatum* L.), where previous modeling efforts have shown its production for bioenergy to be economically feasible (Walsh, 1998). Information associated with the production of other grasses and forbs in the region for potential use as biofeedstocks, such as tall and intermediate wheatgrass [*Thinopyrum ponticum* (Podp.) Z.-W. Liu and R.-C. Wang; *Thinopyrum intermedium* (Host) Barkworth and D.R. Dewey], alfalfa (*Medicago* spp.), sweetclover [*Melilotus officinalis* (L.) Lam.], big bluestem (*Andropogon gerardii* Vitman), and

species mixtures, is limited (Lee et al., 2009). Moreover, previous multisite perennial biofeedstock trials in the northern Great Plains have focused on on-farm environments (Schmer et al., 2008), where management options and experimental controls can be limited and inconsistent across sites.

Species mixtures are purported to induce greater improvements in soil condition relative to monocultures given more abundant and deeper distribution of root biomass in the former (Blanco-Canqui, 2010). Though positive associations between species richness and soil organic C (SOC) have been observed (Skinner and Dell, 2016; Cong et al., 2014; Fornara and Tilman, 2008), outcomes are far from consistent as some studies have observed decreased SOC with increased species richness (Skinner et al., 2006) or an absence of an association altogether (Bonin et al., 2014). Moreover, studies evaluating species mixture effects on soil properties often focus on SOC, with limited information on other properties known to affect soil function.

In this study, we sought to quantify potential changes to soil properties resulting from different PHB at five sites in central and western North Dakota over a 5-yr period. Soil properties investigated in the study were selected for their association with water regulation (soil bulk density), salinity mitigation (electrical conductivity), buffering capacity (soil pH), nutrient cycling (available P), and climate regulation (SOC). This investigation was done in conjunction with a broader study objective that sought to determine appropriate grass and legume species, harvest methods, and management practices to maintain productive perennial biomass stands throughout North Dakota (Wang et al., 2014, 2013). We hypothesized PHB treatments with multiple plant species would induce greater improvements in soil condition compared to monoculture PHB treatments.

2. Materials and methods

2.1. Site descriptions

Sites were located in central and western North Dakota USA at North Dakota State University (NDSU) Research Extension Centers near towns of Carrington, Hettinger, Minot, Streeter, and Williston (Fig. 1). Major land resource areas represented in the study included 53B (Central dark brown glaciated plains; Minot), 54 (Rolling soft shale plain; Hettinger and Williston), and 55B (Central black glaciated plains; Carrington and Streeter), which encompass approximately 17 Mha (USDA-NRCS, 2006). Climate within the study region is characterized as semiarid to sub-humid continental, with cold and dry winters, warm to hot summers, and erratic precipitation (Bailey, 1995). Mean annual temperature and precipitation at the sites ranged from 4.5 to 5.6°C and 381 to 505 mm, respectively (Table 1). Soils at the sites were of moderate to high inherent fertility, characterized by Albolls, Ustolls, and Udolls as taxonomic suborders, and occupied a spatial extent of 45 Mha (USDA-NRCS, 1999).

Previous land management was typical for crop and hay production in central and western North Dakota. A two year crop rotation including small grains and pulse crops was used at Carrington and Minot. Continuous spring wheat (*Triticum aestivum* L.) using no-till practices was used at Hettinger. At Streeter, previously cropped land was fallowed for four years prior to establishment of the study. The site at Williston was used for hay production in grassland dominated by crested wheatgrass [*Agropyron cristatum* (L.) Gaertn.] (Wang et al., 2014). All sites reported in this study were rainfed and did not receive supplemental irrigation.

Ten perennial plant species and species combinations, each split by two harvest schedules (annual and biennial), were seeded at all sites the week of 15 May 2006 (Wang et al., 2014). Plots were seeded with a drill designed for small-seeded grasses and legumes with 0.15 m row centers. Plot dimensions were 4.6 x 9.2 m. The experimental design was a randomized complete

block design with four replications. For purposes of this study, seven of ten perennial plant treatments harvested annually were evaluated for their effects on soil properties. Treatments included four monocultures and three mixtures (Table 2).

Following establishment, plots were sprayed and mowed at least once at all sites except Hettinger, where they received only chemical applications. Due to poor stands from exceptionally dry conditions, plots were reseeded at Hettinger in 2008. Treatments without leguminous species received fertilizer N annually at a rate of 56 kg N ha⁻¹ as NH₄NO₃ or urea from 2007 onward. Additional information on site history, plot establishment, management, and biomass harvest may be reviewed elsewhere (Wang et al., 2014, 2013).

2.2. Sampling protocol and soil analyses

Soil samples were collected between 27 April and 15 May in 2006 prior to seeding of treatments, and again in 2011 between 3 May and 27 September. Excessive soil moisture during the 2011 growing season severely restricted plot access at Minot and Streeter, thereby requiring an extended sampling period. During each sampling, soil samples were collected near the center of each plot using a Giddings hydraulic probe (Giddings Machine Company, Windsor, CO) with an inner tip diameter of 3.54 cm. Depth increments sampled were 0-0.05, 0.05-0.1, 0.1-0.2, 0.2-0.3, 0.3-0.6, 0.6-0.9, and 0.9-1.2 m. Within each sampled plot, six cores were composited from the 0-0.05 and 0.05-0.1 m depths, three cores from the 0.1-0.2, 0.2-0.3 m depths, and two cores from the 0.3-0.6, 0.6-0.9, and 0.9-1.2 m depths. Collection of cores at Williston was hampered by excessive gravel at approximately 0.9 m, which limited sample collection at lower depth increments. Once collected, samples were saved in double-lined plastic bags, placed in cold storage at 5°C, and processed within 6-wk of collection.

Soil samples were dried at 35°C for 3-4 d and then mechanically ground to pass a 2.0 mm sieve. Identifiable plant material (>2.0 mm diameter, >10 mm length) was removed prior to grinding. Electrical conductivity and pH were estimated from a 1:1 soil-water mixture (Whitney, 1998; Watson and Brown, 1998). Plant-available soil P was estimated by bicarbonate extraction (Olson et al., 1954). Total soil C and N were determined by dry combustion on soil ground to pass a 0.106 mm sieve (Nelson and Sommers, 1996). Using the same fine-ground soil, inorganic C was measured on soils with a pH ≥ 7.2 by quantifying the amount of CO₂ produced using a volumetric calcimeter after application of dilute HCl stabilized with FeCl₂ (Loeppert and Suarez, 1996). Soil organic C was calculated as the difference between total C and inorganic C. Gravimetric data were converted to a volumetric basis for each sampling depth using field measured soil bulk density (Blake and Hartge, 1986). All data were expressed on an oven-dry basis.

2.3. Data analyses

To reduce the effects of sampling depth and soil bulk density on SOC, total N, and available P, data from the 2006 and 2011 samplings were recalculated on an equivalent soil mass (ESM) basis assuming profile masses of 470, 970, 2240, 3600, 7300, and 11300 Mg ha⁻¹ following the method of Ellert and Bettany (1995). The six equivalent masses approximated soil within the 0-0.05, 0-0.1, 0-0.2, 0-0.3, 0-0.6, and 0-0.9 m depths, respectively.

Soil attributes in 2011 were analyzed statistically to evaluate effects of PHB treatments after five years using PROC MIXED in SAS (Littel et al., 1996). Plant diversity effects on soil attributes in 2011 were evaluated similarly by contrasting monocultures (Switchgrass, Tall wheatgrass, Intermediate wheatgrass) and mixtures (Tall wheatgrass + Intermediate wheatgrass + Alfalfa + Sweetclover, Switchgrass + Tall wheatgrass, Switchgrass + Big bluestem). Given

modest effects of perennial plant species on soil attributes in 2011 (reviewed below), mean values of soil properties across treatments were calculated and compared to baseline soil conditions in 2006 to evaluate change in soil properties over the five year period. A mixed repeated-measures model was used to evaluate change in soil properties. All statistical analyses were conducted by site and soil depth (or equivalent mass) using a significance criteria of $P \leq 0.05$. Potential associations between measured variables were identified using Pearson correlation analysis. A 3600 Mg ha⁻¹ ESM for SOC stocks was used in correlations as the majority of PHB root biomass has been found to reside in the surface 0.3 m of soil (Frank et al., 2004).

3. Results

3.1. Initial soil conditions and vegetation attributes

Initial soil conditions at the five sites were not limiting to establishment and growth of perennial grasses (Table 3). Values for soil bulk density across sites were below critical threshold values for restriction of root growth (Jones, 1983). All sites were non-saline except for Minot, where electrical conductivity values exceeded 2 dS m⁻¹ for the 0.6-0.9 and 0.9-1.2 m depths. Soil pH at 0-0.3 m depths varied from slightly acid to slightly alkaline (USDA, 1993), and fell within a range for successful switchgrass germination (Hanson and Johnson, 2005). Alkalinity increased with increasing depth at all sites due to presence of inorganic C. Initial available P levels were adequate for switchgrass establishment and growth (Brejda, 2000), though accumulation of P throughout the soil profile was apparent at Carrington due to previous livestock presence at the research site. Values for SOC and TN across sites were characteristic of soils in the region with high inherent fertility (Cihacek and Ulmer, 2002).

Species composition of applied PHB treatments was maintained throughout the study at most sites. In 2011, treatments at Carrington, Minot, and Streeter each contained a majority composition of species for monoculture treatments or a balanced composition of intended species for mixture treatments (see Supplementary Material; Table S1). At Williston, invasion of grassy weeds compromised the intended vegetation composition of all treatments except for the intermediate wheatgrass and CRP mix treatments, which maintained their intended composition through 2011.

3.2. PHB effects on soil properties after five years

Perennial plant species had limited and/or inconsistent effects on soil bulk density, electrical conductivity, and soil pH in 2011 (Table 4). Soil bulk density differed among treatments at Hettinger (0.9-1.2 m), Minot (0-0.05 m), and Streeter (0.2-0.3 m), though no one treatment was consistently lower or higher relative to other treatments across sites (Table 5). Electrical conductivity at Minot under the Conservation Reserve Program (CRP) mix was greater than all other treatments at 0.2-0.3 m, while at 0-0.05 m, the CRP mix was greater than all other treatments except tall wheatgrass (Table 5). At Streeter, electrical conductivity at 0.1-0.2 m was greater under intermediate wheatgrass than all other treatments except switchgrass + tall wheatgrass. Electrical conductivity at the 0.6-0.9 m depth at Williston was greater under switchgrass than all other treatments except Sunburst switchgrass + Sunnyview Big Bluestem. It should be noted, where significant PHB treatment effects on electrical conductivity were observed, all values fell within the non-saline category (USDA, 1993). Plant species effects on soil pH were limited to Minot (0.1-0.2 m), where pH was greater under the CRP mix compared to all other treatments except Sunburst switchgrass + Sunnyview Big Bluestem (Table 5).

Stocks of SOC, TN, and available P did not differ among perennial plant species at any site (Table 6). Moreover, contrasts between monoculture and mixtures yielded no significant differences for any soil property at any site (see Supplementary Material; Table S2 and S3).

3.3. PHB soil properties: 2006 to 2011

Due to the small effects of PHB treatments in 2011, treatments were pooled and the overall effect of perennial crops was evaluated from 2006 to 2011. As reviewed below, small changes in soil bulk density, electrical conductivity, and soil pH were observed (Table 7), soil organic C and total N stocks generally increased over the five years (Table 8 and S4), and available P stocks generally decreased (Table 9).

Changes in soil bulk density, electrical conductivity, and soil pH between the initial (2006) and 5-year (2011) samplings were subtle and varied by site (Table 7). Across sites, changes in soil properties were most prevalent at Williston (6 significant changes observed), followed by Carrington (3), Minot and Streeter (2 each), and Hettinger (0). At select depths <0.3 m, soil bulk density was greater in 2011 than 2006 at Minot and Williston, but the opposite was observed at Streeter (Table 7). Where significant changes were observed, electrical conductivity decreased between 2006 and 2011 at Carrington and Williston. At both sites, electrical conductivity was associated with soil NO₃-N (data not shown), with the decrease over 5 yr reflecting plant uptake of available N. Soil pH did not significantly change between 2006 and 2011 at any site (Table 7).

Changes in SOC were observed at four sites (Table 8). At Hettinger, SOC decreased 11% within the near-surface 470 Mg ha⁻¹ ESM between 2006 and 2011, equating to a decline of -1.7 Mg C ha⁻¹ or -0.3 Mg C ha⁻¹ yr⁻¹. Soil organic C increased at the remaining sites with relative increases greatest at Williston (22-43%), followed by Minot (14-33%) and Streeter (6-

8%). Significant increases in SOC at Minot and Williston occurred throughout the soil profile (470-7300 Mg ha⁻¹ ESM), whereas increases at Streeter were limited to ESMs of 2240 and 3600 Mg ha⁻¹. Ranges of SOC accrual rates at Minot and Williston were 0.9-1.1, 1.3-1.4, 1.7-2.3, 2.0-2.6, and 2.9-5.7 Mg C ha⁻¹ yr⁻¹ for ESMs of 470, 970, 2240, 3600, and 7300 Mg ha⁻¹, respectively. Soil organic C increased by 1.1 Mg C ha⁻¹ yr⁻¹ at Streeter for both 2240 and 3600 Mg ha⁻¹ ESM. Changes in total soil N between 2006 and 2011 closely followed trends in SOC (see Supplementary Material; Table S4).

Changes in SOC were modestly related to initial SOC for the 3600 Mg ha⁻¹ ESM ($r = -0.21$, $P = 0.0377$; Fig. 2). The negative relationship implied greater initial SOC stocks were associated with smaller changes in SOC. Evaluation of PHB aboveground biomass and SOC change at 3600 Mg ha⁻¹ ESM did not confirm a relationship between variables across sites ($P = 0.2051$) (data not shown).

Significant changes in available P were observed at all sites between 2006 and 2011 (Table 9). Available P decreased at Carrington, Hettinger, and Minot, whereas P increased at Streeter and Williston. Among sites with decreasing available P, decreases were most pronounced across ESMs at Minot (53-63%), least at Hettinger (27-38%), and intermediate at Carrington (34-44%). Due to large initial P stocks, absolute decreases in available P at Carrington were substantial, with reductions of 4.8, 6.0, 8.6, 11.0 kg P ha⁻¹ yr⁻¹ for ESMs of 470, 970, 2240, and 3600 Mg ha⁻¹, respectively. In contrast, decreases in available P ranged from 2.6 to 5.8 kg P ha⁻¹ yr⁻¹ at Hettinger and from 1.8 to 13.0 kg P ha⁻¹ yr⁻¹ at Minot, with greater P reductions with increasing ESM at both sites. Reductions in available P at Minot occurred across all ESMs, whereas P reductions at Hettinger were limited to 2240-11300 Mg ha⁻¹ ESM. Available P at Streeter approximately doubled between 2006 and 2011 for ESMs of 7300 and

11300 Mg ha⁻¹ (4.1 and 5.1 kg P ha⁻¹ yr⁻¹, respectively). Increases in available P at Williston were less pronounced than at Streeter, with increases of 0.8, 1.1, and 2.4 kg P ha⁻¹ yr⁻¹ for 2240, 3600, and 7300 Mg ha⁻¹ ESM, respectively.

4. Discussion

Among the seven PHB treatments included in the study, treatment effects were few and no single treatment induced consistent changes in soil properties across sites after five years. Moreover, contrasts of aboveground plant diversity effects on soil properties failed to result in any significant differences between monocultures and mixtures at any site. Accordingly, the hypothesis for the study was not supported. Additional time may serve to distinguish responses among treatments, as management-induced changes in soil properties can take decades to be resolved under semiarid conditions (Mikha et al., 2006). However, the value of future evaluations would be predicated on maintenance of intended vegetation composition within treatments. While fertilizer N did not appear to reduce plant diversity of PHB mixtures in this study, N addition can reduce grassland diversity over time (Harpole et al., 2016). Furthermore, continued invasion of treatments by grassy weeds could potentially confound soil-based outcomes from intended treatments in the future.

Though high-diversity grassland perennials have been associated with increased biomass yields (Tilman et al., 2006; Skinner and Dell, 2016), enhanced aboveground production does not always translate to improvements in soil condition (Skinner et al., 2006; Bonin et al., 2014). Inherent soil attributes, coupled with cross-site variability in biomass production (Wang et al., 2014), likely contributed to subtle and inconsistent PHB treatment effects. With the exception of Williston, soils were inherently fertile at each site. Soils at Carrington, Hettinger, Minot, and Streeter were medium- to fine-textured, possessed deep (>0.2 m) mollic colors, and had high

cation exchange capacity and base saturation (USDA-NRCS, 1999). Such inherent conditions may have masked subtle treatment effects throughout the soil profile. Accordingly, results from this study may not apply to low-input systems on marginal soils. Furthermore, initial soil conditions at sites were highly variable, where coefficients of variation ranged from 8-13% for soil bulk density, 33-149% for electrical conductivity, 6-9% for soil pH, and 22-115% for SOC, TN, and available P (data not shown). Biomass production was also highly variable over the course of the study, with annual yields ranging from 1.7 to 10.6 Mg ha⁻¹ across sites (Wang et al., 2014, 2013). Moreover, a significant site x treatment interaction for yield was observed (P<0.0001), implying site-specific yield responses to treatments. Consequently, if biomass-induced changes to soil properties were observed they would be expected to respond to treatments differently across sites.

Previous assessments of PHB effects on soil bulk density have been mixed, with lower soil bulk density observed under warm-season grasses compared to cropland in the north and central U.S., but the opposite occurring in the south-central U.S. (Blanco-Canqui, 2010). In a 5-yr on-farm evaluation across three U.S. states, Schmer et al. (2011) found soil bulk density under switchgrass to decrease over time at sites in North and South Dakota, but increase at sites in Nebraska. Outcomes in this study were similarly divergent, with soil bulk density increasing at two sites (Minot and Williston) but decreasing at Streeter. Tillage prior to initial soil sampling and subsequent soil resettling following treatment establishment likely resulted in observed soil bulk density increases. An explanation for decreased soil bulk density at Streeter was less clear, as biomass inputs from roots at this site were likely less than at Carrington and Minot (Wang et al., 2014) where soil bulk density either increased or didn't change. Absolute changes in soil bulk density across sites, however, were modest, only exceeding 0.2 Mg m⁻³ at Williston (0-0.05

m). Moreover, significant changes were limited to the surface 0.3 m across sites, where biomass inputs, management, and freeze-thaw effects are most prevalent (Schmer et al., 2011).

Electrical conductivity either did not change or significantly decreased over the 5-yr period. At responsive locations (Carrington and Williston), initial electrical conductivity was $<0.4 \text{ dS m}^{-1}$, suggesting decreases were associated with depletion of extractable nutrients (Johnson et al., 2001; Eigenberg et al., 2006). The lack of increased electrical conductivity across sites was noteworthy, as cropland soils throughout the northern Great Plains are susceptible to increased salinity due to cropping patterns that have facilitated decreased water uptake and associated upward movement of salts in the soil profile (Doering and Sandoval, 1976; Halvorson, 1984). Accordingly, stable or decreasing electrical conductivity under perennial herbaceous biofeedstocks reflect a capacity to diminish potential for increased salinity in this important agricultural region.

Soil pH did not change over the course of the study at any site, reflecting a resistance to acidification under PHB treatments. Despite the prevalence geologically young soils with high buffering capacity (Bluemle, 2000), increased soil acidification has been observed under rainfed, annual cropping systems throughout the northern Great Plains (Sainju et al., 2015; Reeves and Liebig, 2016). The absence of significant soil pH decreases under perennials reflects mitigation of fertilizer-induced acidification resulting from lower fertilizer N application rates and higher N uptake efficiencies compared to annual cropping (Smith and Doran, 1996; Dawson et al., 2008). Accordingly, inclusion of PHB phases in annual cropping systems can serve to stabilize soil pH.

Uptake of available P by PHB has been observed to range between 7 to 14 kg P ha⁻¹ yr⁻¹ in regions with higher precipitation than received at sites in this study (Heggenstaller et al., 2009; Lemus et al., 2009). Decreases in available P at Carrington, Hettinger, and Minot fell into this

range, however, suggesting significant P uptake potential for PHB for select sites in North Dakota. It should be noted the three sites exhibited substantial initial P stocks within the 11300 Mg ha⁻¹ ESM (Range = 78 to 222 kg P ha⁻¹). Removal of available and particulate P by switchgrass in buffer strips is well documented (Eghball et al., 2000; Blanco-Canqui et al., 2004), underscoring the value of PHB for effectively removing nutrients from soil and runoff. Outcomes from this study suggest the potential for field-scale remediation of P-affected areas with PHB.

Many evaluations of soil responses to PHB have focused on soil carbon dynamics, with particular emphasis on accrual rates of SOC (Anderson-Teixeira et al., 2009; Blanco-Canqui, 2010). Increases in SOC under switchgrass across multiple growing conditions have ranged from 1.2 to 10.1 Mg C ha⁻¹ yr⁻¹ for depths of 0.3-1.5 m over 3-10 years (Frank et al., 2004; Al-Kaisi et al., 2005; Lee et al., 2007; Follett et al., 2012), underscoring potential contributions of switchgrass to mitigate GHG emissions from agriculture (Monti et al., 2012). Increased SOC under PHB, however, is not universal, as Schmer et al. (2011) found SOC stocks under switchgrass to decrease after 5-yr at two on-farm sites in South Dakota. Outcomes from this study were similarly mixed, with three sites exhibiting SOC accrual and one site losing SOC over the 5-yr study period. Accrual rates across responsive sites ranged from 0.9 to 5.7 Mg C ha⁻¹ yr⁻¹, and generally increased with increasing soil mass/depth, thereby reflecting root biomass contributions to increased SOC at deeper depths (Blanco-Canqui, 2010). The site losing SOC (Hettinger) did so only within the near-surface 470 Mg ha⁻¹ ESM, and was driven by limited biomass production due to persistent dry conditions over the course of the study (Wang et al., 2013).

The negative relationship between initial SOC and SOC change across sites was particularly apparent at Minot and Williston. Initial SOC was low at these sites, thereby reflecting a greater capacity to accumulate organic matter than other sites within edaphic limitations of the soil (Paustian et al., 1997), assuming growing conditions favor soil C accrual. Increases in both near-surface and deeper cumulative depths at these sites reflects significant potential for improvements in soil function across the entire profile. Near-surface (<0.1 m) increases in SOC influence nutrient conservation, water infiltration, and erosion control (Franzluebbers, 2002), while SOC increases below 0.3 m enhance the role of soil to serve as a repository for atmospheric GHGs given decreased C mineralization with increasing depth (Schmer et al., 2015).

5. Conclusions

Understanding how PHB alter soil properties, and in turn, how such alterations affect ecosystem services is essential for the development and adoption of improved management practices to facilitate a transition toward multifunctional agricultural landscapes (Sanderson and Adler, 2008). Outcomes from this study demonstrated PHB mixtures did not confer improvements in soil condition compared to monocultures. Perennial herbaceous biofeedstocks, however, induced changes in soil properties over the 5-yr study, with substantial declines in available P at sites with high initial P and modest increases in SOC at sites with low initial SOC. These outcomes highlighted the value of PHB to remediate nutrient-laden and/or degraded soils. In contrast to observed changes in available P and SOC, other soil properties changed minimally (electrical conductivity) or not at all (soil pH). Such resistance to change can have important implications for continued soil function (Seybold et al., 1999), and can confer a period of

stability against a backdrop of increased salinity and acidification for rainfed cropping systems in the northern Great Plains.

Acknowledgments

Many people contributed in a technical role during sample collection, processing, and laboratory analyses. We are especially grateful to Jason Gross, Holly Johnson, Johannah Mayhew, Steven Petrik, Angela Renner, Gail Sage, Andrew Schmid, and Becky Wald. Additionally, we thank Karen Kreil and Keith Trego for project management and administrative guidance, and Arnold Kruse for providing the project ‘vision’ in 2004.

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Funding

This work was partially supported by the North Dakota Natural Resources Trust and the North Dakota Industrial Commission.

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555 **List of Figures**

556 Figure 1. Approximate location of sites included in study.

557 Figure 2. Relationship between initial soil organic C and change in soil organic C for perennial
558 biofeedstock treatments at five sites in North Dakota, USA. Outcomes expressed using an
559 equivalent soil mass of 3600 Mg ha⁻¹.

560

561 Table 1. Location, mean annual precipitation and temperature, prevalent soil type, and soil classification for sites included in study.

562

Site	MAP* (mm)	MAT† (°C)	Prevalent soil type	Soil classification
Carrington, ND	505	5.0	Heimdal-Emrick loams, 0-3% slope	Coarse-loamy, Mixed, Superactive, Frigid Calcic and Pachic Hapludolls
Hettinger, ND	394	5.8	Belfield-Savage-Daglum silt loams, 3-6% slope	Fine, Smectitic, Frigid, Glossic and Vertic Natrustolls and Fine, Smectitic, Frigid, Vertic Argiustolls
Minot, ND	473	4.7	Bowbells-Tonka complex, 0-3% slope	Fine-Loamy, Mixed, Superactive, Frigid Pachic Argiustolls and Fine, Smectitic, Frigid Argiaquic Argialbolls
Streeter, ND	434	4.5	Barnes-Svea loams, 0-3% slope	Fine-Loamy, Mixed, Superactive, Frigid Calcic and Pachic Hapludolls
Williston, ND	381	5.6	Lihen loamy fine sand, 0-3% slope	Sandy, Mixed, Frigid, Entic Haplustolls

563 * MAP = Mean annual precipitation.

564 † MAT = Mean annual temperature.

565 Table 2. Species, traits, and seeding rates (kg pure live seed ha⁻¹) for perennial biofeedstock treatments included in study.

566

Variety/species	Species trait	Seeding rate (PLS ha ⁻¹)
Sunburst Switchgrass	Warm-season	11.2
Trailblazer or Dakota Switchgrass [†]	Warm-season	11.2
Alkar Tall Wheatgrass	Cool-season	12.3
Haymaker Intermediate Wheatgrass	Cool-season	11.2
CRP mix (Intermediate + Tall + Alfalfa + Sweetclover)	Cool-season	4.5 + 5.0 + 1.1 + 0.6
Sunburst Switchgrass + Tall Wheatgrass	Warm + Cool-season	5.6 + 5.6
Sunburst Switchgrass + Sunnyview Big Bluestem	Warm-season	7.8 + 2.8

567 [†] Trailblazer was seeded at Carrington, Hettinger, and Streeter and Dakota at Williston and Minot.

Table 3. Initial soil conditions in 2006 for sites included in study. Mean values and corresponding standard error are presented by site and depth.

Site / depth (m)	Soil bulk density (Mg m ⁻³)	Electrical conductivity (dS m ⁻¹)	Soil pH (-log [H ⁺])	Available P (kg P ha ⁻¹)	Soil organic C (Mg C ha ⁻¹)	Soil inorganic C (Mg C ha ⁻¹)	Total N (Mg N ha ⁻¹)
Carrington							
0-0.05	1.17 (0.03)	0.38 (0.02)	7.02 (0.11)	55.8 (3.5)	20.2 (0.4)	NP [†]	1.8 (0.1)
0.05-0.1	1.36 (0.02)	0.28 (0.01)	6.45 (0.12)	35.1 (2.1)	19.1 (0.7)	NP	1.7 (0.1)
0.1-0.2	1.35 (0.03)	0.29 (0.01)	6.61 (0.13)	40.7 (3.6)	33.6 (1.4)	NP	3.0 (0.1)
0.2-0.3	1.32 (0.02)	0.33 (0.02)	6.82 (0.12)	30.4 (4.5)	26.9 (2.2)	NP	2.5 (0.2)
0.3-0.6	1.32 (0.01)	0.30 (0.01)	7.22 (0.07)	65.1 (11.1)	49.7 (3.2)	0.5 (0.3)	4.9 (0.3)
0.6-0.9	1.39 (0.03)	0.22 (0.01)	7.59 (0.13)	40.2 (6.1)	28.5 (1.4)	9.7 (4.0)	2.8 (0.1)
0.9-1.2	1.55 (0.03)	0.19 (0.01)	7.66 (0.17)	33.9 (4.4)	25.1 (2.3)	12.2 (3.7)	1.8 (0.1)
Hettinger							
0-0.05	1.48 (0.04)	0.29 (0.02)	6.88 (0.14)	24.5 (1.5)	15.5 (0.4)	NP	1.4 (0.1)
0.05-0.1	1.60 (0.03)	0.28 (0.02)	7.39 (0.13)	13.6 (1.0)	12.2 (0.8)	0.1 (0.1)	1.1 (0.1)
0.1-0.2	1.49 (0.02)	0.30 (0.01)	7.67 (0.12)	11.4 (0.5)	18.5 (0.8)	1.2 (0.4)	1.7 (0.1)
0.2-0.3	1.51 (0.02)	0.34 (0.01)	8.09 (0.09)	7.6 (1.0)	17.0 (0.9)	3.5 (0.8)	1.5 (0.1)
0.3-0.6	1.45 (0.01)	0.32 (0.01)	8.50 (0.03)	13.9 (1.1)	43.1 (2.0)	17.9 (2.3)	3.5 (0.1)
0.6-0.9	1.46 (0.02)	0.32 (0.01)	8.72 (0.03)	10.8 (0.7)	41.7 (4.8)	20.9 (2.2)	2.6 (0.1)
0.9-1.2	1.46 (0.03)	0.42 (0.03)	8.81 (0.04)	14.1 (1.4)	45.2 (8.7)	19.0 (1.6)	2.3 (0.2)

Site / depth (m)	Soil bulk density (Mg m ⁻³)	Electrical conductivity (dS m ⁻¹)	Soil pH (-log [H ⁺])	Available P (kg P ha ⁻¹)	Soil organic C (Mg C ha ⁻¹)	Soil inorganic C (Mg C ha ⁻¹)	Total N (Mg N ha ⁻¹)
Minot							
0-0.05	1.20 (0.02)	0.22 (0.02)	6.31 (0.13)	14.3 (1.3)	14.6 (0.5)	NP	1.3 (0.1)
0.05-0.1	1.37 (0.04)	0.24 (0.02)	6.16 (0.13)	14.0 (1.5)	14.4 (0.5)	NP	1.4 (0.1)
0.1-0.2	1.50 (0.02)	0.28 (0.02)	6.48 (0.15)	19.6 (2.7)	22.6 (1.5)	NP	2.3 (0.1)
0.2-0.3	1.53 (0.02)	0.41 (0.04)	7.20 (0.16)	12.3 (1.8)	15.2 (0.9)	1.3 (0.7)	1.6 (0.1)
0.3-0.6	1.59 (0.02)	1.43 (0.25)	7.89 (0.15)	29.7 (5.4)	38.6 (4.0)	23.9 (6.5)	3.1 (0.2)
0.6-0.9	1.65 (0.02)	2.34 (0.25)	8.28 (0.10)	36.8 (7.9)	33.1 (2.8)	34.2 (6.4)	2.0 (0.1)
0.9-1.2	1.63 (0.03)	2.54 (0.24)	8.36 (0.07)	39.7 (9.7)	34.4 (2.1)	28.7 (4.4)	1.7 (0.1)
Streeter							
0-0.05	1.20 (0.03)	0.33 (0.02)	7.14 (0.11)	5.6 (0.4)	20.1 (0.4)	0.1 (0.1)	1.9 (0.1)
0.05-0.1	1.38 (0.02)	0.34 (0.02)	7.20 (0.12)	3.7 (0.3)	20.1 (0.5)	0.1 (0.1)	1.9 (0.1)
0.1-0.2	1.30 (0.01)	0.34 (0.02)	7.28 (0.12)	3.5 (0.3)	28.3 (1.0)	0.5 (0.3)	2.7 (0.1)
0.2-0.3	1.39 (0.01)	0.36 (0.02)	7.53 (0.12)	2.3 (0.2)	22.4 (1.0)	2.1 (1.4)	2.1 (0.1)
0.3-0.6	1.37 (0.02)	0.52 (0.07)	8.15 (0.13)	4.5 (0.4)	56.4 (6.0)	38.5 (7.5)	4.4 (0.3)
0.6-0.9	1.46 (0.03)	0.79 (0.16)	8.50 (0.09)	7.5 (1.1)	45.1 (5.7)	78.6 (5.7)	2.3 (0.4)
0.9-1.2	1.59 (0.03)	0.82 (0.10)	8.59 (0.10)	7.3 (1.2)	57.9 (6.7)	54.4 (5.1)	1.2 (0.1)

574 Table 3. Cont'd.

Site / depth (m)	Soil bulk density (Mg m ⁻³)	Electrical conductivity (dS m ⁻¹)	Soil pH (-log [H ⁺])	Available P (kg P ha ⁻¹)	Soil organic C (Mg C ha ⁻¹)	Soil inorganic C (Mg C ha ⁻¹)	Total N (Mg N ha ⁻¹)
Williston							
0-0.05	1.27 (0.02)	0.31 (0.01)	7.51 (0.09)	2.7 (0.1)	13.3 (0.4)	0.1 (0.1)	1.2 (0.1)
0.05-0.1	1.50 (0.02)	0.27 (0.01)	7.48 (0.06)	1.9 (0.1)	13.0 (0.3)	0.1 (0.1)	1.2 (0.1)
0.1-0.2	1.55 (0.01)	0.22 (0.01)	7.59 (0.05)	1.5 (0.2)	19.7 (0.6)	0.1 (0.1)	1.8 (0.1)
0.2-0.3	1.50 (0.01)	0.23 (0.01)	7.87 (0.05)	1.3 (0.2)	15.5 (0.7)	0.2 (0.2)	1.4 (0.1)
0.3-0.6	1.46 (0.03)	0.25 (0.01)	8.43 (0.04)	4.8 (0.8)	53.6 (3.9)	20.2 (3.3)	3.2 (0.2)
0.6-0.9	1.54 (0.04)	0.26 (0.02)	8.83 (0.04)	20.7 (2.1)	78.9 (7.0)	30.8 (3.3)	3.0 (0.5)
0.9-1.2	-- [¶]	--	--	--	--	--	--

575 [†] None present.

576 [¶] Sample not collected due to restrictive layer at 90 cm.

577 Table 4. P-values for perennial biofeedstock treatment effects on soil bulk density, electrical
578 conductivity, and pH at five sites in North Dakota, May 2011. Treatments included four
579 monocultures and three mixtures.

580

Depth (m)	-----Site-----				
	Carrington	Hettinger	Minot	Streeter	Williston
----- Soil bulk density -----					
0-0.05	0.4241	0.9314	0.0161	0.1104	0.1956
0.05-0.1	0.4522	0.8964	0.7600	0.8899	0.3940
0.1-0.2	0.2839	0.7572	0.6749	0.8692	0.1675
0.2-0.3	0.7410	0.2577	0.9914	0.0358	0.8297
0.3-0.6	0.7664	0.9147	0.2589	0.2907	0.9044
0.6-0.9	0.1603	0.1932	0.9087	0.7907	0.3163
0.9-1.2	0.5355	0.0472	0.5818	0.7397	-- [†]
----- Electrical conductivity -----					
0-0.05	0.6448	0.5390	0.0058	0.8114	0.1097
0.05-0.1	0.8087	0.8535	0.1970	0.8696	0.2889
0.1-0.2	0.9787	0.9724	0.0715	0.0267	0.9309
0.2-0.3	0.8595	0.8185	0.0177	0.3111	0.8174
0.3-0.6	0.3556	0.3262	0.1109	0.5653	0.3255
0.6-0.9	0.6124	0.9050	0.2018	0.9099	0.0432
0.9-1.2	0.6829	0.1893	0.6129	0.6749	--
----- Soil pH -----					
0-0.05	0.0556	0.7850	0.0756	0.6822	0.8598
0.05-0.1	0.1129	0.9770	0.0824	0.7149	0.2789
0.1-0.2	0.2298	0.8611	0.0383	0.7061	0.2932
0.2-0.3	0.1007	0.5775	0.2913	0.5810	0.5541
0.3-0.6	0.3916	0.5151	0.3070	0.3621	0.7376
0.6-0.9	0.2925	0.3290	0.4210	0.9940	0.0761
0.9-1.2	0.2357	0.3081	0.6383	0.7242	--

581 [†] Sample not collected due to restrictive layer at 90 cm.

Table 5. Mean values of soil bulk density, electrical conductivity, and soil pH in 2011 affected by perennial biofeedstock treatments at responsive sites and soil depths.

584

Site / Depth (m)	Sunburst Switchgrass	Trailblazer or Dakota Switchgrass [†]	Alkar Tall Wheatgrass	Haymaker Intermediate Wheatgrass	CRP mix [¶]	Sunburst Switchgrass + Tall Wheatgrass	Sunburst Switchgrass + Sunnyview Big Bluestem
----- Soil bulk density (Mg m ⁻³) -----							
Hettinger, 0.9-1.2	1.40 bc [§]	1.49 ab	1.51 a	1.49 a	1.38 c	1.43 abc	1.49 a
Minot, 0-0.05	1.52 a	1.43 ab	1.46 a	1.29 c	1.30 bc	1.31 bc	1.42 abc
Streeter, 0.2-0.3	1.23 bc	1.19 c	1.29 abc	1.27 abc	1.33 a	1.32 ab	1.35 a
----- Electrical conductivity (dS m ⁻¹) -----							
Minot, 0-0.05	0.17 c	0.24 bc	0.27 ab	0.21 bc	0.35 a	0.18 c	0.23 bc
Minot, 0.2-0.3	0.37 b	0.21 b	0.65 b	0.28 b	1.34 a	0.18 b	0.34 b
Streeter, 0.1-0.2	0.26 b	0.23 b	0.27 b	0.37 a	0.20 b	0.29 ab	0.22 b
Williston, 0.6-0.9	0.30 a	0.21 b	0.15 b	0.16 b	0.19 b	0.17 b	0.25 ab
----- Soil pH (-log[H ⁺]) -----							
Minot, 0.1-0.2	6.43 b	6.33 b	6.40 b	6.00 b	7.07 a	6.03 b	6.53 ab

585 [†] Trailblazer was seeded at Carrington, Hettinger, and Streeter and Dakota at Williston and Minot.

586 [¶] Intermediate wheatgrass, tall wheatgrass, alfalfa, sweetclover.

587 [§] Values in a row with unlike letters differ (P≤0.05).

Table 6. P-values for perennial biofeedstock treatment effects on equivalent mass soil organic C, total N, and available P at five sites in North Dakota, May 2011. Treatments included four monocultures and three mixtures.

Soil mass [†] (Mg ha ⁻¹)	-----Site-----				
	Carrington	Hettinger	Minot	Streeter	Williston
	----- Soil organic C -----				
470	0.1892	0.4478	0.8213	0.2476	0.2769
970	0.2930	0.3865	0.9142	0.3146	0.1293
2240	0.4150	0.5632	0.9829	0.5297	0.7525
3600	0.5311	0.2581	0.9147	0.2336	0.9784
7300	0.8732	0.3782	0.5101	0.1787	0.5701
11300	0.6060	0.6915	0.8569	0.2712	-- [‡]
	----- Total N -----				
470	0.2067	0.6803	0.5866	0.1149	0.2591
970	0.3856	0.5373	0.6078	0.6761	0.0929
2240	0.4545	0.6271	0.7415	0.1860	0.6613
3600	0.5789	0.6782	0.7903	0.0672	0.9428
7300	0.8630	0.8126	0.8711	0.1406	0.5847
11300	0.5836	0.9851	0.8723	0.3488	--
	----- Available P -----				
470	0.2587	0.4907	0.5202	0.9527	0.9252
970	0.5370	0.6514	0.3369	0.8990	0.9582
2240	0.9804	0.7244	0.2537	0.7383	0.7238
3600	0.9876	0.7134	0.3130	0.5909	0.7489
7300	0.7870	0.6712	0.4889	0.4733	0.5088
11300	0.7410	0.4746	0.6617	0.6268	--

[†] Equivalent soil mass of 470, 970, 2240, 3600, 7300, and 11300 Mg ha⁻¹ reflect approximate soil depths of 0-0.05, 0-0.1, 0-0.2, 0-0.3, 0-0.6, and 0-0.9 m depths, respectively.

[‡] No data due to restrictive layer at 90 cm.

Table 7. Change in soil bulk density, electrical conductivity, and soil pH by site across all perennial biofeedstock treatments included in study.

Depth (m)	-----Site-----				
	Carrington	Hettinger	Minot	Streeter	Williston
----- Δ Soil bulk density (Mg m ⁻³) -----					
0-0.05	0.13	-0.15	0.19 [*]	-0.14	0.23 [*]
0.05-0.1	0.14	0.02	0.17 [*]	-0.11 [*]	0.08
0.1-0.2	0.15	0.07	0.07	-0.05	0.11 [*]
0.2-0.3	0.10	0.00	0.00	-0.10 [*]	0.05
0.3-0.6	0.05	0.04	0.06	-0.03	0.00
0.6-0.9	0.05	0.05	0.10	0.00	0.04
0.9-1.2	0.15	0.00	0.14	-0.02	-- [†]
----- Δ Electrical conductivity (dS m ⁻¹) -----					
0-0.05	-0.10	0.08	0.01	-0.03	-0.10 [*]
0.05-0.1	-0.07	-0.02	-0.01	-0.08	-0.10 [*]
0.1-0.2	-0.12 [*]	-0.05	0.05	-0.08	-0.08 [*]
0.2-0.3	-0.18 [*]	-0.09	0.07	-0.03	-0.10 ^{**}
0.3-0.6	-0.14 [*]	-0.07	0.24	0.08	-0.05
0.6-0.9	0.00	0.00	0.95	0.02	-0.06
0.9-1.2	0.01	0.08	0.52	0.56	--
----- Δ Soil pH (-log[H ⁺]) -----					
0-0.05	-0.06	-0.15	0.05	-0.19	-0.14
0.05-0.1	0.35	-0.26	-0.03	-0.12	-0.12
0.1-0.2	0.22	-0.08	-0.08	-0.10	0.04
0.2-0.3	0.26	-0.10	-0.34	-0.16	0.00
0.3-0.6	0.10	-0.10	-0.28	-0.10	-0.16
0.6-0.9	0.01	-0.16	-0.12	0.04	-0.20
0.9-1.2	0.13	-0.12	-0.02	-0.18	--

^{*}, ^{**} Change from initial sampling significantly different at P≤0.05 and 0.01, respectively.

[†] Sample not collected due to restrictive layer at 90 cm.

Table 8. Change in equivalent mass soil organic carbon (SOC) by site across all perennial biofeedstock treatments included in study.

Soil mass [†] (Mg ha ⁻¹)	Initial SOC (Mg C ha ⁻¹)	5 yr SOC (Mg C ha ⁻¹)	Δ SOC (Mg C ha ⁻¹)	Δ SOC yr ⁻¹ (Mg C ha ⁻¹ yr ⁻¹)
----- Carrington -----				
470	19.8 (0.4) [‡]	22.1 (0.9)	2.3	0.5
970	38.0 (0.9)	40.9 (1.8)	2.8	0.6
2240	70.8 (2.2)	73.7 (3.7)	3.0	0.6
3600	97.1 (4.1)	97.6 (5.9)	0.5	0.1
7300	144.6 (7.2)	159.3 (13.7)	14.7	2.9
11300	166.8 (8.7)	220.9 (22.0)	54.1	10.8
----- Hettinger -----				
470	15.0 (0.4)	13.3 (0.3)	-1.7 [*]	-0.3
970	26.4 (1.1)	24.6 (0.5)	-1.8	-0.4
2240	44.1 (1.9)	43.4 (1.0)	-0.6	-0.1
3600	59.9 (2.8)	58.6 (1.3)	-1.3	-0.3
7300	101.4 (4.3)	101.8 (2.0)	0.5	0.1
11300	141.7 (8.1)	135.1 (2.7)	-6.5	-1.3
----- Minot -----				
470	14.3 (0.5)	19.0 (0.5)	4.7 ^{**}	0.9
970	28.1 (0.8)	34.5 (1.0)	6.4 ^{**}	1.3
2240	49.5 (1.9)	58.2 (2.1)	8.7 ^{**}	1.7
3600	64.4 (2.7)	74.6 (3.3)	10.2 ^{**}	2.0
7300	102.4 (4.7)	117.1 (4.1)	14.7 [*]	2.9
11300	132.7 (6.0)	155.2 (7.9)	22.5	4.5
----- Streeter -----				
470	19.7 (0.4)	20.3 (0.6)	0.6	0.1
970	38.8 (0.6)	40.7 (0.7)	1.9	0.4
2240	66.5 (1.3)	72.0 (1.3)	5.5 ^{**}	1.1
3600	88.3 (1.9)	93.7 (1.9)	5.4 ^{**}	1.1
7300	143.8 (6.8)	151.8 (4.0)	8.0	1.6
11300	187.6 (9.4)	185.5 (9.6)	-2.1	-0.4
----- Williston -----				
470	13.0 (0.4)	18.6 (0.7)	5.6 ^{**}	1.1
970	25.3 (0.5)	32.4 (1.0)	7.1 ^{**}	1.4
2240	44.0 (0.7)	55.5 (2.0)	11.5 ^{**}	2.3
3600	58.7 (1.1)	71.8 (2.9)	13.1 ^{**}	2.6
7300	110.5 (3.3)	138.8 (6.1)	28.3 ^{**}	5.7
11300	-- [¶]	--	--	--

[†] Equivalent soil mass of 470, 970, 2240, 3600, 7300, and 11300 Mg ha⁻¹ reflect approximate soil depths of 0-0.05, 0-0.1, 0-0.2, 0-0.3, 0-0.6, and 0-0.9 m depths, respectively.

[‡] Values in parentheses represent standard error of the mean.

^{*}, ^{**} Change from initial sampling significantly different at P≤0.05 and 0.01, respectively.

[¶] Sample not collected due to restrictive layer at 90 cm.

Table 9. Change in equivalent mass available phosphorus (P) by site across all perennial biofeedstock treatments included in study.

Soil mass [†] (Mg ha ⁻¹)	Initial P (kg P ha ⁻¹)	5 yr P (kg P ha ⁻¹)	Δ P (kg P ha ⁻¹)	Δ P yr ⁻¹ (kg P ha ⁻¹ yr ⁻¹)
----- Carrington -----				
470	54.9 (3.4) [‡]	30.9 (1.2)	-24.0 ^{**}	-4.8
970	88.4 (4.4)	58.4 (2.3)	-30.0 ^{**}	-6.0
2240	128.1 (5.6)	85.1 (6.4)	-43.0 ^{**}	-8.6
3600	158.0 (8.3)	102.9 (10.9)	-55.1 ^{**}	-11.0
7300	218.6 (18.6)	148.3 (24.0)	-70.3	-14.1
11300	221.5 (21.5)	201.5 (39.3)	-20.0	-4.0
----- Hettinger -----				
470	23.6 (1.4)	22.9 (2.8)	-0.6	-0.1
970	36.2 (2.1)	30.2 (3.3)	-6.0	-1.2
2240	47.3 (2.5)	34.5 (3.3)	-12.8 [*]	-2.6
3600	54.3 (2.7)	36.5 (3.5)	-17.8 [*]	-3.6
7300	67.9 (3.1)	41.9 (3.8)	-26.0 ^{**}	-5.2
11300	78.3 (3.4)	49.1 (3.9)	-29.2 ^{**}	-5.8
----- Minot -----				
470	14.0 (1.2)	5.2 (1.1)	-8.9 ^{**}	-1.8
970	27.4 (2.7)	10.7 (2.0)	-16.8 ^{**}	-3.4
2240	46.4 (5.2)	18.4 (3.3)	-28.0 ^{**}	-5.6
3600	60.3 (7.5)	24.6 (5.1)	-35.7 ^{**}	-7.1
7300	88.5 (13.6)	36.2 (8.0)	-52.4 ^{**}	-10.5
11300	123.3 (20.8)	58.2 (15.8)	-65.1 ^{**}	-13.0
----- Streeter -----				
470	5.5 (0.4)	4.1 (0.5)	-1.3	-0.3
970	9.1 (0.6)	7.4 (0.7)	-1.8	-0.4
2240	12.4 (0.9)	13.7 (1.7)	1.3	0.3
3600	14.8 (1.1)	17.9 (1.7)	3.1	0.6
7300	19.1 (1.5)	39.4 (6.5)	20.3 [*]	4.1
11300	25.3 (2.5)	50.6 (4.7)	25.3 ^{**}	5.1
----- Williston -----				
470	2.7 (0.2)	3.5 (0.3)	0.8	0.2
970	4.4 (0.3)	5.7 (0.5)	1.3	0.3
2240	5.7 (0.5)	9.9 (0.8)	4.2 ^{**}	0.8
3600	7.1 (0.6)	12.4 (0.9)	5.4 ^{**}	1.1
7300	11.6 (1.2)	23.9 (4.2)	12.2 ^{**}	2.4
11300	-- [¶]	--	--	--

[†] Equivalent soil mass of 470, 970, 2240, 3600, 7300, and 11300 Mg ha⁻¹ reflect approximate soil depths of 0-0.05, 0-0.1, 0-0.2, 0-0.3, 0-0.6, and 0-0.9 m depths, respectively.

[‡] Values in parentheses represent standard error of the mean.

^{*}, ^{**} Change from initial sampling significantly different at P≤0.05 and 0.01, respectively.

[¶] Sample not collected due to restrictive layer at 90 cm.

648 Figure 2

649