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Catherine E. Stewart
USDA-ARS, catherine.stewart@ars.usda.gov

Ronald F. Follett
USDA-ARS, ronald.follett@ars.usda.gov

Elizabeth G. Pruessner
USDA-ARS, elizabeth.pruessner@ars.usda.gov

Gary E. Varvel
USDA-ARS, gevarvel@windstream.net

Kenneth P. Vogel
USDA-ARS, kvogel1@unl.edu

See next page for additional authors

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N fertilizer and harvest impacts on bioenergy crop contributions to SOC

CATHERINE E. STEWART[1], RONALD F. FOLLETT[1]*, ELIZABETH G. PRUESSNER[1], GARY E. VARVEL[2]*, KENNETH P. VOGEL[3]* and ROBERT B. MITCHELL[3]

[1]Soil-Plant-Nutrient Research Unit, United States Department of Agriculture-Agricultural Research Service, Suite 100, 2150 Centre Avenue, Building D, Fort Collins, CO 80526-8119, USA, [2]Agroecosystems Management Research Unit, USDA-ARS, Univ. of Nebraska, 251 Filley Hall/Food Ind. Complex, Lincoln, NE 68583-0937, USA, [3]Grain, Forage, and Bioenergy Research Unit, USDA-ARS, Univ. of Nebraska, 251 Filley Hall/Food Ind. Complex, Lincoln, NE 68683-0937, USA

Abstract

Belowground root biomass is infrequently measured and simply represented in models that predict landscape-level changes to soil carbon stocks and greenhouse gas balances. Yet, crop-specific responses to N fertilizer and harvest treatments are known to impact both plant allocation and tissue chemistry, potentially altering decomposition rates and the direction and magnitude of soil C stock changes and greenhouse gas fluxes. We examined switchgrass (*Panicum virgatum* L.) and corn (*Zea mays* L.,) yields, belowground root biomass, C, N and soil particulate organic matter-C (POM-C) in a 9-year rainfed study of N fertilizer rate (0, 60, 120 and 180 kg N ha^{-1}) and harvest management near Mead, NE, USA. Switchgrass was harvested with one pass in either August or postfrost, and for no-till (NT) corn, either 50% or no stover was removed. Switchgrass had greater belowground root biomass C and N (6.39, 0.10 Mg ha^{-1}) throughout the soil profile compared to NT-corn (1.30, 0.06 Mg ha^{-1}) and a higher belowground root biomass C:N ratio, indicating greater recalcitrant belowground root biomass C input beneath switchgrass. There was little difference between the two crops in soil POM-C indicating substantially slower decomposition and incorporation into SOC under switchgrass, despite much greater root C. The highest N rate decreased POM-C under both NT-corn and switchgrass, indicating faster decomposition rates with added fertilizer. Residue removal reduced corn belowground root biomass C by 37% and N by 48% and subsequently reduced POM-C by 22% compared to no-residue removal. Developing productive bioenergy systems that also conserve the soil resource will require balancing fertilization that maximizes aboveground productivity but potentially reduces SOC sequestration by reducing belowground root biomass and increasing root and soil C decomposition.

Keywords: harvest timing, N fertilizer, residue removal, roots, soil C sequestration, soil fractions, soil organic C

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Introduction

Belowground plant biomass is the primary source of soil organic C and yet is rarely quantified and not dynamically represented in C cycle models. Roots can contribute up to 50% of total plant biomass to belowground C in perennial systems that can extend up to 3 m (Ma *et al.*, 2000; Garten *et al.*, 2010, 2011). Corn (*Zea mays*, L) can also have a deep root system that contributes 22–50% or more of aboveground biomass to belowground C as roots and root exudates (De Klein *et al.*, 2006; Johnson *et al.*, 2006). Belowground contributions from NT-corn and switchgrass (*Panicum virgatum* L.) help restore soil quality, reduce erosion potential and increase soil C stocks to marginally productive land (Jin *et al.*, 2015; Stewart *et al.*, 2015b). However, agricultural management practices such as N fertilizer, harvest timing and residue removal can subsequently alter plant allocation and belowground contributions to SOC (Garten *et al.*, 2010).

Annual crops have larger external nutrient requirements compared to perennials, which can remobilize C and N stored in rhizomes and roots (Wayman *et al.*, 2014). Agricultural management including N fertilization and harvest practices can alter plant nutrient allocation between aboveground and belowground plant organs and SOC stocks (Varvel *et al.*, 2008; Heggenstaller *et al.*, 2009; Follett *et al.*, 2012; Ontl *et al.*, 2013). Nitrogen fertilization increases aboveground plant productivity, but many perennial species allocate additional N to aboveground, rather than belowground growth, reducing belowground root production at high

*Retired.

Correspondence: Catherine E. Stewart, tel. 970 492-7270, fax 970 492-7213, e-mail: catherine.stewart@colostate.edu

N fertilizer rates (Heggenstaller *et al.*, 2009; Garten *et al.*, 2011). Switchgrass harvest removes most aboveground biomass and the timing can significantly alter nutrient translocation belowground (Vogel *et al.*, 2002). Harvesting switchgrass postfrost allows nutrient translocation from the aboveground biomass belowground and decreases N content of aboveground plant material, potentially providing better feedstock quality, but at the expense of the amount of harvested biomass (Vogel *et al.*, 2002; Wayman *et al.*, 2014). N availability can also change SOC stability through the decomposition or priming of existing SOC depending on litter chemistry. N fertilizer and residue quality are two primary controls of litter decomposition. N fertilization increases residue decomposition, but not in all cases (Craine *et al.*, 2007; Hobbie *et al.*, 2012). Under scenarios of N fertilization and higher soil N availability, residues with greater lignin content may be more completely decomposed, providing a lower overall contribution to soil C compared to residues with a lower lignin content (Stewart *et al.*, 2015a). In nutrient-limited situations, plants can increase root exudation, stimulating soil organic matter decomposition (Personeni & Loiseau, 2004; Shahzad *et al.*, 2015) for nutrient acquisition (i.e., Craine *et al.*, 2007). Species with a greater root N content could be expected to decompose and transition more quickly into soil C pools, including the more labile particulate organic matter (POM) fraction, which is comprised primarily of partially decomposed plant material. Crop species, N and residue management all influence the amount and turnover of SOC within these C pools (Camardella & Elliot, 1992).

In this study, we evaluate root-derived contributions to belowground C stocks and decomposition into the particulate organic matter soil C pool under potential cellulosic bioenergy crops no-till (NT) corn and switchgrass. Following conversion from conventionally tilled corn production, the soil profile (0–150 cm) SOC stocks increased under both crops after 9 years compared to baseline (Follett *et al.*, 2012). Here, we evaluate N fertilizer rate and harvest management treatment effects for both crops after 9 years. We hypothesized overall contributions to belowground C pools would be greater under switchgrass, but due to greater N content of root biomass, annual production and death of corn root biomass would provide a greater contribution to particulate organic matter.

Material and methods

Site and experimental design

The long-term switchgrass and maize experiment began in 1998 at the University of Nebraska's Agricultural Research and Development Center (ARDC) (41.151W 96.40N) (details in Varvel *et al.*, 2008; Follett *et al.*, 2012). The climate at the site is mesic with a mean annual temperature of 10.5 °C and mean annual precipitation of 765 mm. Field soils were Yutan silty clay loam (fine-silty, mixed, superactive, mesic Mollic Hapludalf) and Tomek silt loam (fine, smectitic, mesic Pachic Argiudoll).

The study was a randomized complete block split-split plot experimental design with two cultivars of switchgrass (Trailblazer and Cave-in-Rock) and NT-corn with three replicates. Due to stand deterioration in Trailblazer, we report data from Cave-in-Rock only.

Three N fertility treatments were randomly assigned within the main plots. Subplots were 30 m long × 18 m wide and are separated by 15 m wide alleys. From 2000 to 2007, N fertilizer rates were N1 = 0, N2 = 60, N3 = 120 and N4 = 180 kg N ha^{-1}. Rates on the switchgrass were N1, N2 and N3 and on NT-corn were N2, N3 and N4. The 0N rate for switchgrass was used as a low input treatment only for switchgrass. In 2001, the switchgrass and corn subplots were split lengthwise into 9 m wide sub-subplots for harvest treatments. The two harvest treatments for switchgrass were August or postfrost (PF) harvest date. The NT-corn stover was either removed at 50% (residue removed, or RR) or no residue was removed (NRR).

Aboveground biomass

Aboveground biomass harvesting methods for switchgrass and corn are described in detail in Varvel *et al.* (2008) and Follett *et al.* (2012). Briefly, for NT-corn, total aboveground dry matter production (stover + grain) was measured annually by hand-harvesting a 4.4 m long section of a nonborder, row from each plot after physiological maturity (September or early October). Ears were removed, dried and weighed. Stalks were cut at ground level, chopped, weighed, and a subsample was dried at 60 °C until constant mass. After dried ears were shelled, cob mass was added to dried stalk mass to obtain total stover production. Corn grain yields were determined with a plot combine equipped with a weighing unit that harvested the center three rows of each plot. In plots with no stover removal, all stover was left intact following grain harvest. In stover removed plots, stover was removed using a field flail harvester. We report only the 2006 aboveground C yields (grain + stover) to approximate a full accounting with belowground C stocks taken in the spring of 2007.

For switchgrass, the plots were harvested only once a year from a 0.9 to 1.8 m wide swath (varied with harvester used) the full 30 m length of the plots using flail harvesters and associated weighing equipment. At time of harvest, subsamples were collected from each subplot, weighed for moisture content, dried at 50 °C for 48 h and reweighed to determine dry matter content. Yields were adjusted to a dry weight basis. The C concentration of the switchgrass samples was determined using near-infrared spectrometer (NIRS) procedures and calibrations (Vogel *et al.*, 2011). Calibration data included samples from 2000 to 2004 from this study reported in Varvel *et al.* (2008) with a standard error of prediction for C of 4.49 (Vogel *et al.*, 2011). A field flail harvester was used to remove all remaining biomass from the plots immediately following the yield harvest using the same harvest height of 10 cm. We report averaged switchgrass yields from 1998 to 2007 here to integrate lower yields during crop establishment and to integrate extremely low aboveground biomass yields in some plots for 2006 due to drought conditions (Follett *et al.*, 2012). Precipitation was lower than average from 1998 to 2007 (Jin *et al.*, 2015), resulting in low biomass production particularly in 2006 (Follett *et al.*, 2012). Averages include planting year (1998) which reduced average yield.

Soil Sampling

Soils and belowground root biomass were sampled between April and May 2007 before corn planting. Plant material was removed from the soil surface and then, using a flat-bladed shovel, soils were undercut and removed from the 0–5, 5–10 and 10–30 cm depths to preserve soil structure for bulk density analyses. Samples were also collected from the 30–60, 60–90, 90–120 and 120–150 cm depths in May 2007 using a 5.08 cm hydraulic probe (Follett *et al.*, 2012). To minimize plot disturbance, one core was collected for each of the three field replicates between corn rows and switchgrass crowns. Soil bulk densities on all depths were determined using the clod method of the USDA-NRCS National Soils Laboratory methods (USDA-NRCS, 2004).

Soils were 2 mm sieved and all >2 mm plant material hand-picked from the soil, oven-dried (55 °C), subsampled, mechanically ground to pass through a 0.2 mm sieve and stored in glass containers until further analysis. All soils were checked for carbonates and in the very few cases where carbonates existed they were removed with a gentle wash in phosphoric acid prior to analyses for organic C using accepted procedures (Follett *et al.*, 1997). All analyses were on an oven dry weight. The methodology is such that both the isotopic C analyses and the analyses for the total SOC are done at the same time for the same sample.

Root separations, soil fractionation and C analyses

Roots were washed from the bulk soil samples from each depth increment (1 core per field replicate) using a hydropneumatic root elutriator (Smucker *et al.*, 1982). Between two and four replicate subsamples, 50 g of moist soil was washed on 250 μm sieves and the retained root material oven-dried at 55 °C. Dried roots were ground to pass through a 0.2 mm sieve for further analysis. These root measurements underestimate corn root biomass due to the spring sampling between corn rows and switchgrass crowns (Allmaras & Nelson, 1971; Johnson *et al.*, 2011), but represent an estimate of standing root stock.

Particulate organic matter (POM) was obtained using a modified method of Cambardella & Elliot (1992). Briefly, after the addition of 0.1 M sodium hexametaphosphate, soil samples were shaken 16 h and separated over a 53 μm sieve (POM > 53 μm). POM was oven-dried at 55 °C and ground to pass through a 0.2 mm sieve for C and N analyses.

Total C and N concentrations and δ^{13}C were determined using a Europa Scientific automated nitrogen carbon analyzer (ANCA-NT) coupled to a Europa 20-20 stable isotope analyzer continuous flow isotope ratio mass spectrometer (Europa Scientific Ltd., Crewe, England). Samples were run in duplicate with an analytical error of 0.14 ‰.

The δ^{13}C was calculated using the equation

$$\partial^{13}\mathrm{C} = \left[\frac{13R_{\mathrm{Sample}} - 13R_{\mathrm{Standard}}}{13R_{\mathrm{Standard}}}\right]1000$$

where R_{sample} is the ratio of $\delta^{13}\mathrm{C}/\delta^{12}\mathrm{C}$ in the soil, and R_{standard} is the ratio of $\delta^{13}\mathrm{C}/\delta^{12}\mathrm{C}$ international Pee Dee Belemnite (PDB).

We partitioned C3- and C4-derived C using the stable isotope mixing model approach (Balesdent & Balabane, 1992):

$$F = \frac{\delta_s - \delta_{c3}}{\delta_{c4} - \delta_{c3}}$$

where $\delta_s = \delta^{13}$C of the soil, $\delta_{C3} = \delta^{13}$C of C3 (−26.78) and $\delta_{C4} = \delta^{13}$C of the C4 vegetation (−12.94).

Statistics

Data were analyzed using a split-split plot design in Proc GLMIX in SAS (Cary, NC). Fixed main treatment effects were crop (corn vs. switchgrass), N(crop) and harvest(crop) within each depth. Replicate, replicate×plant and replicate×N(crop) were considered random effects. Differences between treatment means were estimated with predetermined comparisons using ESTIMATE statements (switchgrass 0N vs. 60N, 0N vs. 120N, 60N vs. 120N and August vs. Postfrost harvest; NT-corn 60N vs. 120N, 60N vs. 180N, 60N vs. 180N and residue retained vs. residue removed). We report least-squared means and standard errors with an n of 3. Differences with a P-value of <0.05 were considered significant, and significance level <0.10 noted in the text. T-tests were used to compare crop effects within N rate for aboveground and belowground C and N.

Results

Aboveground and belowground biomass, C and N

Aboveground biomass in switchgrass ranged from 3.13 to 11.20 Mg ha^{-1} (Table 1) and 8.30 to 12.43 Mg ha^{-1} for NT-corn (Table 2). Increasing N fertilizer rate increased aboveground biomass, and biomass C and N of both crops ($P < 0.0001$ and $P = 0.001$, Tables 1 and 2). Residue removal had no effect on aboveground C in NT-corn (Table 2). However, postfrost harvest increased switchgrass aboveground biomass C by 14% (Table 1, $P = 0.04$) and reduced aboveground biomass N by 33% compared to an August harvest (0.033 vs. 0.049 Mg N ha^{-1}). There was no significant difference between switchgrass and NT-corn in aboveground biomass C when compared within the same N rate for the 60 (switchgrass = 3.39 Mg C ha^{-1} vs. NT-corn

Table 1 Aboveground and belowground (0–150 cm) least-squared means for biomass (Mg ha^{-1}), C and N stocks (Mg C or N ha^{-1}) for switchgrass cultivar Cave-in-Rock under 0, 60 and 120 kg N ha^{-1} fertilizer rates and August or postfrost (PF) harvest treatments. Aboveground yields are averaged from 1998 to 2007. Belowground biomass is from spring 2007 only. Lowercase letters indicate differences between N treatments, and uppercase letters differences between harvest treatments. Italics indicate N or harvest treatment averages

		Switchgrass			
		0 kg N ha^{-1}	60 kg N ha^{-1}	120 kg N ha^{-1}	Harvest average
Aboveground biomass	August	3.36	7.07	9.25	*6.56 A*
	PF	3.13	8.27	11.20	*7.53 A*
	Avg	*3.24 a*	*7.67 b*	*10.22 c*	
Aboveground biomass C	August	1.46	3.12	4.08	*2.89 A*
	PF	1.38	3.67	5.04	*3.36 B*
	Avg	*1.42* a	*3.39* ab	*4.55* b	
Aboveground biomass N	August	0.02	0.04	0.07	*0.05 A*
	PF	0.01	0.03	0.07	*0.03 B*
	Avg	*0.02* a	*0.03* b	*0.07* c	
Belowground biomass	August	18.80	*23.15*	*20.17*	*20.70 A*
	PF	19.09	*30.68*	*27.69*	*23.31 A*
	Avg	*18.94 a*	*26.92* b	*20.17* ab	
Belowground biomass C	August	6.17	7.95	5.10	*6.41 A*
	PF	5.54	9.37	5.76	*6.89 A*
	Avg	*5.85* ab	*8.66* b	*5.43* a	
Belowground biomass N	August	0.07	0.09	0.11	*0.09 A*
	PF	0.06	0.12	0.13	*0.10 A*
	Avg	*0.07* a	*0.11* b	*0.12* b	

Table 2 Aboveground and belowground (0–150 cm) biomass (Mg ha^{-1}), biomass C and N (Mg C or N ha^{-1}) least-squared means for NT-corn under 60, 120 and 180 kg N ha^{-1} fertilizer and no residue removed (NRR) or residue removed (RR) harvest treatments in 2006. Aboveground yields are averaged from 2006. Belowground biomass is from spring 2007 only. Lowercase letters indicate difference between N treatments, and uppercase letters differences between harvest treatments. Italics indicate N or harvest treatment averages

		NT-Corn			
		60 kg N ha^{-1}	120 kg N ha^{-1}	180 kg N ha^{-1}	Harvest average
Aboveground biomass	NRR	9.51	12.26	12.43	*11.40 A*
	RR	8.30	11.17	12.18	*10.55 A*
	Avg	*8.91 a*	*11.71 b*	*12.30 b*	
Aboveground biomass C	NRR	3.72	4.42	5.28	*4.48 A*
	RR	3.43	4.60	5.03	*4.36 A*
	Avg	*3.58* a	*4.51* ab	*5.15* b	
Aboveground biomass N	NRR	0.07	0.11	0.14	*0.11 A*
	RR	0.06	0.10	0.14	*0.10 A*
	Avg	*0.06* a	*0.11* b	*0.14* c	
Belowground biomass	NRR	5.81	10.34	9.55	*8.57 A*
	RR	6.28	5.08	6.99	*6.12 A*
	Avg	*6.04 a*	*7.71 a*	*8.27 a*	
Belowground biomass C	NRR	1.31	1.69	1.90	*1.63 A*
	RR	1.20	1.00	1.36	*1.19 B*
	Avg	*1.25* a	*1.35* a	*1.63* a	
Belowground biomass N	NRR	0.05	0.08	0.09	*0.07 A*
	RR	0.05	0.04	0.06	*0.05 B*
	Avg	*0.05* a	*0.06* ab	*0.08* b	

3.58 Mg C ha^{-1}, $P = 0.787$) and 120 kg N ha^{-1} rates (switchgrass = 4.55 Mg C ha vs. NT-corn 4.51 kg N ha^{-1}, $P = 0.938$). However, NT-corn had 2.4 times more aboveground N stocks at 60 kg N ha^{-1} and 0.7 times more N stocks at 120 kg N ha^{-1} compared to switchgrass ($P = 0.01$, $P = 0.03$, respectively).

Under switchgrass, moderate N fertilizer (60 kg N ha^{-1}) maximized belowground root biomass C, 8.66 Mg C ha^{-1} (0–150 cm, Table 1) but at 120 kg N ha^{-1}, belowground root biomass C was the lowest of all treatments at 5.43 Mg C ha^{-1} (Table 1). There was no effect of increasing N fertilizer rate on belowground root biomass C of NT-corn (Table 2). N fertilizer increased belowground root biomass N stocks under both switchgrass ($P = 0.04$, Table 1) and marginally under NT-corn ($P = 0.098$, Table 2).

When compared within the same N rate, switchgrass had 6.6 and 4.0 times more belowground root biomass C compared to NT-corn in the 60 and 120 kg N ha^{-1} rate, respectively ($P = 0.001$, $P = 0.062$). Switchgrass had more belowground root biomass N (2.6 times) when compared within N rate to NT-corn in the 60 kg N ha^{-1} rate ($P = 0.007$), but not in the 120 kg N ha^{-1} rate ($P = .216$). Harvest (postfrost vs. August) had no effect on total belowground root C or N stocks (0-150 cm). Long-term residue removal in NT-corn reduced belowground root biomass C by 37% ($P = 0.022$) and N by 48% ($P = 0.018$) compared to no-residue removal (Table 2).

Depth distribution of belowground root biomass C and chemistry

Nitrogen and harvest effects on belowground root biomass C were predominantly observed in the 0–60 cm depths (Fig. 1). Moderate N fertilization (60 kg N ha^{-1}) enhanced switchgrass belowground root biomass C in the 10–30 and 30–60 cm depths (Fig. 1a). Increasing N fertilization had little effect on belowground root biomass C under NT-corn, except at 120–150 cm, where belowground root biomass C was greater under fertilizer treatments compared to the 60 kg N ha^{-1} fertilization rate ($P = 0.022$, Fig. 1b). Under switchgrass, postfrost harvest increased belowground root biomass C only in 10–30 cm depth ($P = 0.030$, Fig. 1c). Corn residue removal decreased root biomass C by 30% in the 0–30 cm depth ($P = 0.030$, Fig. 1d).

Belowground biomass from switchgrass roots had a much greater average C:N ratio (80.2) compared to NT-corn (26.0, $P > 0.049$), which increased with depth for both crops (Fig. 2a,b). Increasing N fertilizer rate decreased switchgrass root C:N, with the 120 kg N ha^{-1} rate significantly lower (53.2) than 0 or 60 kg N ha^{-1} (98.2 and 89.3, respectively) averaged over the soil profile ($P < 0.001$). The same was observed under NT-corn, with the C:N ratio under 180 kg N ha^{-1} significantly lower (26.2) than the 60 kg N ha^{-1} (33.4) averaged over the soil profile ($P = 0.006$). Postfrost harvest of switchgrass significantly decreased root C:N compared to August harvest in the 30–60 and 60–90 cm depths (Fig. 2c), but not averaged over the soil profile ($P = 0.497$). Residue removal did not affect C:N under NT-corn ($P = 0.467$, Fig. 2d).

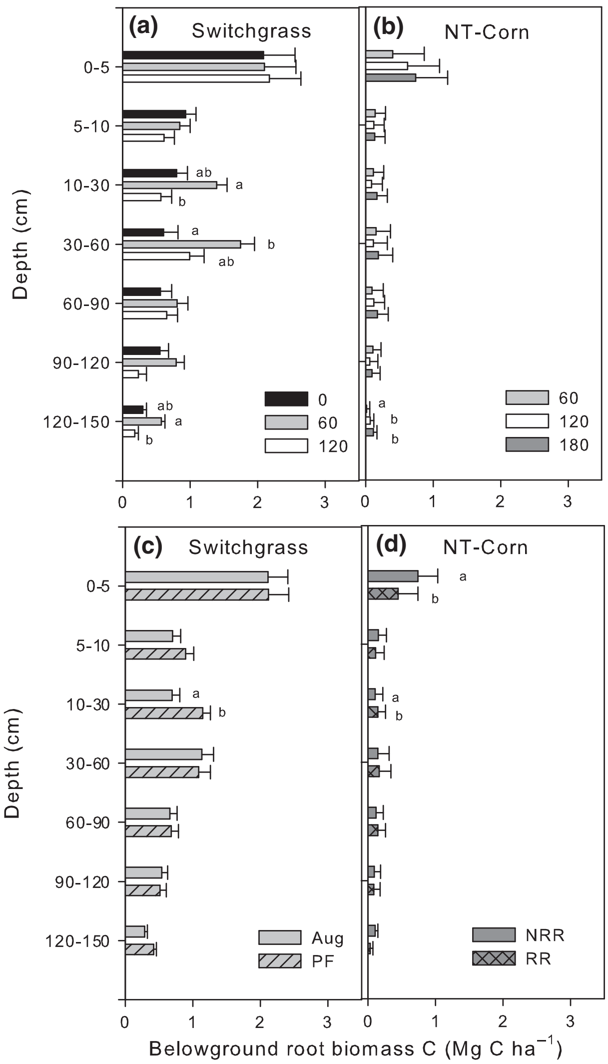

Fig. 1 Least-squared means of belowground root biomass C (Mg C ha^{-1}), for switchgrass under 0, 60 and 120 kg N ha^{-1} fertilizer (a) and August or postfrost harvest treatments (c) and NT-corn under 60, 120 and 180 kg N ha^{-1} fertilizer (b) and no residue removed (NRR) or residue removed (RR) harvest treatments (d). Lowercase letters indicate difference between N or harvest treatments within depth. Error bars represent standard errors ($n = 3$).

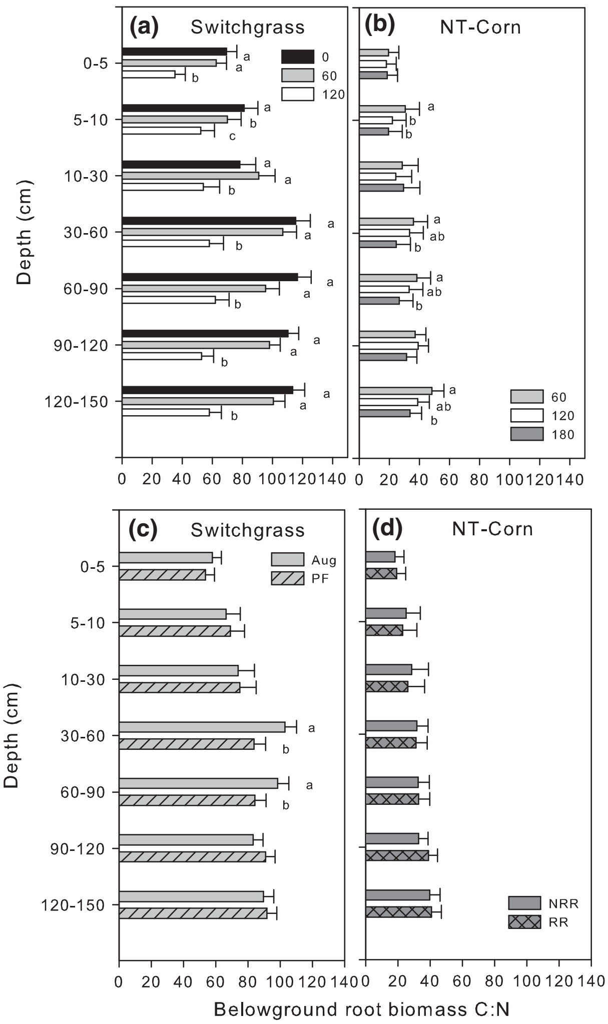

Fig. 2 Belowground root biomass C:N ratios least-squared means, for switchgrass under 0, 60 and 120 kg N ha^{-1} fertilizer (a) and August or postfrost harvest treatments (c) and NT-corn under 60, 120 and 180 kg N ha^{-1} fertilizer (b) and no residue removed (NRR) or residue removed (RR) harvest treatments (d). Lowercase letters indicate difference between N or harvest treatments within depth. Error bars represent standard errors (n = 3).

Particulate organic matter carbon

The majority of POM-C was observed in the 0–60 cm depths (93–95%) with most treatment differences observed in the 0–5 and 5–10 cm depths (Tables 3 and 4). However, these differences in many cases were large enough to drive soil profile (0–150 cm) changes in POM-C stocks. Surprisingly, there was no significant difference between switchgrass and NT-corn in POM-C (g POM C kg^{-1} soil) except for the 5–10 cm depth, where switchgrass had 69% more POM-C (P = 0.034, Table 3).

The middle N rate for both crops had the greatest profile (0–150 cm) POM-C (NT-corn, 120 kg N ha^{-1} 5.08 g POM-C kg^{-1} soil and switchgrass 60 kg N ha^{-1} 6.28 g POM-C kg^{-1} soil) and POM-C was lowest under the high N rate. These differences were driven by N treatment effects in the 0–5 and 5–10 cm depths for each crop. Switchgrass had the greatest POM-C at the 60 kg N ha^{-1} rate in both the 0–5 and 5–10 cm depths (3.61 and 1.53 g POM-C kg^{-1} soil) compared to the 120 N rate (2.08 and 1.21 g POM-C kg^{-1} soil) (P = 0.032, and 0.011, respectively). Under NT-corn, greater profile POM-C under the middle N rate (120 kg N ha^{-1}, 5.08 g POM-C kg^{-1} soil) compared to the 180 kg N ha^{-1} rate (3.62 g POM-C kg^{-1} soil, P = 0.035) was also due to smaller POM-C stocks in the 0–5 and 5–10 cm depths under the 180 kg N ha^{-1} rate.

Residue removal from NT-corn decreased POM-C in the 0–5 cm depth compared to no-residue removal (P = 0.047), resulting in a 22% decrease in POM-C throughout the profile (RR=3.79, NRR=4.89 g POM-C kg^{-1}soil, P = 0.0376, Table 3). Switchgrass postfrost harvest increased POM-C compared to August harvest in the top three depths although it was only significant in the 10–30 cm depth (P = 0.007), a trend that continued for POM-C throughout the soil profile (P = 0.071, Table 3). Switchgrass had greater POM C:N compared to NT-corn in the top three depths (Table 5).

Belowground root biomass C contributions to SOC

The POM-C stocks under both NT-corn and switchgrass were proportional to belowground root biomass C with R2 = 0.72 for NT-corn and R2 = 0.70 for switchgrass (Fig. 3). However, NT-corn had a three times greater conversion to POM-C per unit root biomass compared to switchgrass (Fig. 3).

Discussion

Perennial bioenergy crops contribute substantially more biomass belowground compared to annual crops, can sequester more soil C and, consequently, have a greater greenhouse gas offset potential compared to conventional corn (Davis *et al.*, 2011). This study illustrates two contrasting scenarios of plant allocation, decomposition and incorporation into soil organic matter. Switchgrass produced more belowground root biomass C (5.5–9.3 Mg C ha^{-1}) with a greater C:N ratio compared to NT-corn (1.2–1.9 Mg C ha^{-1}) with a lower C:N ratio, reflecting known differences between crops in belowground root biomass production and tissue chemistry (Johnson *et al.*, 2007; Garten *et al.*, 2011; Ontl *et al.*, 2013). This confirmed our hypothesis that overall contributions to belowground C pools would be greater

Table 3 Least-squared means for C4-POM (g C kg^{-1} soil) for switchgrass harvested in August or in postfrost (PF) under three N treatments (0, 60, 120 kg N ha^{-1}) and NT-corn with (RR) or without residue removal (NRR) under three N treatments (60, 120, 180 kg N ha^{-1}). Bold values indicate significant main effects or contrasts between N fertilizer rates or harvest within crop. Italics indicate N or harvest treatment averages

Plant	N	Harvest	0–5 cm	5–10 cm	10–30 cm	30–60 cm	60–90 cm	90–120 cm	120–150 cm	0–150 cm
			g C4-POMC kg^{-1} soil							
Switchgrass	0	August	2.49	1.02	0.47	0.20	0.10	0.06	0.04	4.38
		PF	3.70	1.38	0.59	0.27	0.15	0.08	0.06	6.23
		0 Average	*3.10*	***1.20***	*0.53*	*0.23*	*0.12*	*0.07*	*0.05*	5.31
	60	August	3.00	1.42	0.55	0.28	0.13	0.09	0.07	5.53
		PF	4.21	1.63	0.74	0.22	0.11	0.07	0.06	7.04
		60 Average	***3.61***	***1.53***	*0.65*	*0.25*	*0.12*	*0.08*	*0.07*	**6.28**
	120	August	2.26	1.37	0.80	0.13	0.07	0.07	0.05	4.75
		PF	1.90	1.05	0.85	0.15	0.11	0.07	0.05	4.19
		120 Average	***2.08***	***1.21***	*0.83*	*0.14*	*0.09*	*0.07*	*0.05*	**4.47**
	August Average		*2.59*	*1.27*	***0.61***	*0.20*	*0.10*	*0.07*	*0.05*	*4.89*
	Postfrost Average		*3.27*	*1.36*	***0.73***	*0.21*	*0.12*	*0.07*	*0.06*	*5.82*
Switchgrass average			2.93	**1.31**	0.67	0.21	0.11	0.07	0.06	5.35
NT-Corn	60	NRR	3.82	0.64	0.28	0.15	0.11	0.07	0.05	5.12
		RR	2.01	0.68	0.33	0.24	0.12	0.06	0.04	3.50
		60 Average	*2.92*	***0.66***	*0.31*	*0.20*	*0.12*	*0.07*	*0.05*	4.31
	120	NRR	3.89	0.96	0.44	0.21	0.13	0.07	0.05	5.76
		RR	2.75	0.88	0.47	0.13	0.11	0.03	0.03	4.41
		120 Average	***3.32***	***0.92***	*0.46*	*0.17*	*0.12*	*0.05*	*0.04*	**5.08**
	180	NRR	2.30	0.72	0.31	0.21	0.13	0.06	0.06	3.78
		RR	2.07	0.75	0.26	0.20	0.08	0.06	0.03	3.46
		180 Average	***2.19***	***0.74***	*0.28*	*0.20*	*0.10*	*0.06*	*0.05*	**3.62**
	NRR Average		***3.34***	*0.78*	*0.34*	*0.19*	*0.12*	*0.07*	*0.05*	***4.89***
	RR Average		***2.28***	*0.77*	*0.35*	*0.19*	*0.10*	*0.05*	*0.03*	***3.79***
NT-Corn Average			*2.81*	***0.77***	*0.35*	*0.19*	*0.11*	*0.06*	*0.04*	*4.34*

Table 4 Main effects, interactions and effect contrasts for C4-POM (g C kg^{-1} soil) for each depth and the soil profile (0–150 cm) for switchgrass harvested in August or postfrost under three N treatments (0, 60, 120 kg N ha^{-1}) and NT-corn with or without residue removal under three N treatments (60, 120, 180 kg N ha^{-1}). Bold values indicate significant main effects or contrasts between N fertilizer rates or harvest within crop. Italics indicate $P < 0.10$

Effect	Num DF	Den DF	0–5 m	5–10 cm	10–30 cm	30–60 cm	0–150 cm
					P-value		
Crop	1	2	0.770	**0.034**	0.148	0.716	0.230
N(Crop)	4	8	0.108	**0.019**	0.644	0.426	**0.043**
Harvest(Crop)	2	12	*0.066*	0.559	**0.023**	0.885	**0.031**
N*harvest(Crop)	4	12	0.426	**0.037**	0.485	*0.073*	0.229
SG 0 vs. 60			0.413	**0.010**	0.615	0.809	0.129
SG 0 vs. 120			0.123	0.933	0.212	0.146	0.186
SG 60 vs. 120			**0.032**	**0.011**	0.429	0.100	**0.014**
Corn 60 vs. 120			0.511	**0.026**	0.519	0.728	0.218
Corn 60 vs. 180			0.248	0.448	0.918	0.893	0.265
Corn 120 vs. 180			*0.089*	*0.090*	0.457	0.631	**0.035**
Switchgrass harvest August vs. Postfrost			0.179	0.291	**0.007**	0.640	*0.071*
NT-corn residue removal			**0.048**	0.978	0.748	0.898	**0.038**

under switchgrass due to greater belowground root biomass. Our measured belowground root biomass underestimated the actual NT-corn root production and reflected a root litter pool due to the spring sampling. Measured root biomass does not account for rhizodeposition, which can contribute an additional 2.5–6 times

Table 5 Least-squared means for the C:N ratio of POM under switchgrass and corn averaged across N rate and harvest. Asterisks indicate significant differences between switchgrass and NT-corn at the 0.05 level

Depth (cm)	Switchgrass C:N	Corn
0–5	18.5	15.3*
5–10	17.8	12.8*
10–30	15.1	10.0*
30–60	11.9	10.2
60–90	10.2	9.7
90–120	8.8	7.1
120–150	7.5	5.6

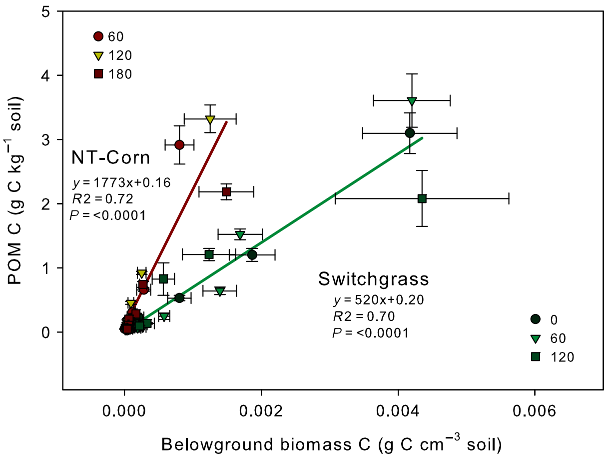

Fig. 3 POM C (g C kg^{-1} soil) as a function of belowground root biomass C (g C cm^{-3} soil) for NT-corn at 60, 120 and 180 kg N ha^{-1} and switchgrass at 0, 60 and 120 kg N ha^{-1}. Error bars represent standard errors (n = 3).

root biomass C to belowground C stocks (Molina *et al.*, 2001).

Despite smaller belowground root biomass C stocks under NT-corn, C was incorporated into the POM fraction three times more per unit of root biomass resulting in no difference in POM-C stocks between the two crops when averaged over the 150 cm soil profile. Faster decomposition of annual corn belowground root biomass into SOC would explain the similar SOC gains under switchgrass and NT-corn after conversion from conventional tillage corn at this site (Follett *et al.*, 2012).

Agricultural management affects switchgrass allocation

Agricultural management significantly altered plant allocation and belowground productivity under switchgrass. The N fertilizer application rate of 120 kg N ha^{-1} maximized aboveground productivity (4.5 Mg C ha^{-1}), but minimized belowground root biomass C (5.4 Mg C ha^{-1}). A moderate fertilizer rate of 60 kg N ha^{-1} maximized belowground root biomass C (8.7 Mg C ha^{-1}); a pattern that has been observed in other studies, although at higher N fertilizer rates. Garten *et al.* (2011) found fertilizer application of 67 kg N ha^{-1} maximized root biomass measured in April (19 Mg ha^{-1}), but was not greater than biomass under the highest N rate of 202 kg N ha^{-1}. Heggenstaller *et al.* (2009) found that a N application at 140 kg N ha^{-1} maximized root production and root N content in Cave-in-Rock in Iowa compared to a N application rate of 220 kg N ha^{-1}. However, Ma *et al.* (2000) found that N fertilizer did not impact belowground root productivity at 112 and 224 kg N ha^{-1} in AL.

Both N fertilizer and harvest timing altered belowground root biomass distribution within the soil profile. Greater belowground root biomass under the 60 kg N ha^{-1} treatment was due to an increase in the 10–60 cm depth, suggesting increased belowground production for additional nutrient acquisition. Garten *et al.* (2011) found increasing N demand under higher rates of N fertilization (67 and 202 kg N ha^{-1}). Reduced belowground root biomass under the 120 compared to the 60 kg N ha^{-1} rate suggests allocation of more N to aboveground compared to belowground productivity, a well-documented finding consistent with other studies (Heggenstaller *et al.*, 2009; Garten *et al.*, 2011). Although there was less belowground biomass under the 120 kg N ha^{-1} rate, the switchgrass root C:N ratio decreased by half compared to the 0N treatment (98.2 vs. 53.2), an effect also observed in other belowground studies (Ma *et al.*, 2000; Garten *et al.*, 2011). Fertilization with 202 kg N ha^{-1} reduced root C:N ratio (55.1 averaged over live and dead 0–90 cm) compared to 0 kg N ha^{-1} (107.6) under 5-years-old Alamo switchgrass grown in Tennessee (Garten *et al.*, 2011). The C:N ratio was lower under 224 kg N ha^{-1} (33) compared to 0 or 112 kg N ha^{-1} (59) under 3-years-old Alamo switchgrass plants in Alabama (0–3 m, Ma *et al.*, 2000).

Switchgrass harvest after senescence can significantly reduce aboveground nutrient content, providing a better biofuel feedstock (Vogel *et al.*, 2002; Johnson *et al.* 2014) and allowing nutrient recycling (Zegada-Lizarazu *et al.*, 2012). Postfrost harvest increased aboveground biomass C (3.3 vs. 2.9 Mg C ha^{-1}) and decreased aboveground biomass N (43 to 33 kg N ha^{-1}). Although we did not observe harvest effects on total belowground root biomass C or N, switchgrass harvested postfrost produced greater belowground root biomass, and 50% more N compared to August harvest in the 10–30 cm depth, suggesting additional C and N storage belowground occurs during senescence. Nutrient storage occurs in crowns (Heggenstaller *et al.*, 2009; Garten *et al.*, 2010, 2011), but our data illustrate that root biomass is also important for N storage. Cumulative C and N storage from root turnover also contributes to soil C stocks. Garten *et al.* (2011) found that dead fine roots increased from 5 to 13% of total plant biomass C from April to November indicating substantial mortality over the growing season. Increased root turnover in addition to greater root biomass under the postfrost harvest treatment likely contributed to the observed greater POM-C stocks in the 10–30 cm depth.

Harvest and N treatments that maximized belowground root biomass incorporated more belowground root biomass C into the soil C pool after 9 years. The moderate fertilizer rate of 60 kg N ha^{-1} maximized POM-C in the top two depths. Postfrost harvest resulted in 18% more POM-C on average compared to August harvest. The POM under switchgrass had a greater C:N ratio in the top 30 cm compared to NT-corn indicating the incorporation of more recalcitrant switchgrass-derived C into soil organic matter. The high belowground productivity of switchgrass roots is the primary reason switchgrass can sequester soil C and improve soil quality (Follett *et al.*, 2012; Stewart *et al.*, 2015b). These results show agricultural management practices that maximize aboveground productivity, such as higher N fertilization, may not maximize SOC sequestration through changes in plant allocation.

Root litter decomposition and N effects

Despite switchgrass having 5 times more belowground root biomass compared to NT-corn, there were only slight differences between switchgrass and NT-corn in POM-C confirming our hypothesis that the annual production and death of corn root biomass would provide a greater contribution to particulate organic matter. This difference suggests slower nutrient cycling under switchgrass with the accumulation of perennial belowground root biomass and under corn, faster and more complete decomposition of belowground root biomass. Residue decomposition generally decreases with increasing C:N ratio and switchgrass had more than double the C:N ratio compared to NT-corn. The relatively low C:N ratio of corn belowground root biomass measured here compared to other studies (Johnson *et al.*, 2007) is probably due to decomposition from fall harvest to early spring sampling. Faster decomposition of higher N residues is well documented (Hobbie, 2005; Johnson *et al.*, 2007; Adair *et al.*, 2008; Cornwell *et al.*, 2008).

N fertilizer effects on soil C storage are a function of indirect effects on C inputs and decomposition. The highest rate of fertilizer application decreased surface POM-C under both NT-corn and switchgrass. This reduction is most likely due to increased decomposition with additional N fertilizer, because belowground root biomass showed no significant reduction with N fertilizer in the surface depths. However, the highest rate of N addition decreased belowground root biomass C:N, which could further accelerate belowground root biomass decomposition. Initial decomposition rate is frequently positively related to residue N content because microbial substrate use can be N limited at low substrate N concentrations (Hobbie *et al.*, 2012). Subsequent analyses at this site found no difference in NT-corn POM-C stocks across N fertilizer rates (Osborne *et al.*, 2014; Jin *et al.*, 2015). However, differences between our results and theirs likely stem from our values only including C4 POM-C from isotope partitioning, which would identify a smaller C pool from recent crop input.

Residue removal effects in NT-corn

Although residue removal did not significantly affect aboveground productivity, we show reduced belowground root biomass C (37%) and N (48%) relative to no-residue removal which resulted in a profile-wide decrease in POM-C. Continued reductions in belowground root biomass C and N input could lead to reduced C and N stocks. Jin *et al.* (2015) also found no impact of residue removal on corn yields from 1998 to 2011 on these plots, although earlier samplings did find a small reduction in yield (Varvel *et al.*, 2008). They suggest that soil moisture conservation under the no residue removed treatment during the drought conditions during the early part of the study boosted yields compared to residue removal treatments. Halvorson & Stewart (2015) found a decrease in soil C and no N accrual after 7 years of residue removal in irrigated NT-corn in the semiarid Great Plains despite an increase in grain yield compared to no-residue removal. These results confirm that deterioration in soil quality under residue removal can be evident without crop yield declines (Jin *et al.*, 2015; Stewart *et al.*, 2015b), although grain yield declines under residue removal have been well documented (Wilhelm *et al.*, 2004) .

The reduction in belowground root biomass was primarily observed in the surface depth (0–5 cm) and corresponded to a reduction in aggregate stability, soil microbial biomass and soil quality (Jin *et al.*, 2015; Stewart *et al.*, 2015b). Roots act as nucleation sites for the formation of soil aggregates (Denef *et al.*, 2004; Six *et al.*, 2006) and are the primary source of belowground C in NT systems (Johnson *et al.*, 2006). Many authors document a decrease in soil C pools (aggregates and POM) as a result of stover removal that has typically been assumed to be a result of reduced aboveground C inputs (Blanco-Canqui, 2013; Osborne *et al.*, 2014; Jin *et al.*, 2015). Additional losses of C inputs through reduced root production could further deplete the soil resource.

Belowground C and SOC stocks

Belowground C stocks are important to quantify and this study illustrates two contrasting scenarios that have the potential for substantial belowground contributions to SOC (Follett *et al.*, 2012). Annual NT-corn produced belowground root biomass C proportional to aboveground biomass C with a low C:N ratio and a relatively quick decomposition trajectory into SOC. Under switchgrass, perennial roots built substantial belowground root biomass with a higher C:N ratio and slower decomposition and incorporation as POM-C. Despite these differences in C and N cycling, NT-corn had a greater conversion to POM-C per unit root biomass such that after 9 years, switchgrass POM-C stocks were similar to NT-corn (Table 3). These results may explain why no differences in SOC sequestration between NT-corn and switchgrass were observed in the soil profile after the initial 9 years (Follett *et al.*, 2012). Aboveground biomass production can be a good predictor of SOC sequestration, but previous work on this site found relatively poor relationships with aboveground biomass production (R^2 = 0.39) (Follett *et al.*, 2012). Adding belowground root biomass C increased R^2 of SOC change (0–30 cm, from baseline) to 0.79 for NT-corn and 0.97 for switchgrass (Fig. 4).

N fertilizer can modify nutrient cycling through changing aboveground and belowground biomass allocation patterns and accelerating decomposition directly and indirectly. A moderate fertilizer application (60 kg N ha^{-1}) maximized switchgrass belowground root biomass production and POM-C. Higher fertilization rates minimized belowground production and C:N ratio, potentially increasing root decomposition, resulting in the lowest POM-C stocks. Nitrogen fertilization had no effect on the belowground biomass C of NT-corn but the highest N rate (180 kg N ha^{-1}) decreased POM-C. Plant allocation patterns and tissue chemistry will have long-term impacts on nutrient cycling in these systems and are rarely investigated over fertility gradients (Garten *et al.*, 2011).

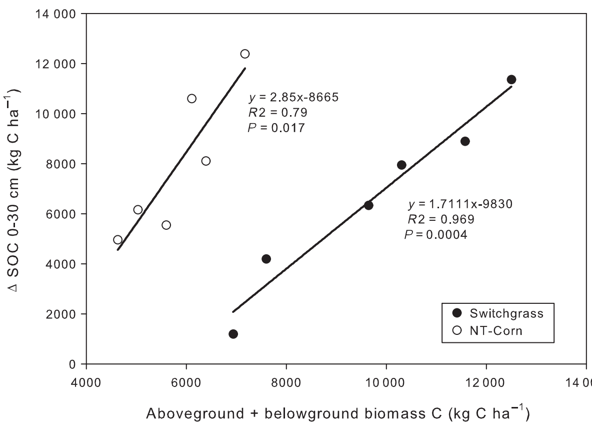

Fig. 4 Change in SOC (kg C ha^{-1}) in the surface soils (0–30 cm) from baseline (1998) to 2007 as a function of plant (aboveground + belowground) biomass C (kg C ha^{-1}).

Modeling simulations using the DAYCent model across the Corn Belt region show substantial ecosystem service benefits could accrue from planting perennial bioenergy species on marginal land, including up to a 473% reduction in greenhouse gas emissions and 22% reduction in N leaching compared to conventional corn production (Davis *et al.*, 2011). Despite the potential of perennial bioenergy crops such as switchgrass to contribute to belowground biomass and SOC sequestration, agricultural management such as harvest timing and N fertilization will moderate this effect. Simulation models will need to incorporate feedbacks between plant allocation, belowground biomass chemistry and decomposition to accurately predict land-use effects from bioenergy production.

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References

Adair EC, Parton WJ, Del Grosso SJ *et al.* (2008) Simple three-pool model accurately describes patterns of long-term litter decomposition in diverse climates. *Global Change Biology*, **14**, 2636–2660.

Allmaras RR, Nelson WW (1971) Corn (*Zea mays* L.) root configuration as influenced by some row-interrow variants of tillage and straw mulch management. *Soil Science Society of America Journal*, **35**, 974–980.

Balesdent J, Balabane M (1992) Maize root-derived soil organic carbon estimated by natural 13C abundance. *Soil Biology and Biochemistry*, **24**, 97–101.

Blanco-Canqui H (2013) Crop residue removal for bioenergy reduces soil carbon pools: how can we offset carbon losses? *BioEnergy Research*, **6**, 358–371.

Cambardella CA, Elliott ET (1992) Particulate soil organic-matter changes across a grassland cultivation sequence. *Soil Science Society of America Journal*, **56**, 777–783.

Cornwell WK, Cornelissen JHC, Amatangelo K *et al.* (2008) Plant species traits are the predominant control on litter decomposition rates within biomes worldwide. *Ecology Letters*, **11**, 1065–1071.

Craine JM, Morrow C, Fierer N (2007) Microbial nitrogen limitation increases decomposition. *Ecology*, **88**, 2105–2113.

Davis SC, Parton WJ, Grosso SJD, Keough C, Marx E, Adler PR, DeLucia EH (2011) Impact of second-generation biofuel agriculture on greenhouse-gas emissions in the corn-growing regions of the US. *Frontiers in Ecology and the Environment*, **10**, 69–74.

De Klein C, Novoa RSA, Ogle S *et al.* (2006) N2O emissions from managed soils, and CO2 emissions from lime and urea application. *Guidelines for national greenhouse gas inventories* pp. 1–54. Institute for Global Environmental Strategies, Hayama, Japan. Available at: http://www.ipcc-nggip.iges.or.jp (accessed 20 Nov. 2015).

Denef K, Six J, Merckx R, Paustian K (2004) Carbon sequestration in microaggregates of no-tillage soils with different clay mineralogy. *Soil Science Society of America Journal*, **68**, 1935–1944.

Follett RF, Paul EA, Leavitt SW, Halvorson AD, Lyon D, Peterson GA (1997) Carbon isotope ratios of great plains soils and in wheat-fallow systems. *Soil Science Society of America Journal*, **61**, 1068–1077.

Follett RF, Vogel KP, Varvel GE, Mitchell RB, Kimble J (2012) Soil carbon sequestration by switchgrass and no-till maize grown for bioenergy. *BioEnergy Research*, **5**, 866–875.

Garten CT, Smith JL, Tyler DD *et al.* (2010) Intra-annual changes in biomass, carbon, and nitrogen dynamics at 4-year old switchgrass field trials in west Tennessee, USA. *Agriculture Ecosystems & Environment*, **136**, 177–184.

Garten CT, Brice DJ, Castro HF *et al.* (2011) Response of "Alamo" switchgrass tissue chemistry and biomass to nitrogen fertilization in West Tennessee, USA. *Agriculture, Ecosystems & Environment*, **140**, 289–297.

Halvorson AD, Stewart CE (2015) Stover removal affects no-till irrigated corn yields, soil carbon, and nitrogen. *Agronomy Journal*, **107**, 1504–1512.

Heggenstaller AH, Moore KJ, Liebman M, Anex RP (2009) nitrogen influences biomass and nutrient partitioning by perennial, warm-season grasses. *Agronomy Journal*, **101**, 1363–1371.

Hobbie SE (2005) Contrasting effects of substrate and fertilizer nitrogen on the early stages of litter decomposition. *Ecosystems*, **8**, 644–656.

Hobbie SE, Eddy WC, Buyarski CR, Adair EC, Ogdahl ML, Weisenhorn P (2012) Response of decomposing litter and its microbial community to multiple forms of nitrogen enrichment. *Ecological Monographs*, **82**, 389–405.

Jin VL, Schmer MR, Wienhold BJ *et al.* (2015) Twelve years of stover removal increases soil erosion potential without impacting yield. *Soil Science Society of America Journal*, **79**, 1169–1178.

Johnson JMF, Allmaras RR, Reicosky DC (2006) Estimating source carbon from crop residues, roots and rhizodeposits using the national grain-yield database. *Agronomy Journal*, **98**, 622–636.

Johnson JMF, Barbour NW, Weyers SL (2007) Chemical composition of crop biomass impacts its decomposition. *Soil Science Society of America Journal*, **71**, 155–162.

Johnson JMF, Archer DW, Weyers SL, Barbour NW (2011) Do mitigation strategies reduce global warming potential in the Northern U.S. corn belt? *Journal of Environmental Quality*, **40**, 1551–1559.

Johnson JMF, Gresham GL (2014) Do Yield and Quality of Big Bluestem and Switchgrass Feedstock Decline over Winter? *BioEnergy Research*, **7**, 68–77.

Ma Z, Wood CW, Bransby DI (2000) Impacts of soil management on root characteristics of switchgrass. *Biomass and Bioenergy*, **18**, 105–112.

Molina JAE, Clapp CE, Linden DR, Allmaras RR, Layese MF, Dowdy RH, Cheng HH (2001) Modeling the incorporation of corn (*Zea mays* L.) carbon from roots and rhizodeposition into soil organic matter. *Soil Biology and Biochemistry*, **33**, 83–92.

Ontl TA, Hofmockel KS, Cambardella CA, Schulte LA, Kolka RK (2013) Topographic and soil influences on root productivity of three bioenergy cropping systems. *New Phytologist*, **199**, 727–737.

Osborne S, Johnson JF, Jin V, Hammerbeck A, Varvel G, Schumacher T (2014) The Impact of Corn Residue Removal on Soil Aggregates and Particulate Organic Matter. *BioEnergy Research*, **7**, 559–567.

Personeni E, Loiseau P (2004) How does the nature of living and dead roots affect the residence time of carbon in the root litter continuum? *Plant and Soil*, **267**, 129–141.

Shahzad T, Chenu C, Genet P, Barot S, Perveen N, Mougin C, Fontaine S (2015) Contribution of exudates, arbuscular mycorrhizal fungi and litter depositions to the rhizosphere priming effect induced by grassland species. *Soil Biology and Biochemistry*, **80**, 146–155.

Six J, Frey SD, Thiet RK, Batten KM (2006) Bacterial and fungal contributions to carbon sequestration in agroecosystems. *Soil Science Society of America Journal*, **70**, 555–569.

Smucker AJM, McBurney SL, Srivastava AK (1982) Quantitative separation of roots from compacted soil profiles by the hydropneumatic elutriation system. *Agronomy Journal*, **74**, 500–503.

Stewart C, Moturi P, Follett R, Halvorson A (2015a) Lignin biochemistry and soil N determine crop residue decomposition and soil priming. *Biogeochemistry*, **124**, 335–351.

Stewart CE, Follett RF, Pruessner EG, Varvel GE, Vogel KP, Mitchell RB (2015b) Nitrogen and harvest effects on soil properties under rainfed switchgrass and no-till corn over 9 years: implications for soil quality. *GCB Bioenergy*, **7**, 288–301.

USDA-NRCS (Natural Resources Conservation Service) (2004) Soil survey laboratory methods manual. Soil Survey Investigations Report No. 42. Version 4.0(ed. Burt R). Update Available at http://soils.usda.gov/technical/lmm/ http://www.nrcs.usda.gov/wps/portal/nrcs/main/soils/ref/ (accessed 3 April 2012, 11 September 2015)

Varvel GE, Vogel KP, Mitchell RB, Follett RF, Kimble JM (2008) Comparison of corn and switchgrass on marginal soils for bioenergy. *Biomass and Bioenergy*, **32**, 18–21.

Vogel KP, Brejda JJ, Walters DT, Buxton DR (2002) Switchgrass biomass production in the Midwest USA: harvest and nitrogen management. *Agronomy Journal*, **94**, 413–420.

Vogel K, Dien B, Jung H, Casler M, Masterson S, Mitchell R (2011) Quantifying actual and theoretical ethanol yields for switchgrass strains using NIRS analyses. *BioEnergy Research*, **4**, 96–110.

Wayman S, Bowden R, Mitchell R (2014) Seasonal changes in shoot and root nitrogen distribution in switchgrass (*Panicum virgatum*). *BioEnergy Research*, **7**, 243–252.

Wilhelm WW, Johnson JMF, Hatfield JL, Voorhees WB, Linden DR (2004) Crop and soil productivity response to corn residue removal. *Agronomy Journal*, **96**, 1–17.

Zegada-Lizarazu W, Wullschleger SD, Nair SS, Monti A (2012) Crop physiology. In: *Switchgrass a valuable biomass crop for energy* (ed Monti A), pp. 55–86. Springer, London.