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Soil carbon stock impacts following reversion of Miscanthus x giganteus and short rotation coppice willow commercial plantations into arable cropping.

Running Title: Bioenergy crop reversion impacts on soil C

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Abstract

There are posited links between the establishment of perennial bioenergy, such as Short Rotation Coppice (SRC) willow and Miscanthus x giganteus, on low carbon soils and enhanced soil C sequestration. Sequestration provides additional climate mitigation, however, few studies have explored impacts on soil C stocks of bioenergy crop removal, thus the permanence of any sequestered C is unclear. This uncertainty has led some authors to question the handling of soil C stocks with carbon accounting e.g. through LCA. Here we provide additional data for this debate, reporting on the soil C impacts of the reversion (removal and return) to arable cropping of commercial SRC willow and Miscanthus across four sites in the UK, two for each bioenergy crop, This article has been accented for publication and undergone full peer review but has not been

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with 8 reversions nested within these sites. Using a paired-site approach, soil C stocks (0-1 m) were compared between 3 and 7 years after bioenergy crop removal. Impacts on soil C stocks varied, ranging from an increase of 70.16 ± 10.81 Mg C ha⁻¹ 7 years after reversion of SRC willow to a decrease of 33.38 ± 5.33 Mg C ha⁻¹ 3 years after reversion of Miscanthus compared to paired arable land. The implications for carbon accounting will depend on the method used to allocate this stock change between current and past land use. However, with, published life cycle assessment values for the lifetime C reduction provided by these crops ranging from 29.50 to 138.55 Mg C ha⁻¹, the magnitude of these changes in stock are significant. We discuss the potential underlying mechanisms driving variability in soil C stock change, including the age of bioenergy crop at removal, removal methods, and differences in the recalcitrant of the crop residues, and highlight the need to design management methods to limit negative outcomes.

Introduction

Bioenergy crops are seen as key resource in the move towards decarbonised energy systems in many parts of the world (Bauer et al., 2018) including the UK (Committee on Climate Change, 2019; Government, 2017; HM Goverment, 2017). Providing a fungible low carbon (C) alternative to fossil fuels, with potential applications in many energy systems including hard to decarbonise sectors such as transport (Government, 2017; HM Goverment, 2017). The primary climate mitigation mechanism for bioenergy crops is the use of the above ground biomass as a replacement for fossil fuels, especially if this can be linked to C capture, utilisation and storage (Committee on Climate Change, 2019; The Royal Society & Royal Academy of Engineering, 2018). In the case of perennial bioenergy crops, there has also been substantial interest in the potential for additional C savings through enhanced soil C sequestration (Qin, Dunn, Kwon, Mueller, & Wander, 2016; Rowe et al., 2016; Whitaker et al., 2018).

Short rotation coppice (SRC) willow and *Miscanthus x giganteus*here after referred to as Miscanthus) are leading perennial bioenergy crops in Europe (Rowe et al., 2016; Walter, Don, & Flessa, 2015). Research has shown that when established on low C soil with less than 60–70 Mg C ha⁻¹ at 0–30 cm depth, these crops have the potential to increase soil C stocks (Don et al., 2012; Harris, Spake, & Taylor, 2015; Rowe et al., 2016; Whitaker et al., 2018). Levels of C sequestration depend on a range of abiotic and biotic factors. Crop age has been positively

associated with soil C sequestration whilst soil type ultimately influences the capacity of soil to accrue C (Harris et al., 2015; Qin et al., 2016; Tiemann & Grandy, 2015). Declines in soil C stocks may also occur if these crops are established on high C soils, such as often found under permanent grassland or forest (Qin et al., 2016; Richards et al., 2017; Rowe et al., 2016)

Previous studies on soil C impacts of perennial bioenergy crop establishment have provided valuable information for policy makers and industry in regard to best practice for the expansion of bioenergy cropping (Committee on Climate Change, 2018). This work has allowed for the inclusion of soil C impacts of bioenergy crop establishment into life cycle assessments (LCA) (Clarke, Sosa, & Murphy, 2019; Parajuli et al., 2017; Whittaker, Macalpine, Yates, & Shield, 2016) and improvements in the modelling C impacts of land use change into bioenergy (Davis et al., 2011; Harper et al., 2018; Richards et al., 2017). This inclusion assumes, however, a degree of permanence to the changes in soil C stock which may not be fully justifiable (Smith, 2004; Whittaker et al., 2016). As noted by Whittaker et al. (2016), calculating the full life cycle soil C budget for these bioenergy crops must include hotspots in the management cycle, including soil C stock changes following crop removal.

Across Europe, SRC willow and Miscanthus removal has occurred due to crops reaching the end of their productive life, in response to reductions in end-user market demand, changes in subsidy regimes and agronomic challenges (see (Bryden, 2019; Helby, Rosenqvist, & Roos, 2006; Mawhood, Slade, & Shah, 2015). Removal may be followed by replanting but reversion to conventional agricultural cropping is common (Welc, Lundkvist, Nordh, & Verwijst, 2017). Whitaker et al., (2016) hypothesised that such reversions could result in a loss of soil C stocks and with negative impacts on the overall C budget.

Despite the need to quantify soil C stock change across the full life cycle of perennial bioenergy crops, there are limited data regarding impacts of their removal or subsequent replacement. Available data falling into two types, comparisons to soil C stock under the bioenergy crop or comparison to the original land use before the bioenergy crop was established (Table S1). These comparisons provide different insights, with comparisons to bioenergy soil C stock isolating the impacts of the reversion process and post removal management, whilst comparisons to the original land use provide an assessment of the impacts on soil C stocks of the full bioenergy crop cycle,

establishment, cultivation, reversion and post reversion management (For discussion see - De Palma et al., 2018). For SRC willow and Miscanthus, only a single study exists for each crop where a paired-site approach has been used to compare to the original arable land use (Dufossé, Drewer, Gabrielle, & Drouet, 2014; Kahle, Möller, Baum, & Gurgel, 2013) (Table S1). These two studies both report higher surface soil C stocks (≤ 40 cm) one year following bioenergy crop reversion compared to the arable land, with soil C stocks being 7.90 and 29.13 Mg C ha⁻¹ higher for SRC willow and Miscanthus reversions, respectively (Table S1). Both studies highlighted that as soil C stock change can be slow to respond to land use change, there is a need for longer term studies (Dufossé et al., 2014; Kahle et al., 2013). Longer term data are available for two SRC poplar reversions and one SRC willow reversion where soil sampling was conducted prior to reversion and then repeated one and four years after, with comparison made to the pre-reversion soil C stock (Toenshoff, Joergensen, Stuelpnagel, & Wachendorf, 2013; Wachendorf, Stuelpnagel, & Wachendorf, 2017) (Table S1). There were no significant changes in soil C stocks at any of these sites one year post reversion, with stocks similar to the pre-reversion soil samples. However, after four years, soil C stocks were significantly reduced in one SRC poplar site (Table S1) thus supporting calls for longer term studies (Dufossé et al., 2014; Kahle et al., 2013).

The majority of existing studies have been conducted on small experimental plots. Whilst experimental studies are valuable, they may not fully reflect how crops are removed during commercial operations, as this management is difficult to replicate within smaller experimental plots (McCalmont et al. 2018). To address the paucity of data quantifying soil C stock change following the end of life removal of commercial bioenergy crops, we conducted a soil survey using a paired-site approach and incorporating sites from across England. Soil sampling was conducted to 1 m depth, to examine changes in C stocks through the soil profile in these deep rooting crops. As these sites were commercial the type of paired-sites available were constrained resulting in two objectives. First, where a bioenergy crop paired land use was available the objective was to provide data on the impact of bioenergy crop removal through comparison to the remaining paired bioenergy crop. Second, where a paired field of the original land use was available the objective was to provide data on the soil C impact of bioenergy crop cultivation from planting through to and including reversion back to the original land use. In addition, to set the magnitude of the soil carbon impacts in broader context we also provide a comparison between the

life cycle soil carbon impacts of bioenergy cultivation and published Life Cycle Assessment (LCA) values for the C offset provided by these crops.

Methods

Site selection and paired-site approach

In cooperation with commercial companies involved in the planting, management and removal of SRC willow and Miscanthus, previously reverted sites across the UK were identified using set selection criteria. Prior to crop removal, sites were required to have been (i) commercial-scale bioenergy plantations, in a commercially productive condition (e.g. not abandoned or in poor condition); (ii) plantations that were not on reclaimed land where topsoil replacement would confound soil C measurements; (iii) fully established at time of removal (> 4 yrs.) and (iv) with availability of paired land use(s) for the comparisons of impact, consisting ideally of both the original land use and a bioenergy crop. A paired site approach was required as these were not available. The paired site approach is commonly used and following best practice, land owners were consulted to facilitate the selection of the most appropriate paired fields for matching the bioenergy reversions with respect to soil type and prior management (De Palma et al., 2018; Rowe et al., 2016).

Using these criteria four suitable sites were identified of SRC willow (2) and Miscanthus (2) within which eight reversions were nested (Table 1). The availability of paired land uses (original arable land use, bioenergy crop) varied between sites (Table 1). Arable land use pairs were present at both the SRC willow reversion sites, but in these cases SRC willow was not present. The Nottingham Miscanthus reversion site had both a Miscanthus and arable land use. Miscanthus land use was also available at the second Miscanthus reversion site in Taunton but no arable pairing (Table 1).

Formal records of removal processes were not available, however, recollections and estimated dates were collated from the landowners. Miscanthus sites were all *Miscanthus x giganteus* established as rhizomes. In the year of removal all fields were harvested as normal in early spring. Following this, for the Nottingham and Taunton Mis -4 yrs. reverted fields, a modified potato harvester was used to harvest rhizomes large enough for resale. This process removed only a

portion of the rhizome from the field although data on fraction of rhizome remaining was not available it was sufficient for some crop regrowth to occur. In Taunton all the Miscanthus reversion fields were allowed to regrow in the spring to approximately 1 m before being sprayed with 4.5L/ha Roundup Bio (Monsanto Europe, Belgium) followed by a second application of 2L/ha about 6 weeks later. The resulting crop was then flail mowed, allowed to dry, baled and removed. The fields were then deep ploughed and left until September before planting a sacrificial crop, followed by standard spring planting. In the Nottingham site herbicide (Glyphosate) was applied earlier to new Miscanthus shoot springs. Fields were then subsoiled (to 30 cm), cultivated and sown. In the willow sites the mix of willow cultivars was unknown, however both sites were established by the same commercial company who reported a routine mix of five cultivars consisting of ~ 30% Tora and equal proportion (20% each) drawn from three of Ulv, Olof, Jorunn or Jorr, and a small amount (10%) of Bowles Hybrid. Removal at all sites occurred after standard winter harvest of the above ground biomass, with the application of herbicide (Glyphosate roundup) to new shoots in early spring. The Gainsborough site was left fallow until June - July before being disked to break up the stools with a double set of disc harrows (front set of mark 3 Sibma Disc Harrows disks with lighter "standard farm set" following to provide weight). Fields were again left fallow until being planted with autumn crops in the September of the same year. In the Doncaster site a more intensive approach was applied, to allow more rapid return to arable cropping, with site mulched with a forest mulcher, ploughed, power-harrowed and disked before being planted with Maize in April.

Sibilet wer the crop beir Table 1: Details of study sites, including soil type and land use, year of crop establishment, removal and years since reversion at time of sampling. All bioenergy crops were established on and returned to arable cropping.

Site name/ nearest town	Land use	Planting year	Age of bioenergy crop at removal (yrs.)	Time since reversion (yrs.)	Land use code	Current land use	Soil Type *	Mean BD 0–30 cm (g cm ⁻³)	Mean pH 0–30 cm	Mean annual temp (°C) 2000 - 2017	Mean annual rainfall (mm) 2000 - 2017
Doncaster	Reverted SRC	2000	13	3	SRC -3 yrs.	Maize for	821b	1.09 ± 0.45	7.06 ± 0.44		
	Reverted SRC	2000	10	6	SRC -6 yrs.	production of cobs **	Blackwood- deep sandy &	1.11 ± 0.56	6.76 ± 0.17	10.5	632
	Arable	Pre-1990	-	-	Arable	Arable rotations	coarse loamy soil	1.15 ± 0.29	7.05 ± 0.19		
Gainsborough	Reverted SRC	2000	10	5	SRC -5 yrs.	Arable rotations	532b Romey-	1.12 ± 0.27	7.39 ± 0.23		
	Reverted SRC	2000	8	7	SRC -7 yrs.	with Silage maize	coarse and fine silty soil with	1.08 ± 0.23	7.12 ± 0.00	10.3	644
	Arable	Pre-1990	-	-	Arable	Arable rotations	areas of Adventurer 2 1024b Fen peat	1.13 ± 0.18	8.24 ± 0.01		
Nottingham	Reverted Miscanthus	2007	6	3	Mis -3yrs.	Arable rotations	551a Bridgnorth-	1.30 ± 0.39	6.77 ± 0.19		741
	Miscanthus	2007	-	-	Mis	Arable rotations	sandy & coarse	1.44 ± 0.33	6.90 ± 0.18	10.1	
	Arable	Pre-1990	-	-	Arable	Arable rotations	loamy soils	1.30 ± 0.37	6.85 ± 0.22		

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Taunton A	Reverted Miscanthus	2006	6	4	Mis -4 yrs.	Arable rotations with Silage	572d/c	1.35 ± 0.27	7.93 ± 0.22		
	Reverted Miscanthus	2006	7	3	Mis -3 yrs.	maize, wheat and barley	Middleton/Hodn et Reddish fine	1.16 ± 0.28	7.65 ± 0.24	10.9	734
	Miscanthus	2006	-	-	Mis	Miscanthus	silty soils & reddish fine	1.24 ± 0.23	7.79 ± 0.12		
Taunton B	Reverted Miscanthus	2007	5	4	Mis -4 yrs. B	Alfalfa	coarse loamy	1.16 ± 0.35	7.89 ± 0.28		
	Miscanthus	2007	-	-	Mis B	Miscanthus	soils	1.30 ± 0.26	7.49 ± 0.39		

* Soil Survey of England and Wales, Rothamsted Research, 1983, Soil Maps 1:2500.

** In contrast silage maize, where all of the above ground biomass (AGB) is removed, at this only the maize cob were removed with all remaining above ground biomass, was returned to the soil.

Soil sampling:

Soil in each field was sampled using a spatial hierarchical design (see Rowe et al., 2016)in order to capture in field variability in soil C stocks across both small and large spatial scales (Figure S1). Five sampling plots per field were randomly selected from intersections of a grid overlaid on a map of the cropped area of field (Figure S1). The resolution of the grid was adjusted to ensure that there were a minimum of 50 grid intersections, with the resolution of the grid not being less than 5 m. A 20 m perimeter buffer was employed to reduce potential edge effects.

Within each of the five sampling plots, three within-plot soil cores were taken using a split-tube soil sampler (Eijkelkamp Agrisearch Equipment BV, Giesbeek, The Netherlands) with an inner diameter of 4.8 cm to a depth of 30 cm. The first core was taken at the grid intersect, with two further cores taken at distances of 1 m and 1.5 m in random compass directions from the intersect (Figure S1). Cores were sectioned into 10 cm increments (0–10, 10–20 and 20–30 cm) in-field and placed into individually labelled bags. This gave a total of 15 spatially nested cores per field.

At three randomly selected sampling plots the 30 cm coring was extended to 1 m by using a window sampler system with a 4.4 cm cutting diameter (Eijkelkamp Agrisearch Equipment BV, Giesbeek, The Netherlands). Immediately after sampling, the window sampler was placed with the hole generated by the 30 cm sampling and coring conducted to a depth of 1 m (giving a 70 cm soil core). If coring to 1 m was not possible, for example due to large stones or bedrock, the depth of the cored hole was recorded.

For each land use a total of 15 surface soil cores (0–30 cm) and nine 1 m cores were taken. At Gainsborough site laboratory inspection showed some of the cores had been taken over a strip of buried fen peat running through the east side of all three adjacent sampled fields. The buried fen peat layer, with high C content, varied in location (from 20 cm and deeper) and in its thickness (20 cm to 70 cm) within affected soil profiles, thus potentially masking impacts of the land use. Therefore, affected cores were removed from the analysis (Table 2)

 Table 2: Numbers of cores removed, reason for removal are given in

 footnote and number of remaining cores are given in parentheses

Site	Land use code	Number of core	e removed
She	Land use code	(remaining core	es)
		0–30 cm	0—1 m
Doncaster	Arable	0 (15)	0 (9)
	SRC -3 yrs.	0 (15)	2* (7)
	SRC -6 yrs.	0 (15)	0 (9)
Gainsborough	Arable	9** ⁽⁶⁾	3** (6)
	SRC -5 yrs.	6** ⁽⁹⁾	3** (6)
-	SRC -7 yrs.	9** (6)	6** (6)
Nottingham	Arable	0 (15)	1* (8)
	Mis	3*** (12)	0 (9)
	Mis -3yrs.	0 (15)	8*
Taunton A	Mis A	0 (15)	0 (9)
	Mis -3 yrs.	0 (15)	0 (9)
	Mis -4 yrs.	0 (15)	0 (9)
Taunton B	Mis B	0 (15)	0 (9)
	Mis -4 yrs. B	0 (15)	0 (9)

*Reduced numbers as cores that did not reach 1 m in field sampling have been removed; see methods.

** Reduced cores due to removal of cores taken in areas with buried fen peat; see methods.

*** Three 0–30 cm cores taken at one of the five sampling points were identified as outliers in the statistical analysis; see text for details.

Laboratory processing

On return to the laboratory the 1 m soil cores were divided into 10 cm increments. Where compression of the core had occurred during sampling the length of the individual sections were reduced to account for the compression, following Rowe et al., (2016) and Walter et al., (2015). All 30 cm and 1 m cores were assessed for fine earth soil mass, moisture content, pH and C content (%).

pH analyses were undertaken to identify soil which may contain inorganic C. Sub-samples (~ 5 g) from each core section were bulked within sampling plot to give a single composite sample per depth. Samples were sieved to 4 mm, mixed at a 1:2.5 weight:volume ratio with deionised water

and left to stand for 30 minutes before measurement using a pH meter (Hanna pH 210 Meter, Hanna Instruments Ltd, Bedfordshire, UK).

The remaining soil, minus a 20 g subsample taken for -80 °C archiving, was weighed, air-dried at 25 °C and sieved to 2 mm with the mass and volume of stones and roots remaining on the sieve recorded. A subsample of the sieved soil (15–18 g) was oven-dried (105 °C for 12 hours) and moisture loss recorded. Mass of each core section was then calculated using values of moisture loss and stone and root volume following methods in the GB Countryside Survey (Reynolds et al., 2013) giving soil mass value corrected to represent the fine earth proportion (Schrumpf, Schulze, Kaiser, & Schumacher, 2011).

For determining soil C concentration, the oven-dried subsample of soil was ground in a ball mill (Fritsch Planetary Mill). For all sites except the reverted willow at the Gainsborough site, pH values (≥ 8) or soil maps suggested the possible presence of inorganic C. To remove any inorganic C present in the samples, 100–200 mg of each oven dried sample was weighed into silver cups (5076, Elemental Microanalysis, Okehampton, UK) with greater mass being used for samples deeper in the profile with lower C concentration, thereby ensuring C concentrations remained within detection range, and 100 µl of 0.5 M HCL was applied. If CO₂ evolution was observed using a binocular microscope at 70× magnification then up to two additional 100 µl applications of 0.5 M HCl were made. If CO₂ evolution continued following the third application, samples were dried to 50 °C and the process was repeated until evolution ceased. Samples were then wrapped in tin cups to aid combustion and C concentration measured using an elemental analyser (Leco Truspec CN, Michigan, USA). Ground soil from the reverted willow at the Gainsborough sites were weighed directly into tin cups, wrapped and C content measured as described above.

Soil C stock calculations

The soil C concentration and core dry mass were used to calculate soil C stock on an equivalent soil mass (ESM) basis, using a reference dry soil mass of 3 Gg ha⁻¹, and 13 Gg ha⁻¹ for the 0–30 cm and 0–1 m soil layer, respectively, following equation 1 in Gifford & Roderick (2003). Reference masses were selected based on the mean dry soil mass of cores across the four sites.

Equation 1:

$$SC_{ESM} = SC_{upper} + (Conc_{Lower}(M_{ref} - M_{upper}))$$

 SC_{ESM} is the soil C stock based on the selected equivalent soil mass (Mg C ha⁻¹), SC_{upper} is the C stock (Mg C ha⁻¹) of the upper soil C section, $Conc_{Lower}$ is the C concentration of the lower layer (% C), M_{ref} is the reference mass selected (Mg ha⁻¹) and M_{upper} is the mass of the upper core sections (Mg ha⁻¹). In the 0–30 cm summed values of the 0–10 cm and 10–20 cm were used as the upper section, and 20–30 cm as the lower section. In the 1 m cores summed values for the 0–80 cm were used for the upper section, C concentration was based on the mean % C from the 80–90 cm and 90–100 cm sections. Cores that did not reach a minimum of 90 cm depth (due to stones or bedrock) were removed for the analysis.

Life cycle comparison

Climate mitigation potential C offset values (the C emission offset or avoided through the replacement of fossil fuels with bioenergy) for SRC willow and Miscanthus were taken from UK specific bioenergy crop studies (Brandão, Milà i Canals, & Clift, 2011; McCalmont et al., 2018; Robertson et al., 2017; Whittaker et al., 2016) (Table S2). Values were converted to Mg C offset ha⁻¹, using values for conversion efficiencies, biomass energy content, transportation and cultivation related emissions, reported in each study but excluding any changes in soil C stocks. Values were normalised to a common crop life span of 15 and 20 years and yield of 10 and 11 oven dry Mg ha⁻¹ for SRC willow and Miscanthus, respectively, based on reported values (Djomo, Kasmioui, & Ceulemans, 2011; McCalmont et al., 2018; Whittaker et al., 2016). These values were compared to the crop cycle soil C stock change at the Doncaster, Nottingham and Gainsborough sites; calculated as the difference between the soil C stock in the reverted bioenergy crop and the arable land use available at these three sites. This accounted for the combined impact of the bioenergy crop establishment, cultivation and reversion.

Statistical analysis

Sites were analysed separately as differences in soil type, available paired land uses, time since reversion, and age of bioenergy crops at removal confounded combined analyses. This includes separate analyses of the data for the land uses in Taunton A and Taunton B due to differences in the date of Miscanthus establishment (Table 1).

Two methods were used to compare soil C stocks between land uses (bioenergy crop, removed bioenergy crop, arable) and thus the inferred impact of bioenergy crop removal. Impacts on overall C stock (Mg C ha⁻¹) based on ESM calculations for the surface soil (0–30 cm) and deeper soil (0–1 m) were examined using mixed effect models with the *nlme* package in the R statistical program (J Pinheiro, D Bates, S Debroy, D Sarkar, 2013). Land use was entered as a fixed effect with sampling core nested within plot as a random effect in all models. The significance of these models was examined using a likelihood ratio test between a null model, including only random terms, and the chosen models with fixed terms. Where variance of land uses were unequal, a weighted variance structure was applied using the *VarIdent* function in the *nlme* package (J Pinheiro, D Bates, S Debroy, D Sarkar, 2013). At the Nottingham site within the Miscanthus field, 0–30 cm data from one of the five in field sampling plots were removed before analysis as outliers following visual inspection of the model residual plots. Mean 0–30 cm soil C stock in this plot of 51.39 ± 1.28 Mg C ha⁻¹ being over 10 Mg C ha⁻¹ greater than the 0–30 cm means for the other plots within this land uses (Table 3). This reduced the total 0–30 cm core number from 15 to 12, but the three 1 m samples were retained in the analysis.

Soil C stock depth profiles for the land uses at each site were also compared and differences tested using a bootstrapped Loess regression (Keith, Henrys, Rowe, & McNamara, 2016). Briefly, soil C stock (Mg C ha⁻¹) was derived for each 10 cm soil section and plotted against matching cumulative soil mass for the full sampling depth of 1 m. Within each site the data for reverted bioenergy crop(s) was then systematically compared to each of the available paired land use. Data for both land uses were grouped before being randomly sampled with replacement. This bootstrapped data was used to produce upper and lower 95 % confidence intervals that represents a null model (i.e. no difference between depth profiles) for the regression of soil C stock and cumulative soil mass. These confidence intervals are compared to the Loess regression line representing individual land uses, with significant change being inferred where the single land use regression falls outside of the confident intervals produced for the combined data. Details of this method are given in Keith et al., (2016). As sites had more than two land uses, this process was repeated for all paired comparisons within a site.

In some cases, for example in arable land use where tillage reduced bulk density, total cumulative core mass (0-1 m) were inconsistent between land uses. In these cases the 0-1 m cumulative soil mass of the lightest of the two land use pairs was calculated and data points greater than this value in the remaining land use were removed prior to analysis. This ensured that confidence intervals for the null hypothesis were calculated using data points from both land uses for the full core mass. Plotting non-incremental soil C stocks against cumulative soil mass allows assessment of differences between land uses on a continuous soil profile without the need for testing of multiple depth increments.

Results:

Total C stocks (ESM) SRC willow removal

At the Doncaster site, soil C stocks were affected by land use in both the surface soil ($\chi_{(2)} = 93.06$ p <0.001, 0–30 cm ESM) and over the full 1 m sampling depth ($\chi_{(2)} = 21.31$ p <0.001, 0–1 m ESM) (Table 3). In surface soil, C stocks were highest in the field which was SRC willow 6 years prior to sampling (SRC -6 yrs.) followed by the SRC -3 yrs. and lowest in the arable (Table 3). The 0–1 m soil C stock was higher than the arable land use in the SRC -6 yrs., but not in the SRC - 3 yrs. (Table 3)

At Gainsborough, surface soil C stocks were also impacted by land use ($\chi_{(2)} = 12.76$, p = 0.002). Surface soil C stocks were again higher in the reverted SRC willow compared to the arable land uses, although there was no significant differences between the two reverted SRC willow plantations (Table 3). Over 0–1 m, soil C stocks were not significantly different between land uses ($\chi_{(2)} = 0.126$, p = 0.93) (see Table S3 for soil C stock change).

Miscanthus removal

Soil C stocks at Nottingham were affected by land use in both the 0–30 cm ($\chi_{(2)} = 44.96$, p <0.001) and 0–1 m depth increments ($\chi_{(2)} = 26.92$, p <0.001). In both 0–30 cm and 0–1 m increments, and in contrast to the SRC willow sites, total soil C stocks were higher in the arable land use than in the Mis -3yrs, with C stocks being highest in the arable followed by the Mis, then the reverted Mis

-3yrs. (Table 3). In contrast to the SRC willow sites, differences in soil C stock between the arable the Mis and Mis -3yrs. increased with depth (Table 3 & Table S3)

At the Taunton site, a paired arable land use was not available and comparisons were made to remaining Miscanthus crops (Mis A & Mis B). In both Taunton A & B, land use affected total soil C stock in the 0–30 cm ($\chi_{(2)} = 63.68$, p = 0.001 and $\chi_{(1)} = 5.463$, p = 0.019, respectively) and 0–1 m depth increments ($\chi_{(2)} = 7.59$, p = 0.022 and $\chi_{(2)} = 4.781$, p = 0.0288 respectively). Post-hoc testing confirmed that, as with the Nottingham site, soil C stock in the Mis A and Mis B were higher than those in the respective paired reverted Miscanthus fields for both the 0–30 cm and 0–1 m depth increments (Table 3). Soil C stocks in the 0–30 cm depth increment in Taunton A were also higher in the field with Mis -3 yrs. prior to sampling than in the Mis-4 yrs., although not to 1 m where soil C stocks were similar in the two land uses (Table 3).

Table 3: Mean soil C stock \pm standard error within the study sites. Values for soil C stock based on ESM reference mass of 3 and 13 G Mg ha⁻¹ for 0–30 cm and 0–1 m depth respectively. Different letters indicate significant differences (p < 0.05) following post-hoc testing of main effect of land use for comparisons within site and depth.

Site	Land use code	Soil C stock based on ESM (Mg C ha ⁻¹)			
		0–30 cm	0–1 m		
Doncaster	Arable	$62.72 \pm 1.37^{\mathbf{a}}$	$124.22\pm4.70^{\mathbf{a}}$		
	SRC -3 yrs.	$93.68 \pm 2.87^{\text{b}}$	$131.21\pm4.54^{\mathtt{a}}$		
	SRC -6 yrs.	$141.57\pm4.75^{\circ}$	$194.38\pm8.95^{\text{b}}$		
Gainsborough	Arable	101.47 ± 2.65^{a}	257.76 ± 14.73*		
	SRC -5 yrs.	$126.60\pm2.72^{\text{b}}$	256.95 ± 20.32^{a}		
	SRC -7 yrs.	$121.02\pm8.90^{\text{b}}$	$263.84\pm7.26^{\textbf{a}}$		
Nottingham	Arable	39.46 ± 1.36^{a}	88.62 ± 4.16^{a}		
	Mis	33.08 ± 2.20^{b}	68.99 ± 3.50^{b}		
	Mis -3yrs.	$27.96\pm0.84^{\text{c}}$	$55.24\pm3.42^{\rm c}$		
Taunton A	Mis A	69.70 ± 2.96^{a}	103.15 ± 5.66^{a}		
	Mis -3 yrs.	$55.58 \pm 1.20^{\text{ b}}$	83.97 ± 1.47^{b}		
	Mis -4 yrs.	$42.31 \pm 1.34^{\circ}$	$85.22\pm3.00^{\text{b}}$		
Taunton B	Mis B	65.66 ± 2.83 ª	103.55 ± 9.85^{a}		
	Mis -4 yrs. B	57.62 ± 1.23 b	78.75 ± 4.52^{b}		

SRC Willow reversions

In the Doncaster site the higher soil C stock in the SRC willow surface soil compared to the arable land use reported in the ESM analysis are clearly apparent in the depth profiles (Fig 1). Both SRC willow reversions have reduced or similar soil C stocks at depth (i.e. at soil mass greater than 5000 Mg ha⁻¹) compared to the arable (Fig 1). This is more marked in the SRC -3 yrs. in which ESM soil C stock (0–1 m) is similar between the arable and the reverted SRC (Table 3). In the SRC -6 yrs. whilst a similar pattern is apparent the higher surface soil C stock extents to a slightly greater depth (Fig 1 c) reflecting the significantly higher soil C stock (ESM) reported over the fully 0–1 m C for this land use when compared to the arable land use (Table 3).

Fig 1

At the Gainsborough SRC willow reversion site, as with the Doncaster site in the upper sections of the profile, soil C stocks are higher in both the reverted SRC willow compared to the arable pairs (Fig 2a and b). In deeper soil, C stocks are either comparable or, in SRC -7 yrs., significantly lower than in the arable; this supports the ESM analysis where soil C stock are higher under the reverted SRC willow plantations in the 0-30 cm but not the 0-1 m depth increment.

Fig 2

Miscanthus reversions

The depth profile for the Mis -3yrs. at Nottingham shows lower soil C stocks in the surfaces soil in comparison to both the arable and Mis land uses (Fig 3). At greater depth, soil C stock is comparable and thus differences in the upper soil profile ($\sim 0-40$ cm) are responsible for the

significant overall changes in the soil C stocks to 1 m reported in the ESM analysis (Fig 3, Table

Fig 3

3)

At the Taunton sites impacts of bioenergy crop reversion on soil C stocks are greatest in the upper soil profile (Fig 4). Within the Taunton A sites, differences in soil C stock between the land uses rapidly decreased with depth becoming non-significant below a soil mass of 400 Mg ha⁻¹, equivalent to a depth of around 40 cm (Fig 4 a and b). In Taunton B whilst soil C stock difference between the reverted Mis -4yrs. B and Mis B again became smaller with increasing soil depth, significant differences between the soil C stocks extended further down the soil profile (Fig 4 c).

Fig 4

LCA comparison

In the Gainsborough site and in the Doncaster SRC -3yrs., crop cycle changes in soil C stock to 1 m calculated through comparison to the paired arable land uses were small (- 0.78 ± 25.10 to 6 .11 ± 22.08 C Mg ha⁻¹ Table S3), resulting in limited impacts on the predicted LCA C offsets based on UK specific bioenergy studies (Fig 5). In contrast, soil C stocks were 70.16 \pm 10.81 Mg C ha⁻¹ higher for SRC -6 yrs. than in the arable land use in the Doncaster site, more than double the lowest predicted fossil offset of 29.50 Mg C ha⁻¹ for offsetting use of heating oil (Brandão et al., 2011) (Fig 5, Table S3). In the Nottingham site, soil C stocks were 33.38 \pm 5.33 Mg C ha⁻¹ lower in the Mis -3 yrs. compared to the arable. This C loss is sufficient to negate the estimated lowest fossil fuel C offset of 29.50 Mg C ha⁻¹ (Robertson et al., 2017) and reduce the highest estimated C offset for replacement of coal from 104.40 Mg C ha⁻¹ to 71.02 Mg C ha⁻¹ (Table S2, Fig 5).

Fig 5

Discussion

This work addressed knowledge gaps in understanding of the impacts of bioenergy cropping and the reversion to arable cropping on soil C to 1 m depth. There was evidence of reversion impacts on soil C stocks in surface soil (0–30 cm) when comparisons were made to either an arable land use or a bioenergy crop. When compared to arable land uses, changes persisted at depth (1 m) in two of the five reverted bioenergy fields sampled. In these two cases, the Doncaster reverted SRC -7 yrs. and the Nottingham reverted Mis -3 yrs., diverging impacts on soil C stock were observed. Higher C stocks (70.15 \pm 10.11 Mg C ha⁻¹) in the reverted SRC willow field compared to the arable land use were double the fossil fuel offset derived from the use of the above ground biomass for bioenergy. In contrast, soil C stocks in the reverted Miscanthus field were 33.38 \pm 5.34 Mg C ha⁻¹ lower than the arable land use, a value greater than the lowest predicted C offset. These results provide valuable data to aid the discussion on handling dynamic soil C stocks within C accounting.

Factors influencing soil C impacts

In studies such as this one where commercial sites are utilised, multiple factors can influence soil C stocks. Perennial bioenergy reversion studies also require consideration of impacts occurring over long timeframes. Utilising the variability between sites within this study, we consider the potential role of some key factors; legacy of previous crop, post-reversion management, residue stability, the age of the bioenergy prior to reversion, soil type and the method of removal.

Legacy of previous crop - SRC willow

Soil C stocks were higher in the surface soil of all the reverted SRC willow fields in comparison to the arable land use. This is consistent with Kahle et al., (2013) who also reported higher soil C stock one year following SRC willow reversion compared to a paired arable land use. Absolute changes in surface soil C stocks (Mg C ha⁻¹) in this study where higher than in Kahle et al. (2013) though in the Gainsborough site the relative change in soil C stock was similar, being 26 % for the Kahle et al., (2013) study and 19 and 24 % in this study (Table S1 & S3). These findings collectively suggest potential for increased C stocks in surface soils following reversion from SRC willow that may persist for years after removal. Wachendroft et al. (2017) proposed that this increase in surface soil C stock may be due to the incorporation of large amounts (19.3–34.1 Mg C ha⁻¹) of coarse SRC residues into the soil, such as roots and stumps which are broken up but left in

field during reversion. The authors suggest that rather than being fully mineralised, a portion of these residues appear to become fragmented and subsequently occluded within soil aggregates (Wachendorf et al., 2017). These inputs are likely to be concentrated in the surface soil, even with tillage. Higher surface soil C stocks have also been reported in studies of SRC willow establishment on arable land (Rowe et al., 2016; Walter et al., 2015). This suggests that C stocks may also be elevated prior to reversion thus there is the potential for an additional legacy effect. Over a greater depth of 1 m, differences in soil C stock between the reverted SRC willow fields and their respective paired arable land uses were, however, reduced and not statistically significant, with the exception of the SRC -6 yrs. at the Doncaster site. The depth profiles suggest that the higher soil C stocks in the surface soil were balanced by reduction at greater depth. Again such changes in soil C distribution have also been reported in studies of SRC willow establishment. Studies have demonstrated soil C stocks in the surface soil to be higher under SRC willow (\leq 30 cm) in comparison to arable land uses, but, potentially due to an absence of tillage which is known to transfer surface C inputs to deeper layers, higher surface soil C stocks are balanced by lower C in the deep layers leading to limited overall change in C stocks (Rowe et al., 2016; Walter et al., 2015). These changes in the distribution highlight the need to consider the full soil profile when assessing impacts on soil C stocks of bioenergy crop reversion.

Post-reversion management

Changes in soil C distribution cannot explain the impacts on soil C stocks in the SRC -6 yrs. at the Doncaster site. Soil C stock in this field was significantly higher over the full 1 m sampling depth when compared to the arable land use (Table 3). Assuming no pre-existing differences between the reverted SRC willow and the arable, the higher soil C stock within the reverted SRC willow must be related to C sequestration occurring either during the lifetime of the SRC willow or after its removal. The higher soil C stock observed in the SRC -6 yrs. compared to the adjacent SRC -3 yrs. field suggests that whilst C inputs during crop removal or C sequestration during the lifetime of the crop may have occurred, factors post removal may have had a greater influence. Subsequent to reversion, the SRC willow fields have been managed using cob-only maize harvesting compared to more conventional arable cropping. This management has been associated with increased C sequestration of up to 23 % (Qin et al. 2016). However, this alone is unlikely to be sufficient to fully account for the ~70 Mg C ha⁻¹ difference in soil C stocks between the SRC willow and the arable, suggesting unknown factors such as additional inputs by the land owner.

This result serves to highlight the large potential impact of post removal land management on soil C stocks and the difficulty in generalising conclusions from individual sites.

Stability of crop residues -Miscanthus

Comparison to both the arable and Miscanthus land uses at the Nottingham sites showed lower soil C stocks in the reverted Miscanthus. Lower soil C stocks were also seen in the reverted Miscanthus at the Taunton site in comparison to the remaining Miscanthus crop. This contrasts with the results of Dufossé et al. (2014) who reported higher surface soil C stock one year after removal of a 20-year-old Miscanthus trial plots in northern France when compared to both remaining Miscanthus and arable land uses. Dufossé et al. (2014) proposed that whilst C losses in the reversion site may be ocurring, they may be obscured by the ~22 Mg C ha⁻¹ of Miscanthus residues (litter, stubble, roots and rhizomes) incorporated into the soil following reversion. Isotopic soil respiration (CO₂) measurements undertaken at the same study site did, however, show that whilst fluxes were increased, suggesting that losses of C from the soil were occurring, the source of this was only partly Miscanthus C, suggesting that losses from alternative soil C pools were being offset by the ~22 Mg C ha⁻¹ of Miscanthus residues (litter, stubble, roots and rhizomes) incorporated into the soil at the time of the reversions (Drewer, Dufossé, Skiba, & Gabrielle, 2016). The partial removal of rhizomes in the Nottingham site and the Taunton Miscanthus site -4yrs in this study would negatively impacted similar offsetting within this study although the level of this impact is unclear as data on the portion of rhizome removed is not available. Rhizomes were also not removed from the remaining two reverted Miscanthus fields in the Taunton site both of which also had significant lower surface soils C stocks that the Miscanthus control. Given the longer time frame between crop removal and sampling in this study, this raises the question on the potential longevity of the offsetting observed in Dufossé et al., (2014), as this will be dependent on the stability of newly incorporated Miscanthus residues over longer time scales.

Laboratory incubation studies suggest that Miscanthus rhizomes, which can make up 79 % of the below ground biomass of this crop (Dohleman, Heaton, Arundale, & Long, 2012), are readily mineralized (Amougou, Bertrand, Machet, & Recous, 2011; Beuch, Boelcke, & Belau, 2000). Incubation studies have reported mineralisation rates for rhizome C of 1.6 % to 2.9 % per week

with the authors noting the higher levels of neutral detergent-soluble (NDS) and lower lignin content in rhizomes compared to more recalcitrant root material (Amougou et al., 2011; Beuch et al., 2000). Whilst processes under field conditions may be more complex, the limited stability of Miscanthus residues, within soils may have also play a role in the differences observed between this study, where sampling was conducted 3–4 years after removal, and that of Dufossé et al., (2014) where sampling was conducted only one year after removal. This also highlights the potential contrasting fate of reversion residues between SRC willow and Miscanthus, with work by Wachendorf et al., (2017) suggesting that, in contrast to Miscanthus, SRC willow residues may become stabilised in soils. Willow roots and stumps also have a higher fraction of lignin compared to Miscanthus (Amougou et al., 2011; Berthod, Brereton, Pitre, & Labrecque, 2015). Lignin acts as a physical barrier for impeding microbial breakdown of plant material, therefore, even without stabilisation, willow residues may persist within the coarse and free particulate organic matter factions for a greater period of time (Austin & Ballaré, 2010). Further work is required but understanding the fate of SRC willow and Miscanthus reversion residues may be key in developing best practices for managing crop reversion.

Pre-reversion factors

The greater age of the Miscanthus prior to the bioenergy crop removal in Dufossé et al., (2014) (20 years versus 5–6 years in this study) may explain differences in the findings. As shown in the meta-analysis by Qin et al., (2016) reduced soil C stocks are not uncommon in young Miscanthus plantations on former arable land. Losses are generally associated with the establishment process and are replaced over time resulting in comparable or higher soil C stock under Miscanthus (Chimento, Almagro, & Amaducci, 2016; Qin et al., 2016). Time taken for soil C stock to recover can vary greatly between sites from only a few years to over 10 years (Qin et al. 2016). In the Nottingham sites it appears this recovery had not yet occurred and a proportion of the impacts seen during reversion can be attributed to unrecovered losses during the bioenergy crop establishment. Differences in soil types may also play a role in determining the response to reversion. Sandy soils, such as at the Nottingham site, are associated with weaker soil C stabilisation due to limited aggregation potential and low availability of mineral surfaces, resulting in soil C being more susceptible to disturbance during the reversion process.

Method of removal

Variation in the method used to remove the crops were present for both crops. For Miscanthus rhizome harvesting occurred in some but not all of the fields samples. Surface soil stock were 13.27–15.31 Mg C ha⁻¹ lower at the Taunton sites in the field where rhizome removal had occurred compared to the two other reverted field where rhizome were not harvested. These impacts were not significant, however, with soil C stock over the 0–1 m sampling depth being highest in the reverted Miscanthus field where removal of the rhizomes had occurred. It is also difficult to make direct comparison between the fields due to differences in removal and planting dates. In the cases of the SRC willow the tillage methods and the time between bioenergy crop removal and cultivation varied between the sites. The sites also varied in soil types, crop age and management post bioenergy crop removal making impacts of removal method impossible to assess. The existence of differences in removal methods do however highlight areas for future research

Conclusions

In this study SRC willow and Miscanthus reversions to arable cropping resulted in a range of impacts from a significantly lower soil C in a Miscanthus reversion (- 33.38 ± 5.34 Mg C ha⁻¹) to significantly higher soil C stocks under a SRC willow reversion (+ 70.15 ± 10.11 Mg C ha⁻¹). Comparison of these impacts to published LCA values show that changes in soil C stock were in some cases of a greater magnitude than that the C offset provided by the crops. Several key factors likely influence the outcome of bioenergy crop reversion impacts on soil C stocks including the stability of crop residue, post removal land management, longevity of the bioenergy crop prior to reversion and soil type. Future work is needed to understand the individual and interactive impacts of these factors, particularly on the processes involved in the stabilisation of Miscanthus and SRC willow crop residues following crop removal. This study also highlights the need to consider the impacts on soil C stocks of bioenergy crop removal in LCA and climate modelling.

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Data Sharing and Accessibility

The data that support the findings of this study are available from the corresponding author, [RR], upon reasonable request. The data on exact site locations will not be available as it contains information that could compromise the privacy of the land owners.

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Figure Legends

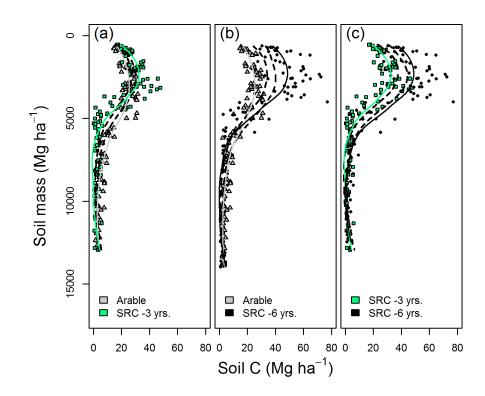
Figure 1: Bootstrapped Loess Regression (BLR) plots of cumulative soil mass versus soil C stock for the Doncaster site, with comparisons between land use pairs a) arable and SRC -3yrs.; b) arable and SRC -6yrs. and c) SRC -3yrs. and SRC -6yrs. For each paired comparison confidence intervals are represented by the area between the dotted lines; this is the null hypothesis of no difference between the land uses; solid lines represent Loess regression of each land use.

Figure 2: Bootstrapped Loess Regression plots of cumulative soil mass versus soil C stock for the Gainsborough site, with comparisons between land use pairs a) arable and SRC -5yrs.; b) arable and SRC -7yrs. and c) SRC -5yrs. and SRC -7yrs. For each paired comparison confidence intervals are represented by the area between the dotted lines; this is the null hypothesis of no difference between the land uses; solid lines represent Loess regression of each land use.

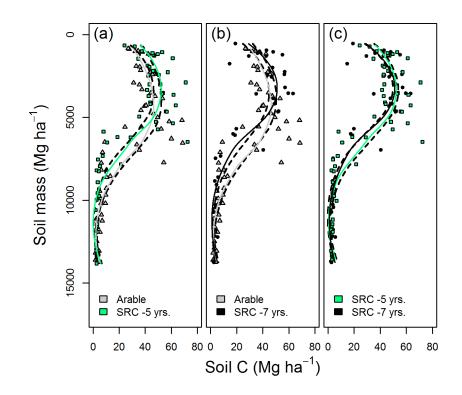
Figure 3 Bootstrapped Loess Regression plots of cumulative soil mass versus soil C stock for the Nottingham site, with comparisons are made between land use pairs a) Arable and Mis -3 yrs.; b) Mis and Mis -3 yrs. and c) Mis and Arable. For each paired comparison confidence intervals are represented by the area between the dotted lines; this is the null hypothesis of no difference between the land uses; solid lines represent Loess regression of each land use.

Figure 4: Bootstrapped Loess Regression plots of cumulative soil mass versus soil C stock for the Taunton A and B site, with comparisons made between land use pairs for Taunton A: a) Mis and Mis -3yrs., b) Mis and Mis -4yrs., and for Taunton B: c) Mis B and Mis -4yrs. B. For each paired comparison confidence intervals are represented by the area between the dotted lines; this is the null hypothesis of no difference between the land uses; solid lines represent Loess regression of each land use.

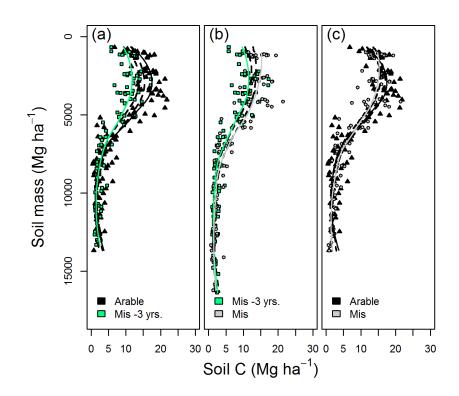
Figure 5: Comparison of potential C offset provided by SRC willow and Miscanthus based on LCA values with and without the inclusion of the soil C stock change 0–1 m (reverted bioenergy crop – arable) for the Doncaster, Nottingham and Gainsborough sites. LCA values are based on published studies, normalised life span of 15 years for Miscanthus and 20 years for SRC willow with yields of 11 Mg ha⁻¹ and 10 Mg ha⁻¹, respectively. Each dot representing the individual scenarios within the published LCAs (see Table S2 for more detail).



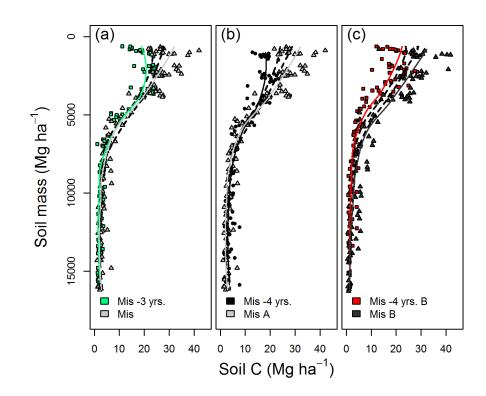
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