Physical Protection of Soil Carbon Stocks Under Regenerative Agriculture

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Keywords: Soil Carbon, Carbon Protection, Stabilised Carbon, Recalcitrant Carbon, Occluded Carbon, Regenerative Agriculture, Aggregate Stability.

Abstract

Regenerative agriculture is emerging as a strategy for carbon sequestration and climate change mitigation. However, for sequestration efforts to be successful, long-term stabilisation of Soil Organic Carbon (SOC) is needed. This can be achieved either through uplift in recalcitrant carbon stocks, and/or through physical protection and occlusion of carbon within stable soil aggregates. In this research soils from blackcurrant fields under regenerative management (0 to 7 years) were assessed. Soils from under the blackcurrant bush crop (*bush* (ca. 40% of the field area)), and the alleyways between the blackcurrant crop rows (*alley* (ca. 60% of the field area) were considered. Soil bulk density (SBD), soil aggregate fractions (proportions of water stable aggregates vs. non-water stable aggregates (WSA and NWSA~~,~~ ~~respectively~~)), soil carbon content, and carbon stability (thermally recalcitrant carbon vs. thermally labile carbon) were assessed. From this, long term carbon sequestration potential was calculated from both recalcitrant and occluded carbon stocks (both defined as stabilised carbon). Results indicated favourable shifts in the ~~proportion~~ percentage of NWSA : WSA with time, increasing from 27.6 % : 5.8 % (control arable field soil) to 12.6 % : 16.0 % (alley soils), and 16.1 % : 14.4 % (bush soils) after 7 years. While no significant ($p \geq 0.05$)) changes in whole field (area weighted average of alley and bush soils), recalcitrant carbon stocks were observed after 7 years, labile carbon stocks increased significantly ($p \leq 0.05$) from 10.44 t C ha^{-1} to 13.87 t C ha^{-1}. Furthermore, as a result of the occlusion of labile carbon within the WSA fraction, total stabilised carbon increased by 1.7 t C ha^{-1} over the 7 year period. This research provides valuable insights into the potential for carbon stabilisation and long-term stability prognoses in soils managed under regenerative agriculture practices, highlighting the important role ~~in~~ which soil aggregate stability plays in the physical protection of carbon, and potential therein to deliver long-term carbon sequestration.

1. Introduction

Land use change, conventional land management practice, and aggressive agricultural techniques remain key drivers of soil damage and degradation (Lal, 2001; Lambin et al., 2001; Foley et al., 2005; Pearson, 2007; Smith, 2008; Al-Kaisi and Lal, 2020). Without a shift to more sustainable approaches future agricultural productivity will be endangered, and with it the loss of food and economic security for many around the world (Zika and Erb, 2009; Tilman et al., 2011; Sundström et al., 2014).

The effects of soil degradation can greatly reduce environmental and ecosystem quality and function (Dominati et al., 2010; Power, 2010; Lal, 2015; Montanarella et al., 2016; Sanderman et al., 2017; Ipbes, 2018). ~~Soil erosion and loss of soil organic carbon (SOC), structural damage (destruction of soil aggregates and compaction), contamination, salinisation, and nutrient depletion all contribute to soil degradation (Lal, 2015; Montanarella et al., 2016; Sanderman et al., 2017); undermining the provision of key ecosystem services that underpin wider environmental health and function (Dominati et al., 2010; Power, 2010).~~

At landscape scales, soil degradation compounds and threatens desertification and biodiversity loss (Zika and Erb, 2009; Power, 2010; Orgiazzi and Panagos, 2018; Huang et al., 2020), while making significant contributions to greenhouse gas emissions and climate change (Lal, 2004; Smith et al., 2020). Globally, agriculture is associated with roughly a third of total land use and nearly a quarter of all global greenhouse gas emissions each year (Foley et al., 2011; Smith et al., 2014; Newton et al., 2020). To date it is estimated that more than 176 Gt of soil carbon has been lost to the atmosphere (Ipbes, 2018), with approximately 70-80% of this *(~130 - 140 Gt*) as a direct consequence of anthropogenic land management and soil cultivation (Sanderman et al., 2017; Lal et al., 2018; Smith et al., 2020). Meanwhile the area of land affected by desertification globally has been reported to exceed 25% and is expanding each year (Huang et al., 2020).

A key mechanistic step in the wider degradation of soil and soil carbon loss, is through the loss and destruction of stable soil aggregates and associated SOC~~,~~ mediated by conventional agricultural practice and soil disturbance (Smith, 2008; Baveye et al., 2020).

Soil aggregates that remain stable and resist disaggregation when exposed to water (*water stable aggregates*) are key determinants of soil structure and stability (Whalen et al., 2003), and act as an important indicator of overall soil quality due to their influence on wider soil properties (Lehmann et al., 2020; Rieke et al., 2022). Soil aggregate formation, as facilitated by SOC, assists the stabilisation and storage (through occlusion and physical protection) of carbon and imparts resilience to soils against erosion and climate change while providing hydrological benefits and enhancing soil fertility (Lal, 1997; Abiven et al., 2009; Kasper et al., 2009; Chaplot and Cooper, 2015; Veenstra et al., 2021; Rieke et al., 2022).

The formation and persistence of stable soil aggregates is instrumental in soil carbon sequestration (Lal, 1997; Six et al., 1998; Abiven et al., 2009), in particular~~.~~ ~~Particularly~~ due to the physical protection of labile carbon within soil aggregates which minimise biogenic and oxidative decay of SOC (Brodowski et al., 2006; Smith, 2008; Schmidt et al., 2011; Berhe and

Kleber, 2013). Soil aggregates can be classified by their formation conditions; *biogenic* (decomposition of organic matter and action of soil fauna), *physicogenic* (soil physical and chemical processes) and *intermediate* (a combination of both ~~biogenic and physicogenic~~ factors)(Ferreira et al., 2020). Additionally, land management practice can ~~further~~ influence these formation conditions and the stability or destruction of soil aggregates (Lal, 1997; Mikha et al., 2021).

It is important, when considering ~~viewed through the lens of~~ carbon sequestration that we acknowledge not all carbon is equal,~~. The potential for long-term carbon sequestration is governed by the resistance of the carbon to degradation. This~~ with long-term resistance to degradation being conferred through; i) inherent recalcitrance of the carbon, and ii) physical protection of the carbon ~~and~~ (occlusion within soil aggregates). Thus, when considering soil carbon sequestration ~~potentials~~ as a solution~~s~~ to climate change it is imperative that we differentiate between ~~soil~~ carbon which is transient and ~~soil~~ carbon which endures.

By adopting more sustainable management practices, agriculture can transition from a negative to a positive force for the environment; providing and enhancing a variety of key ecosystem services (*water regulation, soil propertiles~~y~~ regulation, carbon sequestration and biodiversity support*) (De Groot et al., 2002; Dominati et al., 2010; Power, 2010; Baveye et al., 2016; Keenor et al., 2021). ~~Th~~Herein, regenerative agriculture offers opportunities to produce food and other agricultural products with minimal negative, or even net positive outcomes for society and the environment; potentially~~-~~ improving farm profitability, increasing food security and resilience, and ~~helping to~~ mitigatinge~~e~~ climate change (Al-Kaisi and Lal, 2020; Newton et al., 2020).

~~Despite having no single definition or prescriptive set of criteria, r~~Regenerative agriculture practice may ~~is widely understood to~~ include the key concepts of: (i) reducing/limiting soil disturbance; (ii) ~~maintaining~~ continuous soil cover (~~as~~ perennial crops/vegetation, litter or mulch~~es~~), (iii) increased use of~~ing~~ ~~quantities of~~ organic matter inputs~~returned to the soil~~; (iv) maximised~~ing~~ crop nutrient and water-use efficiency ~~in crops~~; (v) integrating livestock; (vi) reducing or eliminating synthetic inputs (fertilisers and pesticides etc.); and (vii) increasing and broadening stakeholder engagement and employment (Newton et al., 2020; Paustian et al., 2020; Giller et al., 2021).

Adoption of regenerative practices such as no/minimum-till techniques increases the extent of soil aggregation and improves long-term carbon storage potential (Lal, 1997; Gál et al., 2007; Ogle et al., 2012; Lehmann et al., 2020). Furthermore, in addition to providing physical protection to more labile forms of soil carbon, the improved soil aggregation enhances resilience to the effects of drought and erosion, and provides better hydrological function and structure to the soil (Abiven et al., 2009; Bhogal et al., 2009; Baveye et al., 2020; Ferreira et al., 2020; Martin and Sprunger, 2022). No/minimum till techniques have been adopted worldwide and in a variety of agricultural contexts to help reduce soil erosion and SOC mineralisation, increase crop yields and minimise input costs all while building soil organic matter (Sisti et al., 2004; Pittelkow et al., 2015; Ferreira et al., 2020; Kan et al., 2021). Adoption of ~~minimum-till and~~ no/minimum ~~-~~till ~~methods~~, coupled with the incorporation of perennial crops ~~compared with conventional tillage~~ has been reported to significantly increase SOC content within the top 30cm of a soil when compared with conventional tillage and yearly biomass harvest and removal (Gál et al., 2007; Ogle et al., 2012; Ledo et al., 2020). ~~However, these potential SOC increases depend on agricultural context, climate and soil type (Lal, 2004).~~

Conversion of agricultural land from conventional to regenerative ~~approaches~~ management may increase macro-aggregation and aggregate stability (Lal, 1997), and by extension, provide the means to protect labile soil carbon; thus, enhancing long-term soil carbon sequestration efforts (Six et al., 1998; Brodowski et al., 2006; Smith, 2008; Schmidt et al., 2011; Berhe and Kleber, 2013). Furthermore, adoption of regenerative methods ~~such as no-till or reduced till~~ can also lessen machinery costs, working hours and direct carbon emission (Kasper et al., 2009). Indeed, resulting from the adoption of no-till methods, it is estimated that global emission reductions of approximately 241 Tg CO_2e have been achieved ~~globally~~ since the 1970s (Al-Kaisi and Lal, 2020).

To evaluate the influence of transitioning from conventional agricultural management to regenerative soft fruit production~~to soft fruit production under regenerative principles, from a regime of conventional cropping and tillage~~, a field experiment was undertaken on a commercial blackcurrant farm in Norfolk, UK. The experiment evaluated 5 blackcurrant fields managed regeneratively ~~under regenerative principles~~ for increasing lengths of time (0 – 7 years of establishment), and contrasted against a conventionally managed arable field, evaluated as a datum. The research assessed carbon stocks across the regimes and thereafter the proportion of carbon stocks associated with the~~ soil fractions: sand,~~ water stable aggregate~~s~~ (WSA) and non-water stable aggregate~~s~~ (NWSA) soil fractions, with respect to the soil under the blackcurrant bush crop (bush soil) and in between the rows of the blackcurrant crop (alley soils)~~ respectively~~, and at ~~the~~ field scale (~~both~~ alley and bush soils collectively). Thermogravimetric Analysis (TGA) was used to differentiate thermally labile and thermally recalcitrant carbon pools, and their association to the respective soil fractions, a proxy for wider carbon stability (Plante et al., 2005; Plante et al., 2011; Gregorich et al., 2015; Nie et al., 2018; Mao et al., 2022). The research sought to test the hypothesis that a switch from a high soil disturbance conventional arable ~~farming~~ system to a no soil disturbance regenerative ~~and~~ perennial ~~regenerative~~ soft fruit production system, would increase total soil carbon stock with time~~,~~ and that this carbon stock would become increasingly stabilised. With this increased carbon stored~~,~~ either as occluded carbon (held within WSA, conferring physical protection to these stocks), and/or with greater inherent resistance to degradation (i.e. thermally recalcitrant carbon). A glossary of terms defining different soil carbon pools and soil fractions considered in this research is provided in the supplementary information (**Table SI 1**).

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2. Methods

2.1 Field E~~e~~xperiment

This research was undertaken at Gorgate Farm, Norfolk, UK (52°41'58"N 0° 54'01"E). The farm is part of the wider Wendling Beck Nature Recovery ~~Environment~~ Project (WBNRP, 2024) a regenerative farming and environmental landscape management program set in approximately 750 ha. The field experiment comprised 5 blackcurrant fields established on sandy-loam soils in 2019, 2017, 2015, and 2013 (these representing 1, 3, 5, and 7 years since soil disturbance, respectively) and a conventionally managed arable field drilled with winter wheat as a datum (0 years since soil disturbance) ~~drilled with winter wheat~~. Soil samples were collected in late June 2021, immediately prior to the harvest of the blackcurrant crops and a month prior to harvest of the winter wheat crop.

Field cropping history in both the blackcurrant and the arable regimes (2014-2021) is shown in **Fig. 1**.

<table>
<tr><th></th><th>2014/15</th><th>2015/16</th><th>2016/17</th><th>2017/18</th><th>2018/19</th><th>2019/20</th><th>2020/21</th></tr>
<tr><td>Control Field</td><td>Wheat</td><td>Barley</td><td>Wheat</td><td>Barley</td><td>Wheat</td><td>Barley</td><td>Wheat</td></tr>
<tr><td>Year 1 Field</td><td>Wheat</td><td>Barley</td><td>Wheat</td><td>Barley</td><td>Wheat</td><td>Barley</td><td>Blackcurrant</td></tr>
<tr><td>Year 3 Field</td><td colspan="3">Blackcurrant</td><td>Wheat</td><td colspan="3">Blackcurrant</td></tr>
<tr><td>Year 5 Field</td><td colspan="2">Blackcurrant</td><td colspan="5">Blackcurrant</td></tr>
<tr><td>Year 7 Field</td><td colspan="7">Blackcurrant</td></tr>
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Figure 1: *Field cropping history for the arable control, and regenerative blackcurrant fields (2014-2021). Discrete Boxes represent one full cropping cycle and where applicable re-planting of new bushes.*

The regeneratively managed blackcurrant fields ~~under regenerative management~~ were planted using a conservation strip tillage approach;~~,~~ ~~with the blackcurrant~~ bushes are planted as field length strips, leaving alleyways approximately 2m wide. Blackcurrant bushes occupied approximately 40% of the field and the alleyways between the crops~~,~~ approximately 60%. Once planted, the blackcurrant crop required~~d~~ minimal intervention~~s~~ beyond the yearly harvest, pruning, fertilisation and sowing of cover crops in the alleyways ~~and fertilisation~~. Soil ~~Fields~~ remained covered year-round between the blackcurrant crop, with a diverse grazing cover crop through the autumn and winter months, and a summer fallow covering crop during the spring and summer months, both directly drilled (**Table SI 2**). Furthermore, the blackcurrant crop was treated with bi-yearly sprays of compost tea and urea fertiliser (78 kg ha^{-1} and 100 kg ha^{-1} pre and post flowering of the currants, respectively). Plant trimmings and residue were left on the soil surface in situ following yearly pruning and seasonal leaf drop. Comparatively the control comprised a conventionally managed arable field adjacent to the blackcurrant fields and occupying the same sandy-loam soil type. This field was cultivated yearly to 30cm depth and had been drilled with winter wheat in a wheat/barley rotation for the preceding 6 years. Furthermore, during cultivation the previous crop stubble was re-incorporated, and the crop was subsequently treated with a urea-based fertiliser at a rate of 100 kg ha^{-1} post emergence.

In the case of blackcurrants being replanted into previously used fields (e.g., the year 5 field (**Fig. 1**)), these soils were no longer classified as under continuous regenerative management. The removal of previously established bushes and the subsequent soil disturbance caused through cultivation, and replanting constituted a clear disruption to ongoing regenerative practice, and goal of no soil disturbance. As such, this site was more accurately characterised by the initiation of a new cycle of regenerative management, reflecting this transition and the accompanied soil disturbance, rather than as a continuation of the previous management.

2.2 Soil Sampling

Soil core samples (0 - 7.5cm; n = 5) were collected from beneath the blackcurrant bushes and at the centre of the alleyways of each blackcurrant field using a Dent soil corer. Whilst we acknowledge that collection and measurement of samples to a greater depth would be advantageous for determination of potential changes in soil composition of different strata, given the physiological differences between these different soil layers (Penman et al., 2003; Rovira et al., 2022), in practice the investigation was subject to limitations to our sampling protocol. As a result, the discussion of soil compositional characteristics and changes is kept relative to the topsoil (7.5cm soil depth). Further soil core samples (n = 5) were randomly collected from a conventionally managed arable field. Soil samples were sealed and retained in cold storage (≤ 4 °C) prior to laboratory analysis. Soil cores were subsequently oven dried (40 °C for 24 hrs) and soil bulk density calculated from the dry sieved (2 mm) bulk soil prior to soil fractionation (n = 5).

2.3 Soil Fractionation

Soil fractionations, namely, Water Stable Aggregates (WSA), Non-Water Stable Aggregates (NWSA) and sand (**Table SI 1**), were established using a capillary-wetting wet sieving method, adapted from Seybold and Herrick (2001). To generate these different soil fractions, the previously dried bulk density samples (n = 5) were dry sieved (2 mm) to remove all debris and material ≥2 mm, yielding the bulk soil fraction. Subsequently, this 2 mm sieved bulk soil (100 g) was placed on 63 µm sieves and slowly wetted with de-ionised water. Once damp, samples were submerged and oscillated under de-ionised water (manually agitated at 30 oscillations per minute in 1.5 cm of water for 5 minutes). Material that passed through the 63 µm sieve was collected and dried (40 °C for 24 h~~ou~~rs) and then weighed, yielding the~~-~~ NWSA fraction. The soil retained on the 63 µm sieve was further processed using ~~in~~ sodium hexametaphosphate (HMP) solution (0.02 M), to disaggregate any water stable aggregates from the remaining material and separate these from the sand and inorganic material present in the sample. The material remaining on the 63 µm sieve was then dried (40 °C for 24 h~~ou~~rs); and designated as the sand fraction (the overall change in sand fraction has been discounted to focus reporting on NWSA or WSA fractions). The WSA fraction (That which passed through the 63 µm sieve after receiving the HMP treatment) was subsequently established by calculation (**Eq. 1**):

Eq.1 $$\% \, \boldsymbol{WSA} = \left(\frac{\boldsymbol{Bulk\ Soil\ Mass_{dry}} - (\boldsymbol{Sand\ Mass_{dry}} + \boldsymbol{NWSA\ Mass_{dry}})}{\boldsymbol{Bulk\ Soil\ Mass_{dry}}}\right) \times \mathbf{100}$$

2.4 Total Carbon~~,~~ ~~and N~~ ~~c~~Content by ~~e~~Elemental ~~a~~Analysis

Dry bulk soil, and the separated soil fractions (sand ~~fraction~~ and NWSA~~ fraction~~), were milled to produce a fine powder, and subsequently samples (20 mg; n = 4) packed in 8 × 5 mm tin capsules. An elemental analyser (Exeter CHNS analyser (CE440)) was used to determine elemental abundance of C~~ and N~~. Instruments were pre-treated within conditioning samples (acetanilide 1900 µg), a blank sample (empty capsule) and an organic blank sample (benzoic acid 1700 µg) prior to ~~sample~~ analysis, and standard reference materials (acetanilide 1500 µg)

Formatted: Font: +Body (Calibri), Not Bold were run alongside samples (every 6th run) for QA/QC (a precision threshold of ± 1SD of the mean from the standard reference material) (Hemming, N.D.). Subsequently, WSA fraction carbon contents wasere subsequently established by calculation (variation of **Eq 1**).

2.5 Thermogravimetric Aassessment of SOC Sstability

Thermal stability of the SOC in the bulk soil, and the separated soil fractions (sand fraction and NWSA fraction) were assessed using a thermo-gravimetric analyser (Mettler Toledo TGA/DSC 1). Samples (n = 2) were contained in 70 µl platinum crucibles. Samples were heated, in an inert atmosphere, at a rate of 10 °C min^{-1} from 25 °C to 1000 °C. TGA data was subsequently used to ascribe the thermally labile and thermally recalcitrant carbon contents (hereafter referred to as just labile/recalcitrant, respectively) of the bulk soil and soil fractions, as well as any inorganic carbon within the samples. Data was split into 43 distinct phases by temperature range according to organic matter attrition windows as stated in Mao *et al.* (2022): i) 25 °C – 125 °C (moisture evaporation), ii) 125 °C – 375 °C (labile components) and, iii) 375 °C – 700 °C (recalcitrant components), iv) 700 °C – 1000°C (inorganic components). Subsequently, WSA fraction carbon stabilities were subsequently established by calculation (variation of **Eq 1**).

2.6 Carbon Assessment

Soil carbon was assessed as total SOC, soil fraction C (NWSA associated carbon, and WSA associated carbon respectively), total labile and recalcitrant C, occluded carbon (physically protected) and unstabilised C (**Table SI 1**). These fractions were not defined relative to particulate organic matter (POM) or mineral associated organic matter (MAOM), due to the method of sample preparation being unsuitable to accurately prescribe these fractions (Lavallee et al., 2020). In addition, C was further assessed on a total field carbon stock basis (in t C ha^{-1}). To calculate ~~the total field carbon stock in t ha^{-1} (for all carbon measures),~~ the C content of both the alley and bush soils (or the sum of their relative fractions) was multiplied by the relevant soil bulk density measure and the depth of sampling (ca. 7.5cm) and subsequently added together with acknowledgment of their proportion of the field (60% alley and 40% bush, respectively), as set out in (**Eq. 2**):

Eq.2 $$C\ tha^{-1} = \left(0.6\left(C_{Alley} \times SBD_{Alley} \times Depth\right)\right) + \left(0.4\left(C_{Bush} \times SBD_{Bush} \times Depth\right)\right)$$

2.7 Statistical A~~a~~nalysis

Significant differences between the field sites were determined using *post hoc* tests on one-way ANOVA with Tukey's HSD, and data significance set to 95 % ($p \leq 0.05$) (ANOVA; IBM SPSS 28). Significant differences between the individual regimes within field sites (alley soil vs. bush soil) were determined using two tailed T-tests, with data significance set at two levels of confidence; 95 % ($p \leq 0.05$), and 99 % ($p \leq 0.01$) (independent samples T-test; IBM SPSS 28).

3. Results and Discussion

3.1 Bulk Density

When considering soil stability, soil bulk density (SBD) provides significant insights into the arrangement and structure of soil particles, and the extent of soil aggregation (Al-Shammary et al., 2018). As SBD accounts for the total volume that soils occupy (including the mineral, organic and pore space components), it is a key indicator of soil condition (Chaudhari et al., 2013; Allen et al., 2011). Furthermore, SBD maintains a close correlation to concentrations of organic matter and carbon within the soil, where soils become depleted in carbon, SBD tends to increase, potentially leading to compaction of soil structures (Allen et al., 2011).

SBD was observed to decrease significantly ($p \leq 0.05$) in both the alley ~~soils~~ and bush soils of~~in~~ all regeneratively managed fields relative to the conventional control (1.75 g cm^{-3}) (**Fig.**

2). ~~The highest overall SBD was measured in the control soil (1.75 g cm^{-3}) and the lowest SBD in the year 3 bush soil (1.07 g cm^{-3}) (**Fig. 2**).~~

In the alley soils, SBD was observed to decrease successively with each additional year under regenerative management; from 1.35 g cm^{-3} in the year 1 soil, to 1.15 g cm^{-3} in the year 7 soil, with no significant differences observed between the regeneratively managed fields ($p \geq 0.05$). ~~significantly ($p \leq 0.05$) in all regeneratively managed soils compared to the conventional control (**Fig. 2**).~~ While ~~Between the regeneratively managed soils SBD was observed to decrease (not significantly ($p \geq 0.05$)) successively with each additional year under regenerative management; from 1.35 g cm^{-3} in the year 1 alley soil, to 1.15 g cm^{-3} in the year 7 alley soil (relative to 1.75 g cm^{3} in the conventional control soil) (**Fig. 2**). I~~in the bush soils, SBD ~~was also observed to decrease significantly ($p \leq 0.05$) in all regeneratively managed soils relative to the conventional control~~decreases were not successive between the regeneratively managed fields (**Fig. 2**). ~~Between the regeneratively managed soils SBD was observed to generally decrease with time, however this was not successive;~~ Intra-regenerative field comparison showed the greatest decrease in bush soil SBD (significant ($p \leq 0.05$)) was observed between the year 1 and year 3 ~~soils~~fields, reducing from 1.32 g cm^{-3} in to 1.07 g cm^{-3} , before increasing (not significantly ($p \geq 0.05$)) in the year~~s~~ 5 and 7 fields (to 1.18 g cm^{3} and 1.16 g cm^{3} respectively)(**Fig. 2**), likely a consequence of increased stoniness (thus reducing core mass and volume) in the year 3 samples (**Table SI 3**). When compared pairwise, SBD in the alley ~~soils~~ and the bush~~es~~ soils of the regeneratively managed fields were observed to be broadly similar, with only one pair (*year 3*) ~~showing~~ measuring a significant difference (p ~~<~~≤ 0.05) of ~~between the alley and bush soils, measuring~~ 1.27 g cm^{-3} and 1.07 g cm^{-3} respectively (**Fig. 2**). However, this difference likely related more to the underlying soil

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physiology and stoniness of the samples collected than to differences in management practice (Fig. SI 3)~~.~~

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None of the soils measured in this investigation were observed to exceed the root limiting soil density factor of 1.8 g cm^{-3} in sandy soil types, suggesting no significant detriment to ~~the growth of~~ plants growth from soil compaction (Kaufmann et al., 2010; Shaheb et al., 2021). Furthermore, the overall trend of soil bulk density reduction seen over the course of the 7-year period (**Fig. 2**) is likely a consequence of both increased aggregate stability and quantity of stable aggregates (**Sect. 3.2**) alongside increases in soil carbon stocks (**Sect. 3.3**) (Topa et al., 2021; Rieke et al., 2022; Kasper et al., 2009).

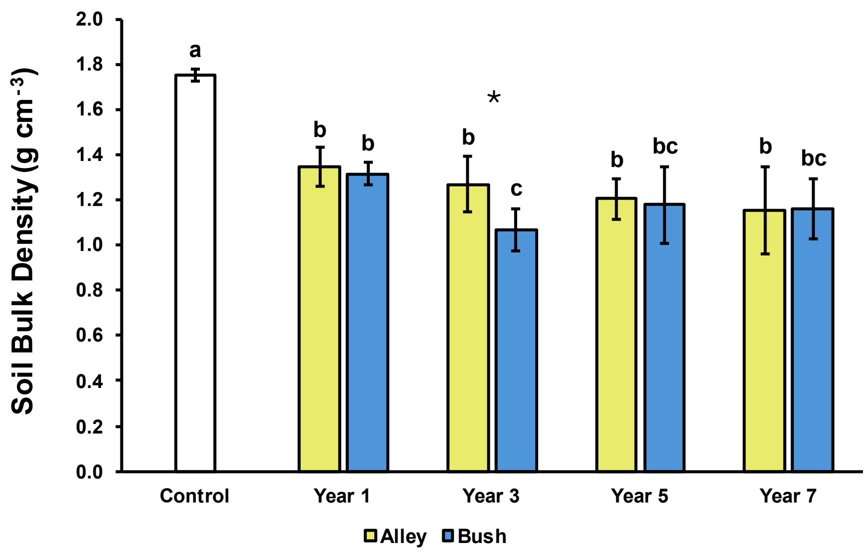

Figure 2: Soil bulk density (n=5) of alley (yellow) and bush (blue) regimes with increasing years of establishment. Error bars represent ± 1SD. For a given regime (alley or bush) dissimilar lower-case letters indicate significant ($p \leq 0.05$) differences across the timeseries. At a given timepoint, * indicates a significant difference ($p < 0.05$) between the alley and bush regimes.

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3.2 Soil Fractionation

Proportions of WSA and NWSA in both the alley and bush soils were seen to change significantly (p ≤ 0.05) in ~~both the alley and bush soils~~ with increased time under regenerative management (**Fig. 3**).~~. While the sand fraction also observed significant changes (p ≤ 0.05) between some of the alley and bush soils (**Fig. 3**), the overall change in sand fraction has been discounted to focus reporting on NWSA or WSA fractions.~~

~~Soil WSA and NWSA fractions in both the alley soils and bush soils observed opposing trends with age of establishment. With~~ NWSA in both the regimes reduced~~ing~~ in fractional share significantly (p ≤ 0.05) over the 7 years of establishment, while the WSA fractional share ~~proportion~~ increased significantly over time (p ≤ 0.05) (**Fig. 3; Table SI 3**). ~~.~~Such changes were likely due to halting of soil tillage (*with a decrease in NWSA, and commensurate increase in WSA evident in the first year of no-till adoption*) and further enrichment with increasing time since soil disturbance. Furthermore, these shifts in NWSA vs WSA proportions were noted to be ~~proportionate~~ commensurate with soil carbon increases (**Sect. 3.3**) and SBD decreases (**Sect. 3.1**), Collectively these changes may suggest enhanced soil aggregate stability and cohesion (Abiven et al., 2009; Six et al., 2004; Kasper et al., 2009).

NWSA fractions in the alley soils decreased successively with time, from a total of 27.6% in the control soil to 12.6% in the year 7 soil, with significant reductions (p ≤ 0.05) measured between the control soil and all regeneratively managed soils (**Fig. 3; Table SI 3**). Additionally, NWSA in the year 7 soil was measured to be significantly lower (p ≤ 0.05) than all other regeneratively managed soils (**Fig. 3; Table SI 3**). In the bush soil, NWSA fractions were also observed to decrease significantly (p ≤ 0.05) in all regeneratively managed soils relative to the control, ~~ranging~~ reducing from~~between~~ 27.6% in the control to 16.1% in the year 7 soil ~~15.2% in the year 1 soil~~ (**Fig. 3; Table SI 3**). However, this decrease was not successive, with ~~as~~ the

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greatest reduction ~~was~~ measured in the year 1 soil (15.2%), however, ~~soil and increased (not significantly ($p \geq 0.05$)) to then broadly plateau in subsequent years (**Fig. 3; Table SI 3**). Furthermore,~~ no significant differences ($p \geq 0.05$) were observed between any of the regeneratively managed soils. When compared pairwise significant differences ($p \leq 0.01$) between the alley and bush soil~~s~~ NWSA were observed in the year 5 and year 7 soils, ~~(**Fig. 3; Table SI 3**). NWSA content of the alley soils was measured to be~~ significantly ($P \leq 0.01$) lower in the alley soils than ~~that of~~ the bushes (15.9% vs. 18.8% in year 5; 12.6% vs. 16.1% in year 7, ~~in the alley and bush soils~~ respectively) (**Fig. 3; Table SI 3**).

Conversely WSA fractions in the alley soils increased broadly with age of establishment, from 5.8% in the control soil to 16.0% in the year 7 soil, with significant increases ($p \leq 0.05$) measured between the control soil (5.8%) and both the year 5 and year 7 soils (10.3% and 16.0% respectively), (**Fig. 3; Table SI 3**). Additionally, the WSA fraction in year 7 was observed to be significantly greater (p ~~<~~≤ 0.05) than in all other regeneratively managed soils (**Fig. 3; Table SI 3**). In the bush soils, the WSA fraction was also observed to generally increase with time, from 5.8% in the control soil to 14.4% in the year 7 soil; with significant increases ($p \leq 0.05$) measured in the year 5 and year 7 soils (11.0% and 14.4% respectively) (**Fig. 3; Table SI 3**). Within the regeneratively managed soils, significant differences ($p \leq 0.05$) were also observed between the year 5 soil and the year 3 soil, and between the year 7 soil and years 1 and 3 soils (**Fig.3; Table SI 3**). When compared pairwise no significant differences ($p \geq 0.05$) were observed for the WSA content of the alley and bush soils in each year of regenerative management (**Fig. 3; Table SI 3**).

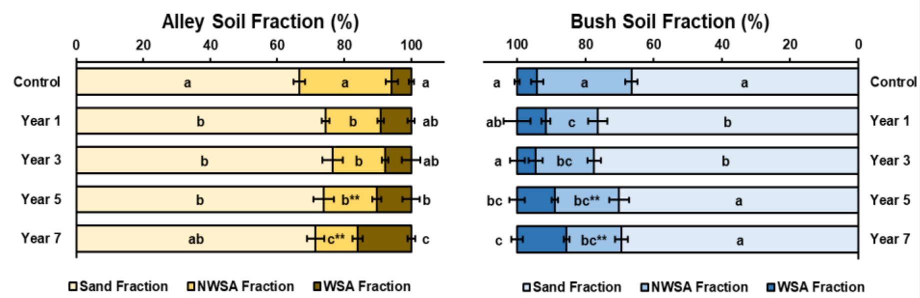

Figure 3: *Sand, Non-Water Stable Aggregates (NWSA), and Water-Stable Aggregates (WSA) fractions (% total mass)) (n=5) of alley (left) and bush (right) regimes with increasing years of establishment. Error bars represent ± 1SD. For a given regime (alley or bush) dissimilar lower-case letters indicate significant ($p \leq 0.05$) differences across the timeseries. At a given timepoint, the * indicates a significant difference ($p \leq 0.05$) between the alley and bush regimes. ** indicates a significant difference ($p \leq 0.01$), between the alley and bush regimes.*

3.3 Soil Carbon and Thermal Stability

SOC was observed to increase in both the alley and bush soils over time (**Fig. SI 1**), with significant increases ($p \leq 0.05$) in the year 5 bush soil (22.3 g kg^{-1} C) and both the alley and bush soils of year 7 (29.9 g kg^{-1} C and 23.8 g kg^{-1} C respectively) relative to the control soil (16.6 g kg^{-1} C) (**Fig. SI 1**). Such increases in SOC likely pertaining to lower carbon turnover from reduced soil disturbance, and increased carbon input; from the perennial root systems of the blackcurrant bushes and alley conservation strip, and litter/residue derived from crop pruning and seasonal leaf fall, relative to the yearly removal of biomass in the conventionally managed field (Ledo et al., 2020; Kan et al., 2021). While increases in SOC were more pronounced in the alley soils than in the bush soils, no significant ($p \geq 0.05$) differences were observed when compared pairwise (**Fig. SI 1**).

The relative stability of soil carbon is an underlying feature of its inherent environmental value: biological function and soil biodiversity rely heavily upon easily degradable carbon pools with short residence times, while services such as carbon sequestration and long term storage rely upon the more stable recalcitrant carbon pools that can resist degradation

(Dell'abate et al., 2003; De Graaff et al., 2010; Kleber, 2010; Keenor et al., 2021; Martin and Sprunger, 2022). Thermal techniques such as thermogravimetric analysis can provide effective means of characterising these organic matter pools in the soil, defining the profile of SOC stability (Dell'abate et al., 2000; Dell'abate et al., 2003; Plante et al., 2005; Plante et al., 2011; Mao et al., 2022). Furthermore, this thermal stability can provide a proxy for biogenic decay and degradation of soil organic matter and carbon stocks (Plante et al., 2005; Plante et al., 2011; Gregorich et al., 2015; Nie et al., 2018; Mao et al., 2022).

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Total labile and recalcitrant carbon pools were observed to increase in a broadly stepwise manner over the 7 year period relative to the control soil, with marginally more labile carbon than recalcitrant carbon measured measured in both alley soils and bush soils and in each field across all years (**Fig. 4**). However, Additionally, while increases in labile carbon the content of labile carbon were increased significantly (p ≤ 0.05) in both the alley and bush soils with time (years 5 and 7), while no significant differences (p ≥ 0.05) were observed in betweenthe recalcitrant carbon pools of either the alley or bush soils were observed (**Fig. 4**).

Labile soil carbon measured in the alley soils increased in all regeneratively managed soils relative to the control soil. broadly stepwise with increasing period of establishment, with labile carbon increasing in all regenerative managed soils relative to the control soil (**Fig. 4**). These increases were sSignificant increases (p ≤ 0.05) were measured in both the year 5 and year 7 soils relative to the control relative to the control (increasing from 7.9 g kg^{-1} C $_{labile}$ (control) to 13.6 g kg^{-1} C $_{labile}$, and 17.6 g kg $^{-1}$ C $_{labile}$, years 5 and 7 respectively)(**Fig. 4**). , i.e., an increase of 9.7 g kg^{-1} C $_{labile}$ (**Fig. 4**). Additionally, significant differences (p ≤ 0.05) in the labile carbon pool measured in the year 7 soil wereas observed between the year 7 and to be significantly greater (p ≤ 0.05) than that of the years 1 and 3 alley soils (**Fig. 4**).

-In the bush soils, ~~the labile soil carbon pool followed the same trend of broadly stepwise increase in all regeneratively managed soils relative to the control. S~~significantly greater ($p \leq 0.05$) labile carbon stocks were also measured in the year 5 and year 7 soils relative to the control (increasing from 7.9 g $kg^{-1}C_{labile}$ to 12.4 g $kg^{-1}C_{labile}$ and 13.9 g kg^{-1} C_{labile}, respectively) ~~i.e., an increase of 4.0 g kg^{-1} C_{labile}~~ (**Fig. 4**). Furthermore, significant differences ($p \leq 0.05$) were measured between regeneratively managed soils (year 5 and 7 vs. year 3; and year 7 vs. year 1) (**Fig. 4**). When compared pairwise, labile carbon in the alley soil increased by a total of 9.7 g kg^{-1} C_{labile}, vs. Increase of 4.0 g kg^{-1} C_{labile} in the bush soil after 7 years of regenerative management, suggesting enhanced labile carbon stock growth in alley soils relative to bush soils (**Fig. 4**). However, no significant differences ($p > 0.05$) were observed between alley and bush soils of the same field ~~years~~ (**Fig. 4**).

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Recalcitrant carbon measured in the alley soils increased broadly stepwise ~~with increasing~~ relative to the control with time, from 8.7g kg^{-1} $C_{recalcitrant}$ (control) to 12.3 g kg^{-1} $C_{recalcitrant}$ (year 7 soils), however none of these increases were significant ($p \geq 0.05$)(**Fig. 4**). In the bush soils, recalcitrant carbon was also observed to generally increase with time (not significantly ($p \geq 0.05$)). These increases were smaller than those observed within the alley soils, increasing (not significantly ($p \geq 0.05$)) from 8.7 g kg^{-1} $C_{recalcitrant}$ (control) to 9.9 g kg^{-1} $C_{recalcitrant}$ in the year 7 soil (**Fig. 4**). When compared pairwise, recalcitrant carbon stocks in the alley soil increased by a total of 3.6 g kg^{-1} $C_{recalcitrant}$, compared with -1.2 g kg^{-1} $C_{recalcitrant}$ in the bush soil after 7 years of regenerative management, again suggesting increased carbon stock growth in the alley soils relative to the bush soils (**Fig. 4**). However, no significant differences ($p > 0.05$) were observed between alley and bush soils of the same field (**Fig. 4**).

Considering both labile and recalcitrant carbon collectively, for labile and recalcitrant carbon stocks in the alley soils and bush soils, no significant differences ($p \geq 0.05$) were observed between any of the given years.

period of establishment, with all regeneratively managed soils increasing relative to the conventional control, however none of these increases were significant ($p \geq 0.05$) (**Fig. 4**).

Over the 7 year period recalcitrant carbon in the alley soils increased (not significantly ($p \geq 0.05$)) by 3.6 g kg^{-1} C$_{recalcitrant}$ (from 8.7g kg^{-1} C$_{recalcitrant}$ (control) to 12.3 g kg^{-1} C$_{recalcitrant}$ (year 7 soils) (**Fig. 4**).In the bush soils, recalcitrant carbon was also observed to generally increase with time (not significantly ($p \geq 0.05$)). However, these increases were smaller than those observed within the alley soils (**Fig. 4**). Recalcitrant carbon in the bush soil increased (not significantly ($p \geq 0.05$) from 8.7 g kg^{-1} C$_{recalcitrant}$ (control) to 9.9 g kg^{-1} C$_{recalcitrant}$ (year 7(**Fig. 4**).) i.e., a difference of 1.2 g kg^{-1} C$_{recalcitrant}$ (**Fig. 4**). When compared pairwise for labile and recalcitrant carbon stocks in the alley soils and bush soils, no significant differences ($p \geq 0.05$) were observed between any of the given years. However, it was observed that both alley and bush soils followed the same trend, with a greater proportion of both labile and recalcitrant carbon stored within the alley soils (**Fig. 4**). Bby year 7, the alley soil was observed to contain a total carbon content of 29.9 g kg^{-1} C (split as 17.6 g kg^{-1} C$_{labile}$ and 12.3 g kg^{-1} C$_{recalcitrant}$), while the bush soil contained a total carbon content of 23.8 g kg^{-1} C (split as 13.9 g kg^{-1} C$_{labile}$ and 9.9 g kg^{-1} C$_{recalcitrant}$). In contrast, total carbon content in the control soil was 16.6 g kg^{-1} C (split as 7.9 g kg^{-1} C$_{labile}$ and 8.7 g kg^{-1} C$_{recalcitrant}$) (**Fig. 4**).

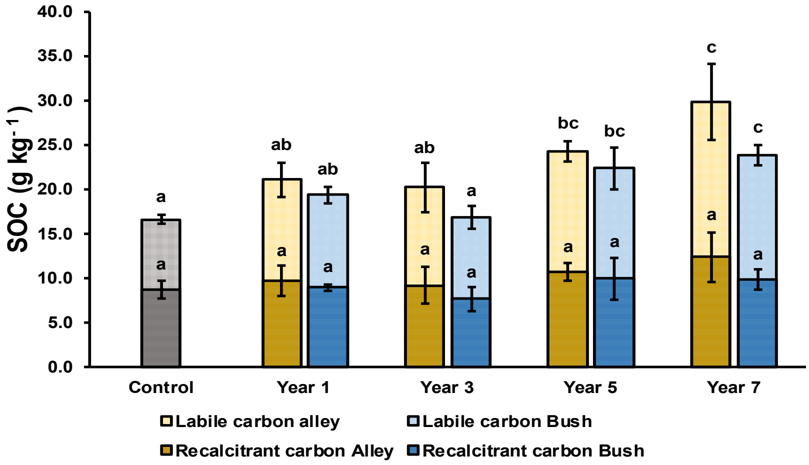

Figure 4: Soil Organic Carbon (SOC) split by thermally recalcitrant (hashed) and thermally labile (plain) carbon pools (n=5) in the alleyway (yellow) and bush (blue) regimes. Error bars represent ± 1SD. For a given regime (alley or bush) dissimilar lower-case letters indicate significant ($p \leq 0.05$) differences across the timeseries. At a given timepoint, * indicates a significant difference ($p < 0.05$) between the

3.4 Carbon Thermal Stability in Aggregate Fractions

Total labile and recalcitrant carbon pools, when split by soil fraction, were found to diverge over the 7 year period, with greater proportions of carbon (*both labile and recalcitrant*) observed in the WSA fraction while diminishing in the NWSA fraction with time. It is highlighted that despite their smaller fractional share (**Sect. 3.2**), WSA were substantially enriched in carbon relative to the NWSA fraction.

Labile carbon in the alley soils was observed to shift ~~between dominance in the NWSA fraction~~ toward dominance of the WSA fraction with time, with significant decrease ($p \leq 0.05$) in the NWSA fraction and a non-significant increase ($p \geq 0.05$) in the WSA fraction over the 7 year period (**Fig. 5A**). Alley ~~When analysed by aggregate fraction, the labile carbon pool in the~~ NWSA fraction labile carbon ~~was observed to~~ significantly decreased ($p \leq 0.05$) ~~with increased time under regenerative management,~~ from 33.7% ~~(~~in the control~~)~~ to 17.5% in the year 7 soil~~(year. 7).~~ However, no significant differences ($p \geq 0.05$) were measured between the

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Formatted: Normal, Indent: First line: 0 cm, Don't add space between paragraphs of the same style control and the other regeneratively managed soils (**Fig. 5A**). While alley ~~Within the~~ WSA fraction ~~the~~ labile carbon ~~pool was observed to~~ increased (not significantly ($p \geq 0.05$)) from 45.5% in the ~~conventional~~ control to 61.3% in the year 7 soil (**Fig. 5A**). additionally, ~~I~~initial reductions in ~~the~~ labile carbon ~~pool~~ were observed in year 1 and year 3 relative to the control (reducing to a low of 38.1% in the year 3 soil), before rebounding in years 5 and 7. However no significant differences ($p \geq 0.05$) were observed between any of the regeneratively managed alley soils (**Fig. 5A**).

Labile carbon in the bush soils was similarly observed to shift ~~from~~ toward ~~dominance in the NWSA fraction to~~ dominance of ~~in~~ the WSA fraction with time under regenerative management, culminating in reduced NWSA and increased WSA ~~fraction~~ associated labile carbon by year 7. However, this trend was less pronounced than within the alley soil, with a ~~and no~~ significant difference~~s~~ ($p \geq 0.05$) only observed ~~were observed~~in the year 7 WSA fraction ~~overall~~ (**Fig. 5B**). Bush NWSA fraction labile carbon was observed to decrease (not significantly ($p \geq 0.05$)) between the control and year 7 soil, reducing from 33.7% to 23.7% respectively, additionally, no significant differences $p \geq 0.05$) were measured between the other regeneratively managed soils (**Fig. 5B**). ~~Within the NWSA fraction no significant differences ($p \geq 0.05$) were observed between the control and any regeneratively managed soil (**Fig. 5B**). Labile carbon initially decreased in year 1 relative to the control (from 33.7% to 24.8%) before converging with the control in years 3 and 5 (33.6% and 33.8% respectively) and subsequently reducing again in year 7 (23.7%) (**Fig. 5B**).~~While bush ~~In the~~ WSA fraction ~~the~~ labile carbon ~~pool~~ increased (not significantly ($p \geq 0.05$)) between the control and year 7 soil (45.5% to 54.8%), ~~. H~~however~~,~~ these changes were not as substantial as those observed in the alley soils (**Fig. 5B**). Additionally, while WSA associated labile carbon decreased in the year 3 soil to 28.2%, (~~while this decrease was~~ not significantly ($p < 0.05$)) ~~relative to the control~~, this ~~labile carbon content~~ was observed to rebound significantly ($p \leq 0.05$) from year 3 to year 7, showing overall increase in WSA associated labile carbon (**Fig. 5B**).

When compared pairwise, a significant difference ($p \leq 0.05$) was observed between the NWSA fraction of year 5 alley and bush soils, with 23.7 % ~~of the~~ labile carbon ~~pool contained with~~in the ~~NWSA fraction of the~~ alley soil relative to 33.8 % in the bush soil; no further significant differences ($p \geq 0.05$) were observed (**Fig. 5 A/B**).

Recalcitrant carbon in the alley soils was also observed to enrich in WSA relative to the NWSA fractions over time, with the decrease in NWSA being significant ($p \leq 0.05$), while the increase in WSA was not significant ($p \geq 0.05$) over the 7 year period (**Fig. 5C**).

~~When analysed by fraction, the recalcitrant carbon pool in the~~Alley NWSA fraction recalcitrant carbon ~~was observed to~~ decreased broadly stepwise, with a significant decrease ($p \leq 0.05$) measured between the 7-year and control soil~~s~~ (from 33.2% to 18.9%) (**Fig. 5C**). Significant differences ($p \leq 0.05$) were also observed between the year 3 and year 7 soils, where NWSA fraction proportion increased to converge with the control in the year 3 soil (32.2 %), and thereafter decreasing in year 5 and year 7 (**Fig. 5C**).

~~In the~~Alley WSA fraction ~~the~~ recalcitrant carbon ~~pool~~ was observed to increase (not significantly ($p \geq 0.05$)) with time, ~~increasing~~ from 50.1% in the control to 64.5% in the year 7 soil (**Fig. 5C**). Initial decreases in recalcitrant carbon were measured ~~observed~~ in the year 1 soil ~~relative to the control (decreasing~~ (not significantly ($p \geq 0.05$)) to 41.0 %), following ~~.~~ ~~T~~thereafter subsequent stepwise increases in all other regeneratively managed soils ~~were observed~~ (**Fig. 5C**).

Recalcitrant carbon in the bush soils was also observed to increase ~~in~~ the WSA fraction (not significantly ($p \geq 0.05$)) and decrease (not significantly ($p \geq 0.05$)) within the NWSA fraction with time, from the control soil to the year 7 soil (**Fig. 5D**).

~~When analysed by fraction, the recalcitrant carbon pool in the~~ Bush NWSA fraction recalcitrant carbon was observed to decrease overall by year 7 (from 33.2% in the control to 26.2%),~~.~~ ~~H~~however, no significant differences ($p \geq 0.05$) were measured between any of the regeneratively managed soils and the control (**Fig. 5D**).

Conversely, bush ~~Within the~~ WSA fraction~~,~~ recalcitrant carbon was observed to increase overall from the control to year 7~~,~~ (**Fig. 5D**). ~~with i~~Initially ~~reductions~~ (not significant ($p \geq 0.05$)) reductions were measured in year 1 and 3 relative to the control soil~~,~~ (decreasing from 50.1% in the control to 36.4% in the year 3 soil), ~~(**Fig. 5D**). WSA was~~before subsequently ~~observed to~~ increasing~~e~~ stepwise to a total of 56.4% in year 7 (however, this was not significantly different ($p \geq 0.05$) to the control) (**Fig. 5D**).

When compared pairwise significant differences ($p \leq 0.05$) were observed between the ~~in the recalcitrant carbon pools of the~~ NWSA fraction recalcitrant carbon stocks of ~~in~~ both year 5 and year 7 soils, with 23.9% and 18.9% stored in the alley soils, vs. 34.1% and 26.2% stored in the bush soils respectively (**Fig. 5 C/D**).

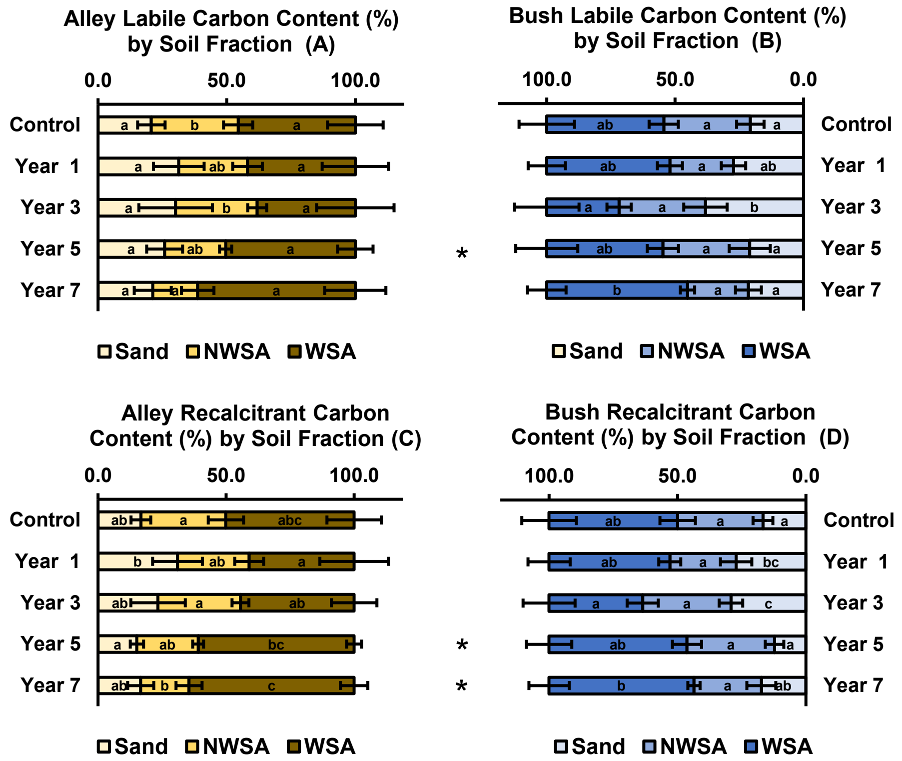

Figure 5: Thermally, ~~L~~labile (top) and thermally recalcitrant (bottom) SOC split by soil aggregate fraction (Sand, Non-Water Stable Aggregates (NWSA) and Water-Stable Aggregates (WSA)) as a total % of soil mass (n=5), of alley (left) and bush (right) soils with increasing years of establishment. Error bars represent ± 1SD. For a given soil fraction (sand, NWSA, WSA) dissimilar lower-case letters indicate significant ($p \leq 0.05$) differences across the timeseries. At a given timepoint, the * indicates a significant difference ($p \leq 0.05$) between the alley and bush regimes. ** indicates a significant difference ($p \leq 0.01$), between the alley and bush regimes.

3.5 Aggregate Occlusion of Carbon

Despite the inherent degradability of the labile carbon stocks of in both NWSA and WSA aggregate structures, these can be considered as distinct carbon pools for the purpose of long-term carbon storage and stability (Six et al., 1998; Mclauchlan and Hobbie, 2004). The ascribed ~~O~~occluded carbon pool considered the stabilised labile carbon stocks held within the WSA fraction (**Sect. 3.4**).~~,~~ Considered as such due to the long-term storage potential conferred by physical protection within the aggregate structures, and the physical~~ly~~ separation~~g~~ of the carbon from its potential vectors of degradation - ~~and~~ inhibiting the breakdown and decomposition of the carbon stored within (Schrumpf et al., 2013; Gärdenäs et al., 2011; Six and Jastrow, 2002; Dungait et al., 2012; Plante et al., 2011; Mclauchlan and Hobbie, 2004; Smith, 2008). Conversely unstabilised carbon considered the labile carbon that contained within the NWSA fraction (**Sect. 3.4**), and thus with greater potential for degradation, due to the enhanced potential for carbon oxidation and decomposition by soil biota (Smith, 2008; Berhe and Kleber, 2013; De Gryze et al., 2006; Six et al., 1998; Dungait et al., 2012). Additionally, recalcitrant carbon (**Sect. 3.3**), was considered stabilised regardless of the soil aggregate pool in which it was contained (both WSA and NWSA) due to the relative stability of this carbon fraction.

Occluded carbon in the alley soils ~~was observed to~~ increased broadly stepwise with time, with ~~measuring~~ increased occluded carbon content in all regeneratively managed soils relative to the ~~conventional~~ control. However, this increase was only significant ($p \leq 0.05$) in the year 7 soil, ~~(~~increasing from 3.64 g kg^{-1} C in the control to 10.99 g kg^{-1} C ~~in the control and year 7 soil~~over the 7 year period ~~)~~ (**Fig. 6**). In the bush soil, occluded carbon was observed to follow a similar trend to that in the alley, increasing significantly ($p \leq 0.05$) from 3.64 g kg^{-1} C in the control to 7.66 g kg^{-1} in the year 7 soil (**Fig. 6**). However, a decrease (not significant ($p \geq 0.05$)) in the occluded carbon content of the year 3 bush soil was measured relative to the control soil, reducing to 2.64 g kg^{-1} C, before rebounding in years 5 and 7 (**Fig. 6**). When compared pairwise, no significant differences ($p \geq 0.05$) were observed between the occluded carbon contents of either the alley soils or bush soils, however ~~with~~ a greater quantity of occluded carbon was measured ~~stored~~ within the alley soils relative ~~than~~to the bush soils in all but year 1 (**Fig. 6**).

Unprotected carbon in the alley soils was observed to increase (not significantly ($p \geq 0.05$)) in all ~~the~~ regeneratively managed soils relative to the control soil, ~~. However, this increase remained broadly similar across all regeneratively managed soils,~~ ranging between 6.4 g kg^{-1} C and 6.7 g kg^{-1} C, compared with 4.2 g kg^{-1} C in the control soil (**Fig. 6**). In the bush soil, unprotected carbon was observed to increase broadly stepwise, with significant increases ($p \leq 0.05$) in the years 3, 5 and 7 soils relative to the control, and increasing to a maximum of 6.6 g kg^{-1} (in the year 5 soil) relative to 4.2 g kg^{-1} C in the control soil (**Fig. 6**). When compared pairwise no significant differences ($p \geq 0.05$) were observed between the regeneratively managed soils, with unprotected carbon measuring similarly in both the alley soils and bush soils (**Fig. 6**).

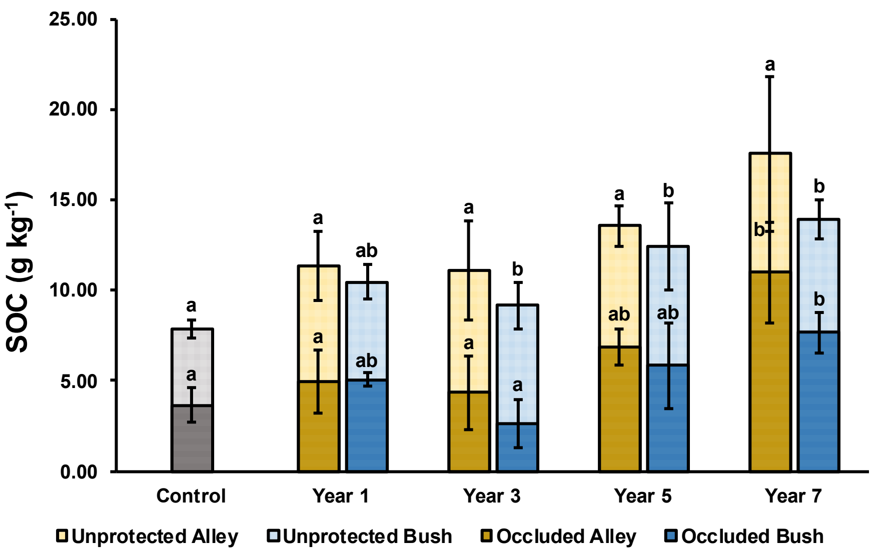

Figure 6: Thermally ~~L~~labile SOC split by occluded (hashed) and unprotected (plain) carbon pools (n=5) in the alley (yellow) and bush (blue) regimes. Error bars represent $\pm$ 1SD. For a given regime (alley or bush) dissimilar lower-case letters indicate significant ($p \leq 0.05$) differences across the timeseries. At a given timepoint, * indicates a significant difference ($p < 0.05$) between the alley and bush regimes.

3.6 Carbon Stability at Field Scale

Acknowledging proportions of alley and bush soils (60% and 40% of field area, respectively) and accommodating the influence of SBD (**Sect. 3.1; Fig. 2**), soil carbon contents (in g C kg^{-1}) (**Sect. 3.3; Fig. SI 1**) were converted to carbon stocks (t C ha^{-1}). These field scale soil carbon stocks were observed to increase (not significantly ($p \geq 0.05$)) by 1.74 t C ha^{-1} over the 7 year period relative to the control soil (from 21.98 t C ha^{-1} to 23.72 t C ha^{-1}) (**Fig. SI 2**).

When considering carbon stocks as split by labile and recalcitrant carbon pools, both were initially observed to decrease between the control and year 3 soil (**Fig. 7A**), likely in response to lower soil carbon inputs, arising from small infrequent litter drop of the young plants compared with the yearly incorporation of crop residues in the conventional system, and additionally soil disturbance during planting. The majority of this decrease occurred in the recalcitrant carbon stock, decreasing significantly ($p \leq 0.05$) from 11.54 t C ha^{-1} to 7.62 t C ha^{-1}, while labile carbon stock was observed to decrease gradually (not significantly ($p \geq 0.05$) from 10.44 t C ha^{-1} to 9.22 t C ha^{-1} (**Fig. 7A**). Following this initial decrease, ~~in~~ both labile and recalcitrant carbon stocks were observed to~~,~~ subsequently ~~yearly~~ increase~~s were observed~~ in ~~both~~ years 5 and 7, by which point labile carbon stocks were observed to exceed those in the control (**Fig. 7A**).

Over the 7 year period recalcitrant carbon stock was observed to decrease (not significantly ($p \geq 0.05$) to 9.85 t C ha^{-1} (from 11.54 t C ha^{-1}), while labile carbon stocks were observed to increase significantly ($p \leq 0.05$) to 13.87 t C ha^{-1} (from 10.44 t C ha^{-1}). Highlighting that the overall net 1.75 t C ha^{-1} increase observed in soil carbon stock over the 7 year period was comprised entirely of labile carbon (**Fig. 7A ; Fig. SI 2**).

While recalcitrant carbon stocks were observed to increase in later years, this rate of increase was less than that of the labile carbon pool (**Fig. 7A**). However, it is likely that recalcitrant carbon stocks would recover to the level of the control and possibly increase further with additional time under regenerative management. Furthermore, It is likely that the initial decreases observed in both labile and recalcitrant carbon pools related to soil disturbance and changing/reduction of organic input (crop residue) when initially transitioning from an arable to blackcurrant crop, alongside a soil priming effect from the increase in labile carbon content increasing the diversity and abundance of soil microbial communities that promote decomposition (De Graaff et al., 2010; Amin et al., 2021; Yazdanpanah et al., 2016; Lal et al., 2018). ~~Additionally~~ Indeed, it has been observed that significantly increasing labile carbon inputs to the soil can undermine the stability of recalcitrant carbon due to this enhanced priming effect (De Graaff et al., 2010), potentially causing the recalcitrant carbon loss initially observed.

Occluded carbon stocks were observed to increase marginally (not significant ($p \geq 0.05$)) between the control and year 1 soil (from 4.81 t C ha^{-1} to 4.98 t C ha^{-1}), before decreasing relative to both in the year 3 soil (not significantly ($p \geq 0.05$)) (to 3.23 t C ha^{-1}) (**Fig. 7B**). Subsequently, occluded carbon stocks were observed to increase in the years 5 and 7 soils (to 5.82 t C ha^{-1} (not significantly ($p \geq 0.05$)), and 8.21 t C ha^{-1} (significantly ($p \leq 0.05$)) ~~respectively)~~). An overall significant ($p \leq 0.05$) increase in the occluded carbon pool between the control and year 7 soils, almost doubling from 4.81 t C ha^{-1} to 8.21 t C ha^{-1} (**Fig. 7B**). While unstabilised carbon was observed to remain broadly consistent across all soils with no significant differences ($p \geq 0.05$) measured (**Fig. 7B**). Indeed, unstabilised carbon remained relatively unchanged between the control and year 7 soil (5.63 t C ha^{-1} and 5.67 t C ha^{-1} respectively). However, a small increase was observed in the year 1 soil following cultivation, increasing to 6.02 t C ha^{-1}, before converging (**Fig. 7B**). It is highlighted that the significant ($p \leq 0.05$) increase in occluded carbon corresponds to the almost identical increase in labile carbon measured in the same time period (3.40 t C ha^{-1} and 3.42 t C ha^{-1} respectively) (**Fig.**

7A/B). As such, it can be concluded that virtually all the uplift in labile carbon measured over the 7 year period had been physically protected within the stable aggregate fraction as occluded carbon. This result is important as it confirms regenerative practices have been effective in cultivating aggregate stabilities~~ty~~ capable of physically protecting what would otherwise be potentially degradable, labile, carbon. Thus, when viewed as total stabilised carbon (inclusive of recalcitrant carbon and occluded carbon) a total 1.7 t C ha^{-1} increase (not significant ($p \geq 0.05$) of potentially sequesterable carbon was observed after 7 years of regenerative management relative to the control (**Fig. 7C**).

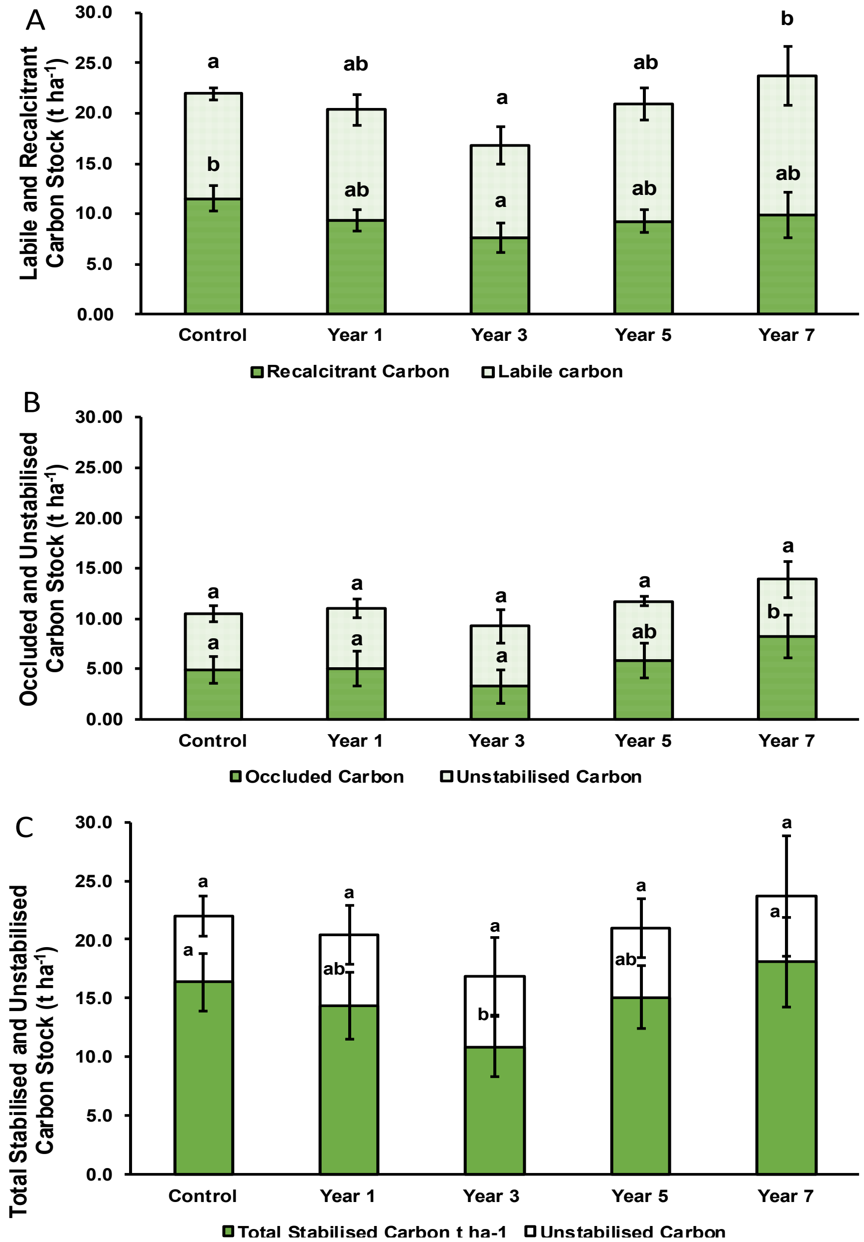

Figure 7: Carbon stock (n = 5) split by thermally recalcitrant carbon (hashed) and thermally labile carbon (plain)(A) and occluded carbon (hashed) and unstabilised carbon (plain)(B); and total stabilised carbon (Green) and unstabilised carbon (plain). Total stabilised carbon considered both recalcitrant and occluded carbon stocks. Error bars represent ± 1SD. Dissimilar lower-case letters indicate significant ($p \leq 0.05$) differences across the timeseries.

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3.6 Carbon sequestration

Efforts to increase soil carbon stocks, through methods such as regenerative agriculture, have become increasingly important strategies to support climate change mitigation (Lal, 1997; Lal et al., 2004; Lal, 2004; Smith, 2008; Smith et al., 2020; Soussana et al., 2019; Baveye et al., 2020; Keenor et al., 2021). However, it is important that we acknowledge not all carbon is equal in terms of its long-term sequestration potential: The results presented herein highlight the important nuances of both recalcitrant carbon pools and the physical protection of carbon (labile and/or recalcitrant) within soil aggregates. Given the physical protection conferred by stable soil aggregates even relatively labile carbon structures may be stabilised and physically protected in the long term as a result of their occlusion from degradative forces; with the aggregate stability governing the carbon residence time rather than its inherent stability (Schrumpf et al., 2013; Gärdenäs et al., 2011; Dungait et al., 2012; Six and Jastrow, 2002; Plante et al., 2011; Mclauchlan and Hobbie, 2004)(**Sect. 3.4; Sect. 3.5**). While the average mean residence time of aggregate stabilised carbon can range from decades to centuries, similarly to that of recalcitrant carbon, the permanence of this carbon can vary greatly between different land use types (as a result of soil management practice) (Six and Jastrow, 2002; Rabbi et al., 2013). As such It is highlighted that carbon protection is only conferred for as long as the carbon is occluded – i.e. activities that damage and destroy soil aggregates (*soil disturbance and ploughing*) can reverse these physical protections and allow for the re-entry of this carbon to the degradative labile carbon pool from which it had previously been isolated (Pandey et al., 2014; Six et al., 1998; Mclauchlan and Hobbie, 2004). Within a no till rotational system, carbon storage within stable aggregates has been observed to range between 27 – 137 years (Six and Jastrow, 2002). Thus providing significant means of stabilising and sequestering carbon in the medium- to long-term, within regeneratively managed systems (Lal, 1997, Abiven et al., 2009), and potentially on par with that of recalcitrant carbon stocks (Mao et al., 2022).

Additionally, for accurate carbon sequestration accounting to be realised, focus must be placed on the role soil bulk density plays in carbon sequestration calculations; as changes in soil carbon content often culminate in commensurate changes to the bulk density of a soil (Ruehlmann and Körschens, 2009; Smith et al., 2020; Rovira et al., 2022). Simply, as soil bulk density changes, the total volume that the soil occupies also changes (the total amount of soil remains the same, but its structure and arrangement in 3D space does not). Where soil bulk density decreases, the mass of soil per unit volume decreases. Consequently, to increase field-scale carbon stocks (assessed to a prescribed depth), SOC (g kg^{-1}) must increase at a greater rate than bulk density decreases.

In this research, SBDs oil bulk density (**Sect. 3.1**), was observed to decrease in the top 7.5cm with increased period of time under regenerative agricultural management practices, meanwhile soil carbon content (**Sect. 3.2**) was observed to increase with time. However, when changes in carbon stocks were considered on a t C ha^{-1} basis (with the a prescribed soil depth of 7.5cm), carbon stocks did not increase incrementally with increasing time (**Sect. 3.6; Fig. SI 2**). In effect there was a trade-off, as the rate of SBD decrease outpaced that of SOC increase. Consequentially, where soil carbon stocks are considered, while carbon content of the soil increased by ~65% between over the 7 year period (increasing from 16.6 g kg^{-1} in the control to 27.5 g kg^{-1} after 7 years (alley and bush soil collectively)), the total field scale increase in carbon stock was only ~8% (increasing from 21.98 t ha^{-1} to 23.72 t ha^{-1}) over the 7.5cm depth measured (**Fig. SI 2**).

Our results highlight the antagonism that exists between SBD and SOC where a prescribed soil depth is applied to soil carbon stock calculations. Thus, it is arguably more appropriate to acknowledge the depth of horizon transitions within a soil profile, and where SBD is increasing (e.g. with time under regenerative practices) to in effect increase the volume of the original soil, this new soil depth of the horizon should be used in carbon stock calculation.

Yet it is often the case that soil analysis reports do not acknowledge these changes in SBD; rather they present absolute soil carbon content (%). As a consequence, the credibility of both on-farm emissions reductions and creation of soil carbon credits is undermined, creating low integrity carbon sequestration and may lead to the abandonment of potentially significant transitional technologies due to a lack of trust. As such, the standardisation of soil carbon accountancy methods, (alongside robust validation and verification) is imperative to restoring confidence and boosting the integrity of soil based carbon sequestration (Keenor et al., 2021).

Thus, accounting for recalcitrant carbon and total stabilised carbon with respect to the SBD measured, potentially sequesterable soil carbon was measured to increase over the 7 year period by 1.7 t C ha^{-1} (**Sect. 3.6; Fig. 7C**); offering significant benefit and potential to long term carbon storage at the farm and landscape scale. When calculated against the scale of regenerative blackcurrant production at Gorgate Farm (50.3 hectares) a total potential of 314 t CO_2e could be sequestered in the top 7.5cm of soil over a 7 year period, with carbon residence on a decadal timescale.

As perennial plants, soft fruit and orchard crops offer significant opportunities for investment, engagement, and adoption of regenerative agriculture principles for soil enhancement and climate change mitigation, due to their low maintenance - long-term growing cycle and ~~the~~ minimised ~~al need for~~ soil disturbance. Were the same regenerative methods as practiced at Gorgate Farm to be applied to all UK soft fruit production (total of 10,819 hectares (Defra, 2023)), this could provide a total UK wide sequestration potential of 67,500 t CO_2e (after 7 years of continuous management).

4. Conclusion

The results of this research highlight the potential for regenerative agriculture practices to increase SOC, increase the proportions of WSA, ~~enrichment~~ and physically protect labile carbon within these aggregates - ~~and~~ thus affording opportunities~~y~~ for long-term carbon sequestration as stabilised soil carbon stocks. However, our results also bring to the fore important factors relating to soil carbon stock assessment. In particular, the antagonism between SBD decreasing at a rate greater than SOC increases; this creating a trade-off where soil carbon stocks are calculated to a standard prescribed depth, not an equivalent mass.

Thus, we highlight further research and practical guidance is needed to enable more robust soil carbon stock assessment that acknowledges i) a full pedogenic soil horizon, ii) further delineation of soil carbon pools (POM vs. MOAM) iii) the inherent recalcitrance of SOC, and iv~~ii~~) the proportion of SOC physically protected by association with soil aggregates.

Authorship contribution

BJR was the Principal Investigator and SGK the Senior Researcher for this research. Together SGK, BJR and RL undertook the investigation planning and fieldwork. Laboratory work was led by SGK with assistance in preliminary laboratory study and WSA method development from RL. SGK undertook the soil data and carbon stability analysis, statistical analysis, literature review, and the drafting of the manuscript. SGK and BJR undertook review and editing to deliver the final manuscript.

Acknowledgments

This Research was supported by the Natural Environment Research Council and ARIES DTP [grant number NE/S007334/1] with additional support provided by Greenworld Sales Ltd, Norfolk.

Competing Interests

The authors have no competing interests to declare.

Data Availability Statement

Data can be made available from the corresponding author upon request.

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