

Investigating the Effect of Oak Woodland Expansion on Soil Carbon Sequestration in Upland Pastures

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Cover image: Aerial photograph showing boundary between the ancient oak woodland and pasture on the study site, Cornwall.

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ABSTRACT

To mitigate global warming requires urgent action to both reduce greenhouse gas emissions and increase carbon sequestration. The afforestation of unproductive agricultural land is a prominent strategy for achieving this, yet considerable uncertainty surrounds the impact this will have on soil organic carbon (SOC) stocks. In an effort to address this uncertainty, this study compared SOC stocks (0-15cm) across a chronosequence, ranging an upland pasture (P) through a roughly 40-year-old upland oak woodland (RW), to an ancient oak woodland (AW). Soil samples were taken from each plot (alongside in-situ soil pH and temperature readings) and analysed for moisture content and SOC content using loss on ignition. Contrary to expectations, SOC stocks were greatest in the RW (146.17 tonnes C ha⁻¹), followed by the AW (120.93 tonnes C ha⁻¹) and lowest in the P (97.42 tonnes C ha⁻¹). These values are within the ranges previously reported in the UK, and differences between plots are largely explained by changes soil moisture and woodland management. **Ultimately, the afforestation of upland pastures with oak woodland may be an effective strategy to enhance SOC sequestration, providing average SOC gains of 26%.**

1.0 INTRODUCTION

1.1 The role of soils in climate change mitigation

The climate crisis is a global challenge that requires immediate and sustained action to mitigate future warming. To achieve this will require (at the very least) net-zero emissions by 2050 (IPCC, 2019). Indeed, almost all future scenarios consistent with limiting warming below 2°C assume a significant near-term reduction in anthropogenic greenhouse gas (GHG) emissions alongside large-scale CO₂ removal (Smith, P. *et al.*, 2015). In this context, protecting and restoring soils emerges as a vital Nature-based Solution (NbS; Griscom *et al.*, 2017). Soils contain around two-thirds of terrestrial organic carbon (1700 Gt; Friedlingstein *et al.*, 2023) and the role of Soil Organic Matter (SOM) in climate regulation has long been acknowledged (Jenkinson *et al.*, 1991). Research has highlighted significant historical losses of C from this pool (116 Gt; Sanderman *et al.*, 2017) and the potential for accelerated losses under future warming (Hicks Pries *et al.*, 2017; Jenkinson *et al.*, 1991). Building Soil Organic Carbon (SOC) therefore, serves as a NbS by not only replenishing a C sink but also by safeguarding against additional climate and land-use change driven CO₂ emissions.

1.2 Soil carbon change following afforestation

Globally, afforestation of agricultural land has the potential to sequester significant amounts of C (around 205 Gt; Bastin *et al.*, 2019). While it is well established that this would create a large C store in the form of aboveground biomass, the effects on SOC are much more uncertain (Guo and Gifford, 2002). By definition, afforestation is the conversion of non-forested land into forest. When this occurs, the change in SOC is governed by a number of factors, including prior land use (e.g., pasture or arable), tree species, management practices, soil properties (e.g., clay content, pH, moisture, and temperature), and topography (Ashwood *et al.*, 2019; Bárcena *et al.*, 2014). There has been considerable effort globally to investigate the change in SOC following afforestation, yet results are highly inconsistent. For example, studies have reported no change in SOC stocks (Degryze *et al.*, 2004), to increases (Guo and Gifford, 2002) or declines (Poeplau *et al.*, 2011). Nevertheless, many of these have shown the same general trend, with small losses in SOC immediately following afforestation before reversing and potentially increasing with stand age until pre-afforestation SOC levels are reached or surpassed (Laganière *et al.*, 2010; Paul *et al.*, 2002).

SOC is an important component of SOM, making up around 50-58% of it (Pribyl, 2010). SOM itself is the non-living part of organic matter and accumulates in soils when organic matter inputs from litterfall and rhizodeposition outweigh losses through microbial

decomposition, erosion, or leaching (Jandl *et al.*, 2007). When forests are cleared for agriculture, SOC depletion may be caused by (1) changes in soil moisture and temperature conditions that accelerate the decomposition of organic matter, (2) increased soil erosion, (3) decreased soil aggregation and physical protection of SOM, and (4) an overall reduction in above- and below-ground biomass inputs (Lal, 2005). For this reason, many agricultural soils contain SOC stocks 20-50% lower than their pre-conversion equivalents (Bárcena *et al.*, 2014; Lal, 2005). By extension, the opposite is true following afforestation, with greater organic matter inputs, reduced soil erosion, increased SOC aggregation, and changes in soil microclimate (Gaiser and Stahr, 2013; Li *et al.*, 2012). Ultimately, however, while there is considerable global pressure to convert unproductive agricultural land back to native forest (FAO, 2020), only a very small proportion of existing research has investigated the impact of this conversion on SOC stocks outside the context of commercial forestry.

1.3 UK Woodland expansion potential

There is a growing body of research demonstrating the environmental and public health benefits of trees (Ciccarese *et al.*, 2012; Griscom *et al.*, 2017). Along with the continued loss of forests worldwide, this has resulted in greater public demand for the expansion of UK woodlands. Spurred by its status as one of the least wooded countries in Europe (Forestry Commission, 2017), UK government policy now backs this public demand for woodland expansion. This is demonstrated by the implementation of a woodland carbon guarantee scheme alongside other initiatives outlined in the UK's 25-year environment plan (Defra, 2018a). However, to fulfil its climate change commitments, the UK will require significant woodland expansion in the order of 30,000 hectares annually by 2025 (Ares *et al.*, 2021). Yet, the UK landscape is a crowded landscape where most land parcels serve a range of land covers and uses. Thus, afforestation on a scale large enough to make a meaningful contribution to carbon budgets may come with significant trade-offs. For example, the trade-offs with food security and associated risks of carbon leakage (Bateman *et al.*, 2023; Doelman *et al.*, 2020). Additionally, there are both logistic and economic costs associated with large-scale tree planting (NCC, 2020), as well as the impact afforestation may have on ecosystem service (ESS) provision (Friggens *et al.*, 2020; Seddon *et al.*, 2019). Ultimately, the facilitation of natural colonisation may be a more effective, environmentally sensitive, and cheaper alternative to planting (Cook-Patton *et al.*, 2020; Crouzeilles *et al.*, 2020), although its viability is not well understood. As home to fragments of a once much more extensive woodland type, and as areas of marginal agricultural productivity, the UK uplands are a prime candidate for afforestation.

1.4 A case for woodland expansion into upland pastures

Uplands cover around 38% of UK land area (Bunce *et al.*, 2018) and could present a major opportunity for woodland expansion. Despite being internationally recognised for their heathland biodiversity and carbon stocks in the form of peatlands (Billett *et al.*, 2010; Reed *et al.*, 2009), these uplands are often ecologically and economically marginal areas, largely underutilised in terms of their potential for ESS provision (Bonn *et al.*, 2014; O'Neill *et al.*, 2020). One significant underutilised aspect of the uplands is their role as the headwaters of many UK river catchments and areas of greatest precipitation (Curtis *et al.*, 2014). This makes them key areas for floodwater attenuation efforts (Burt and Holden, 2010; Wal and Ross, 2011). A practical example of such efforts is the planting of native woodlands to slow runoff by improving soil structure, increasing landscape roughness, and reducing hydrological connectivity (Nisbet *et al.*, 2011; Robinson *et al.*, 2003). Indeed, a large part of the flood attenuation effect of woodlands is derived from increases in SOC (Burton *et al.*, 2018). Nevertheless, competing land use interests (such as livestock grazing) have caused a significant degradation of this upland landscape, diminishing its soil carbon stocks and hydrological function (Bonn *et al.*, 2009; Rowney *et al.*, 2022). However, recent shifts in agricultural policy, especially following the UK's departure from the EU, open the door to refining upland management (Bateman and Balmford, 2018). The phasing out of the EU-derived Basic Payment Scheme signals a move towards a 'public money for public goods' model for land management that prioritises key ESSs (Bateman and Balmford, 2018). This shift is expected to significantly impact upland farms, which have historically depended on these subsidies for financial viability (Defra, 2017; Hanley *et al.*, 2007).

1.5 Upland oak woodlands

Upland oak woodlands cover an estimated 70,000 to 100,000 hectares across the UK (Baarda, 2005) yet represent a fraction of a once much more widespread European habitat. Indeed, pollen records show that oaks dominated in the uplands prior to clearance in the late neolithic (6400-6000 years BP; Woodbridge *et al.*, 2014). Since then, these woodlands have undergone significant reduction following agricultural expansion across Europe, only now existing on land that is unsuitable for agriculture (Roberts *et al.*, 2018). Today, the fragments have global recognition for supporting specialist ferns, bryophyte, lichen, and animal assemblages (Baarda, 2005). Where these fragments occur along the oceanic west coast (high rainfall low temperature extremes), they support the richest bryophyte flora in Europe (Rothero, 2005). As with most upland woodlands, the principal constraint on the health and establishment of oak woodlands has been livestock and deer grazing (Barkham, 1978; Palmer *et al.*, 2004). For example, while seeds do germinate, they are often overgrazed and rarely make it past the seedling stage (Humphrey *et al.*, 2004). Given the observed decline these

woodlands across Europe, their regeneration is of heightened significance (Denman *et al.*, 2014). However, in terms of their SOC sequestration potential, little is known. Still, these woodlands typically establish on steeper sites with shallow soils, thus if sensitively encouraged the expansion of oak woodlands into upland pastures could complement rather than compete with other carbon rich habitats such as peat- and heathlands (Fletcher *et al.*, 2021; Murphy *et al.*, 2022).

1.6 How the afforestation of pasture affects soil carbon stocks

The precise agricultural land use prior to afforestation is a critical determinant of SOC sequestration rates. For example, while afforestation of former croplands is likely to enhance SOC stocks, the outcomes of afforestation on grassland or pasture are less clear and more variable (Bárcena *et al.*, 2014; Guo and Gifford, 2002). A global meta-analysis found that often, the conversion of pasture or grassland to forest produces negligible (broadleaf) or negative (coniferous) rates of SOC sequestration (Laganière *et al.*, 2010), with 75% of temperate grassland conversions showing SOC losses even after 140 years (Poeplau *et al.*, 2011). Typically, these losses occur within the initial years of conversion (Paul *et al.*, 2002). Multiple surveys of afforested mineral soil sites throughout England and Wales support this, reporting SOC losses in the 0-30 cm layer after 30 years (Hannam *et al.*, 2016). These losses are often attributed to a combination of site disturbance and a cessation of herbaceous root litter C inputs which are finer and have faster turnover rates than tree roots (Kuzyakov and Domanski, 2000). Nevertheless, the primary body of evidence for these findings is dominated by studies of non-native conifer plantations and thus making any robust predictions about how SOC stocks will respond to broadleaf afforestation is difficult.

Of the few studies that have considered broadleaf afforestation, many also report no clear difference between pasture and woodland soil C stocks. For example, an Ireland-wide study by Wellock *et al.*, (2011) found no significant difference in mineral SOC stock (0-30cm) between (broadleaf) afforested and adjacent non-forested sites (paired pre-afforestation habitat: pasture and rough grazing). Consistent with this finding, Upson *et al.*, (2016) report no net change in C stocks in the mineral soil layers for 15 years following afforestation of lowland pastures. A similar study of an ash chronosequence in Ireland found a continuous decline in SOC stocks for the first 27 years, from which point SOC stocks began to increase (Wellock, 2011). However, rates of SOC accumulation were slower than initial losses, with SOC stocks in 47-year ash woodlands only 79% of pre-afforestation grasslands. The only UK-based review of upland grasslands reports inconclusive evidence for the magnitude or direction of SOC change following afforestation, citing a lack of relevant UK datasets (Reynolds, 2007). SOC stocks of

woodlands established on arable soils are shown to increase with stand age, as the trees continually add litter to the soil (Ashwood *et al.*, 2019). This is unlikely to be the case for pasture as baseline SOC stocks are often similar to or greater than those in mature forests and ancient woodlands (Ashwood *et al.*, 2019; Garten and Ashwood, 2002; Post and Kwon, 2000). Ultimately, it remains uncertain to what extent findings from lowland soils can be extended upland agricultural systems, particularly in light of inconsistencies in sampling methodology.

1.7 Outline of research questions and hypotheses

Given the lack of clarity on the impact that afforestation of pasture has on SOC stocks, as well as the general lack of research on native broadleaved afforestation, this study aims to address the following questions:

- (1) Can the expansion of oak woodland into upland pastures effectively increase topsoil SOC stocks?
- (2) Does the SOC sequestration potential of these woodlands increase over time?

Furthermore, given that upland pastures are generally located on unproductive sloped land this study will also address the following question:

- (3) How does the relationship between slope gradient and SOC change following afforestation?

2. METHODOLOGY

2.1 Study area

Located on the western fringe of Bodmin Moor, Cornwall, Cabilla manor farm is a 300-acre hill farm with 100 acres of woodland and 200 acres of grade 4 pasture – grazed intensively since 1972 and exclusively by highland cattle and sheep since 2001. The site is bisected by the river Bedalder, a second order tributary of the river Fowey (**Figure 1**). The site averages 210m in elevation and is subject to a cool-temperate oceanic climate with an annual mean temperature range of 7.21 to 13.45°C. The region receives 1431.7 mm y⁻¹ in rainfall, of which ~20% falls over the summer months (June to August; Met Office, 2020). Soils on the estate comprise freely draining acid loamy soils over granite (**Appendix A1**). The three plots chosen for sampling are a grazed pasture (P), an adjacent ancient oak woodland (AW) and a nearby naturally regenerated 30 to 40-year

GB national outlines [Shape], Scale: 1:250,000, Updated 2005, Ordnance Survey, GB. Using EDINA Digimap Ordnance Survey Service Downloaded: July 2023.
High Resolution (25 cm) Vertical Aerial Imagery [Tiff], Scale: 1:500, Updated 2019, Ordnance Survey, GB. Using: EDINA Digimap Aerial, Downloaded: July 2023.
OS Open Roads [Shape], Scale 1:25,000, Updated April 2022, Ordnance Survey, GB. Using: EDINA Digimap Ordnance Survey Service, Downloaded: July 2023.
OS VectorMap@ Local [Shape], Scale 1:3000–1:20000, Updated April 2022, Ordnance Survey, GB. Using: EDINA Digimap Ordnance Survey Service, Downloaded: July 2023.

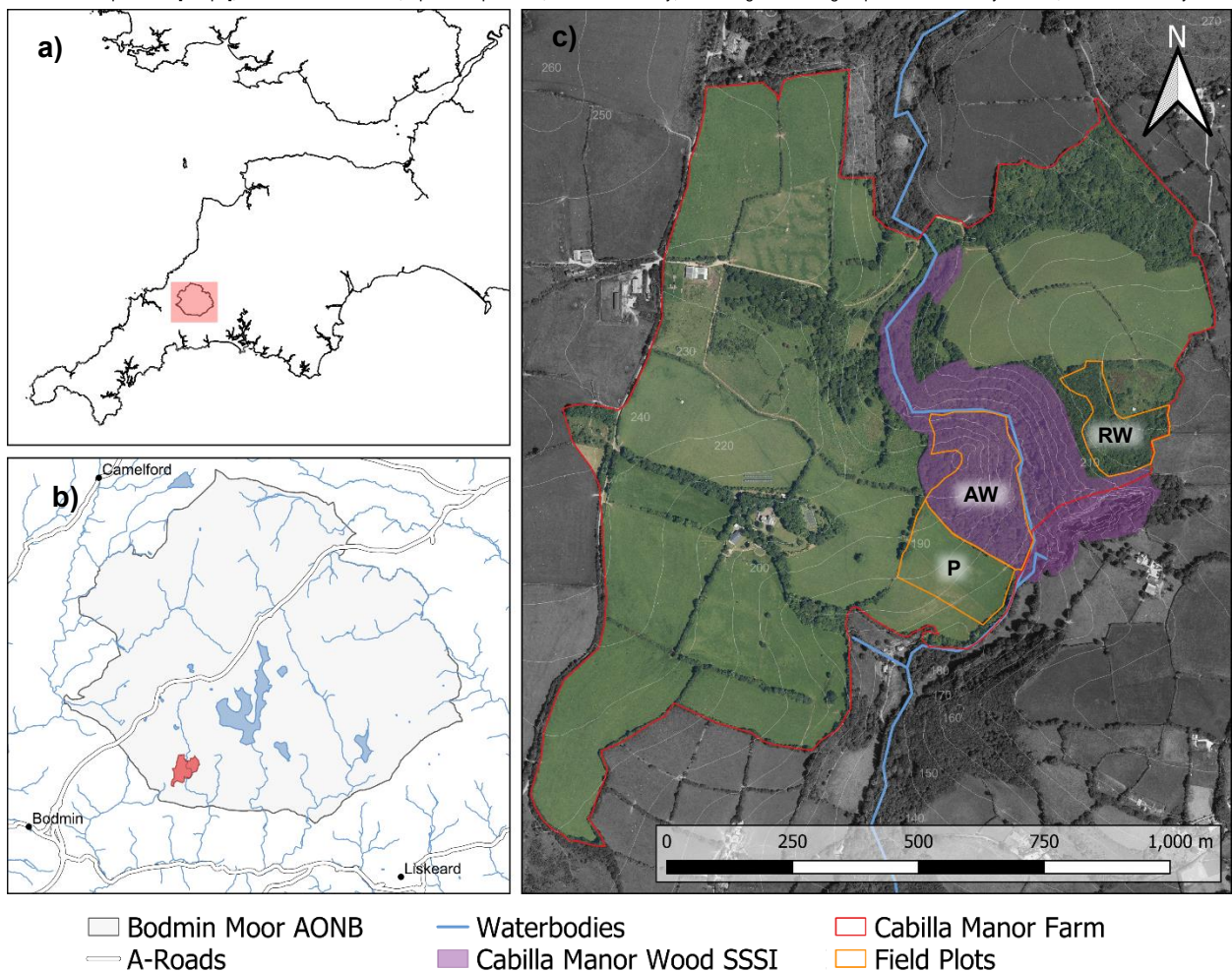


Figure 1: a) Location of Bodmin Moor within Cornwall. b) Location of Cabilla Manor Farm within Bodmin Moor. c) Site map including contours to show topography. Elevation ranges from 250 to 180 m at river level.

oak woodland, referred to as recent woodland (RW). Both woodlands fall under the W1 (UKHab) community of upland oakwood and have similar understories dominated by bracken and bramble in places. Both woodlands are dominated by pedunculate oak (*Quercus robur*) and have low species and structural diversity. The AW has a previous management history of oak timber and coppice production. For the RW, historical mapping (Epoch 1) classifies the plot as open rough ground (1875-1901). The plot is last mapped as pasture/open ground with gorse and bracken in 1980 (Epoch m7). Subsequent maps classify the plot as broadleaved woodland.

2.2 Experimental design

Woodland soils have greater variability and more possible sources of error than agricultural soils for SOC estimation. The following methodology for sample site selection, collection, and laboratory analysis, therefore, was designed primarily to alleviate these sources of error as much as possible. It is based on the recommendations laid out by Vanguelova *et al.*, (2016). In addition, topographic factors like slope aspect and gradient have a significant impact on SOC variability within a landscape, making them important factors to control for (Zhu *et al.*, 2019). To achieve this, National LiDAR Programme DTM (1m resolution) data were downloaded from Defra (Defra, 2018b), and QGIS 3.28.8 was used to analyse slope characteristics and establish sampling points based on a stratified random sampling strategy (**Figure 2**).

To determine the location and number of soil sampling points, a stepwise process was followed. First, slope aspect in each plot was compared. Only South facing slopes were present in two plots, so north facing slopes were excluded from further analysis. Similarly, plot RW had a maximum slope gradient of 20 degrees, so no samples were taken on slopes greater than this. To ensure all slope gradients were sampled, plots were divided into four slope classes by equal interval classification: 0-5, 5-10, 10-15, and 15-20. To account for woodland soil variability, Vanguelova *et al.*, (2016) recommend a minimum of 4 sample points per 0.25 ha woodland plot. With this in mind, the largest plot when filtered by south facing slopes was 2.38 ha. Therefore, a minimum of 38 samples from each plot are required. To select sampling locations, the QGIS feature 'Random Points in Polygons' was used to select 10 points from each slope class, totalling 40 per plot. As collecting and processing bulk density (BD) samples is time intensive (Vanguelova *et al.*, 2016), only one sample per slope class was taken. Once processed these would be averaged out providing a single BD for each plot.

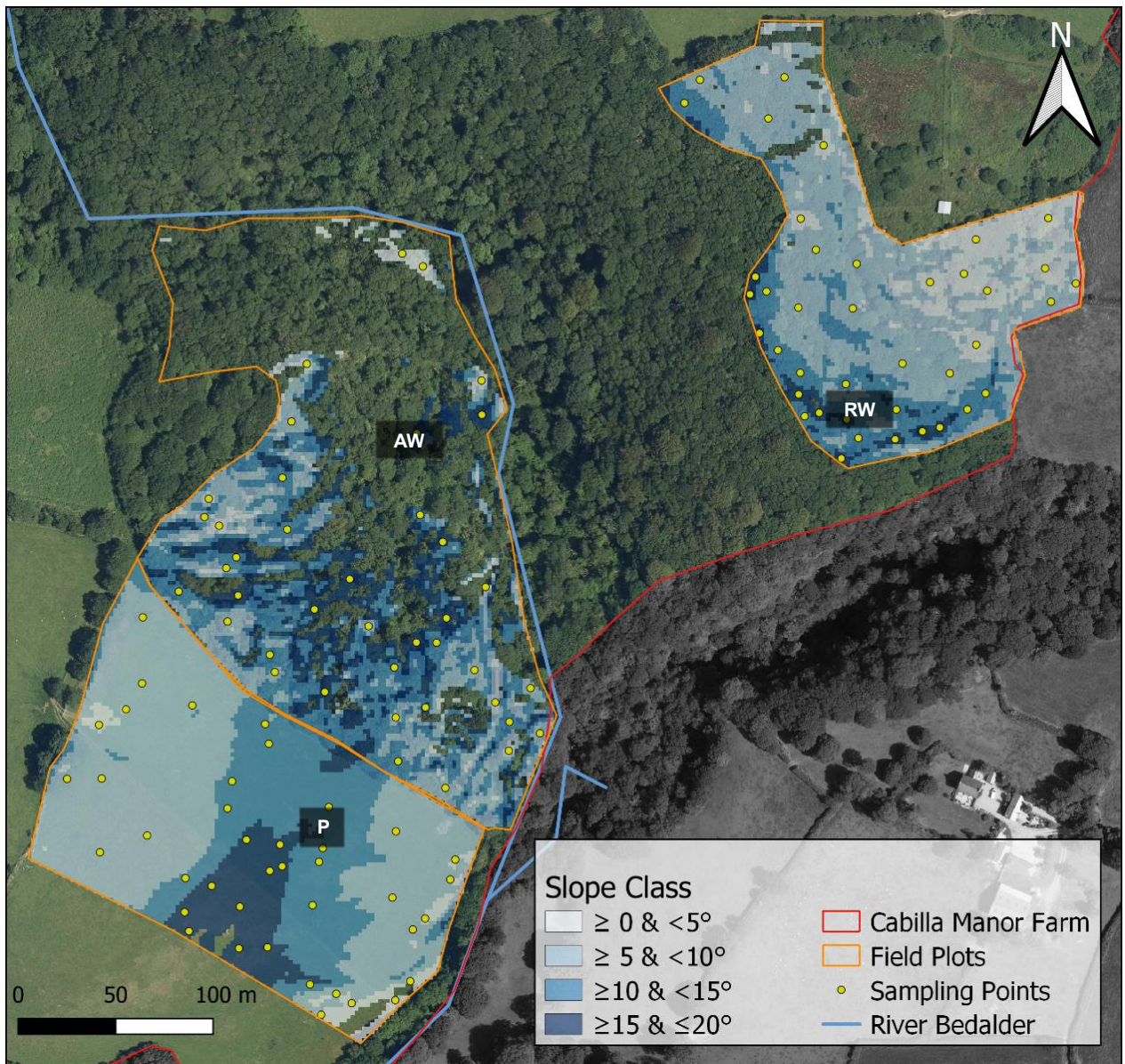


Figure 2: Location of soil sampling points. At each point, soil cores were taken, and soil temperature and pH were measured in situ. The reason a large portion of the AW is excluded from sampling is that these areas are either north-facing or have a slope gradient $>20^\circ$.

2.3 Sample collection

Samples were collected throughout August 2023 when the forest floor is at its minimum mass (i.e., before autumn litterfall; **Figure 3**). At each sample site, surface vegetation and all recognisable leaf litter were cleared to expose bare soil which then defined 0 cm depth. For SOM samples, a soil gouge was used to extract the upper 15 cm of soil (**Figure 4**). Once extracted, samples were placed in airtight polythene bags and frozen until laboratory analysis. Additionally, at each sample point measurements of soil pH and temperature were taken using a HI-99121 waterproof pH & temperature meter. Finally, for BD samples, a 50.16 mm internal diameter and 49.52 mm height (97.86 cm³ volume) stainless steel cylinder was pressed into the ground, carefully removed and then trimmed

to remove excess soil. The soil core was extruded into a sample bag and frozen until laboratory analysis. This was repeated for three depths at BD sampling points (e.g., 0-5, 5-10, 10-15 cm).



Figure 3: Pictures of site conditions at the time of sample collection (August 2023) in the P (left), RW (middle), and AW (right). While understories of woodlands are similar, the AW has evidently greater coverage of bracken.



Figure 4: Picture of soil gouge used with vegetation scraped away in the P (left) and a sample to 15 cm depth (right), authors photography.

Deeper sampling (subsoil) is sometimes recommended to better reflect long-term change in SOC following afforestation (Shi *et al.*, 2013), however, preliminary soil pits in each plot were found to be <30-40cm deep before reaching substantial pieces of

underlying bedrock, which were impenetrable by hand tools. Sampling from the upper 0-10 cm depth is the most common (Vanguelova *et al.*, 2016) and would be sufficient in this instance. However, a lack of standardised sampling protocols in the literature, alongside the variability of soils makes comparisons between studies unreliable. Sampling from the upper 0-15 cm depth instead, reflects a nationally representative topsoil layer (Carey *et al.*, 2008) and allows for comparisons to many UK-based studies (Gregg *et al.*, 2021; Vanguelova *et al.*, 2013) as well as to established countryside survey soil health benchmarks in the UK (Feeney *et al.*, 2023). Nevertheless, the upper 15 cm is shown to store greater proportions of SOC compared to deeper layers and better reflects recent management changes (Bárcena *et al.*, 2014; Mayer *et al.*, 2020; Paul *et al.*, 2002).

2.4 Laboratory analysis

Loss on Ignition (LOI) was chosen for estimating SOM for its accuracy, simplicity, and affordability. To alleviate the effect that mineralogical composition has on SOM/LOI relationships (Kasozi *et al.*, 2009), samples were taken from plots with the same underlying soil type. As with sampling depth, a general agreement on the methodology for estimating soil carbon content using LOI is lacking. Therefore, the methodology used here is synthesised from similar studies while considering potential sources of error (**Figure 5**). For example, while it has been suggested that sieving samples to 2 mm may underestimate the carbon content of forest soils (Laganière *et al.*, 2010), the majority of studies still use and recommend 2 mm.

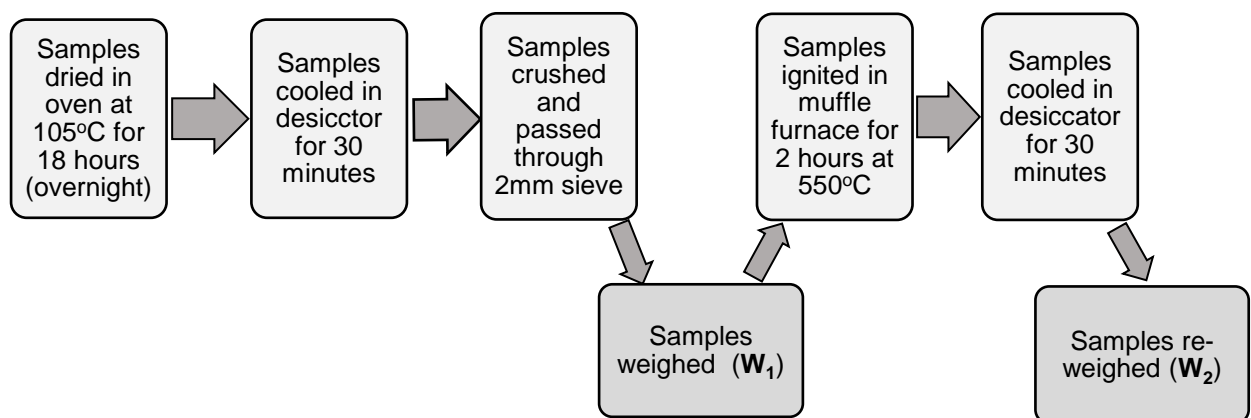


Figure 5: Flow chart demonstrating methodology used to determine SOM content from each soil sample.

Soil moisture content was calculated for each sample as a function of oven dried sample weight (**Equation 1**). SOM was calculated using sample weights prior to and after ignition (**Equation 2**). For bulk density, samples were oven dried for 18 hours at 105°C. The

sample dry weight was divided by 97.86 (sample volume) to give a dry soil weight by unit volume in g cm^{-3} (**Equation 3**).

$$\text{Eq. (1)} \quad \text{Soil moisture (\%)} = \frac{\text{Loss in soil weight (g)}}{\text{Weight of oven dried soil (g)}} \times 100$$

$$\text{Eq. (2)} \quad \text{SOM (\%)} = \frac{W_1 - W_2}{W_1} \times 100$$

$W_1 = \text{Weight before ignition}$

$W_2 = \text{Weight after ignition}$

$$\text{Eq. (3)} \quad \text{BD (g cm}^{-3}\text{)} = \frac{\text{Weight of oven dried soil (g)}}{\text{Sample volume (cm}^3\text{)}}$$

2.5 Data analysis and interpretation

2.5.1 Carbon stock calculation

SOC was derived by employing a conversion factor of 1.818 to SOM (i.e., assuming organic carbon constitutes 55% of SOM; **Equation 4**). The most frequently used conversion factor is, 1.724 (58%), although this may often overestimate SOC in woodland soils (Pribyl, 2010). Instead, the conversion factor chosen here was to allow for comparison to other UK-wide soil surveys (Emmett *et al.*, 2010), and is within the appropriate range observed by other studies of forest soils (Bhatti and Bauer, 2002; De Vos *et al.*, 2005). Next, topsoil (0-15cm) carbon stocks for each plot were calculated using average SOC and BD measurements (**Equation 5**).

$$\text{Eq. (4)} \quad \text{SOC (\%)} = 0.55 \times \text{SOM (\%)}$$

$$\text{Eq. (5)} \quad \text{Soil Carbon Stock} = \text{SOC} \times \text{BD} \times \text{soil depth}$$

$(\text{tonnes ha}^{-1}) \quad (\%) \quad (\text{g cm}^{-3}) \quad (\text{cm})$

2.5.2 Woodland composition

Woodland species composition was determined using point-centred quarter data. This included tree density, species dominance, and importance values. A canopy height

model was created in QGIS 3.28.8 using ~3 cm resolution aerial photogrammetry of the site, captured in September 2023 and provided by the site owner. By excluding cells with a height value <1 m, this model was used to derive canopy cover (%) and average canopy height of each woodland plot.

2.5.3 Statistical analysis

Analyses were performed using R version 4.3.1 (R Core Team, 2017) and interpreted using a 95% confidence interval (significance criterion $p \leq 0.05$). All figures were produced using base R packages and ggplot2 (Wickham, 2009). First, each soil property (SOC, pH, temperature, moisture) in each plot (P, RW, AW) was tested for parametricity using the Shapiro-Wilkes test alongside visual inspection of histograms and QQ plot. Depending on their distribution, different statistical tests were used to compare soil properties between plots. Where these were non-parametric, a Wilcoxon rank-sum test was applied. For parametric variables, where variance (determined by F-tests) was equal (unequal) an independent (Welch's) t-test was performed to determine any significant differences. Soil properties were plotted against each other, and correlations tested using Spearman's correlation test if parametric, and Pearson's if non-parametric. If correlations were significant, linear regression was used to determine the presence and strength of an explanatory relationship. Where necessary, multivariate regression analysis was conducted to identify the combination of variables that had the greatest impact on the dependent variable.

3. RESULTS

3.1 Comparison of carbon stocks and explanatory variables

Every measured soil property was significantly different between plots, with the exception of pH and BD which showed no significant difference between the RW and AW (**Table 1**). On average, samples from the pasture contained the lowest SOC (7.22%), followed by the ancient woodland (11.52%), and then recent woodland (19.12%). Accounting for BD ($P = 0.90$, $RW = 0.51$, $AW = 0.70$), C stocks in the upper 15 cm of soil increased sequentially from P (97.42 tonnes C ha⁻¹) to AW (120.93 tonnes C ha⁻¹) and then RW (146.17 tonnes C ha⁻¹; **Figure 6**). This represents a 19.44% increase from P to AW, and a 33.35% increase from P to RW. The same trend is observed in soil moisture, being lowest in the P (54.44%), followed by AW (61.23%), and then RW (69.16%). Soil temperature was greatest in P (20.35 °C), followed by RW (17.55 °C), and then AW (16.33 °C). Soil pH, likewise, was greatest in P (5.90) although indistinguishable between woodlands (both 5.38).

Table 1: Results of T-tests between each of the measured variables in each plot. Blue text represents independent (not welch's) t-test, and black p values are from Wilcoxon rank sum t-tests.

	Site	P	RW	AW		P	RW	AW
SOM	P				Moisture			
	RW	p <0.001				p <0.001		
	AW	p <0.001	p <0.001			p <0.001	p <0.001	
C Stock	P				Temp			
	RW	p <0.001				p <0.001		
	AW	p = 0.041	p = 0.002			p <0.001	p <0.001	
BD	P				pH			
	RW	p <0.001				p <0.001		
	AW	p <0.001	p = 0.064			p <0.001	p = 0.797	

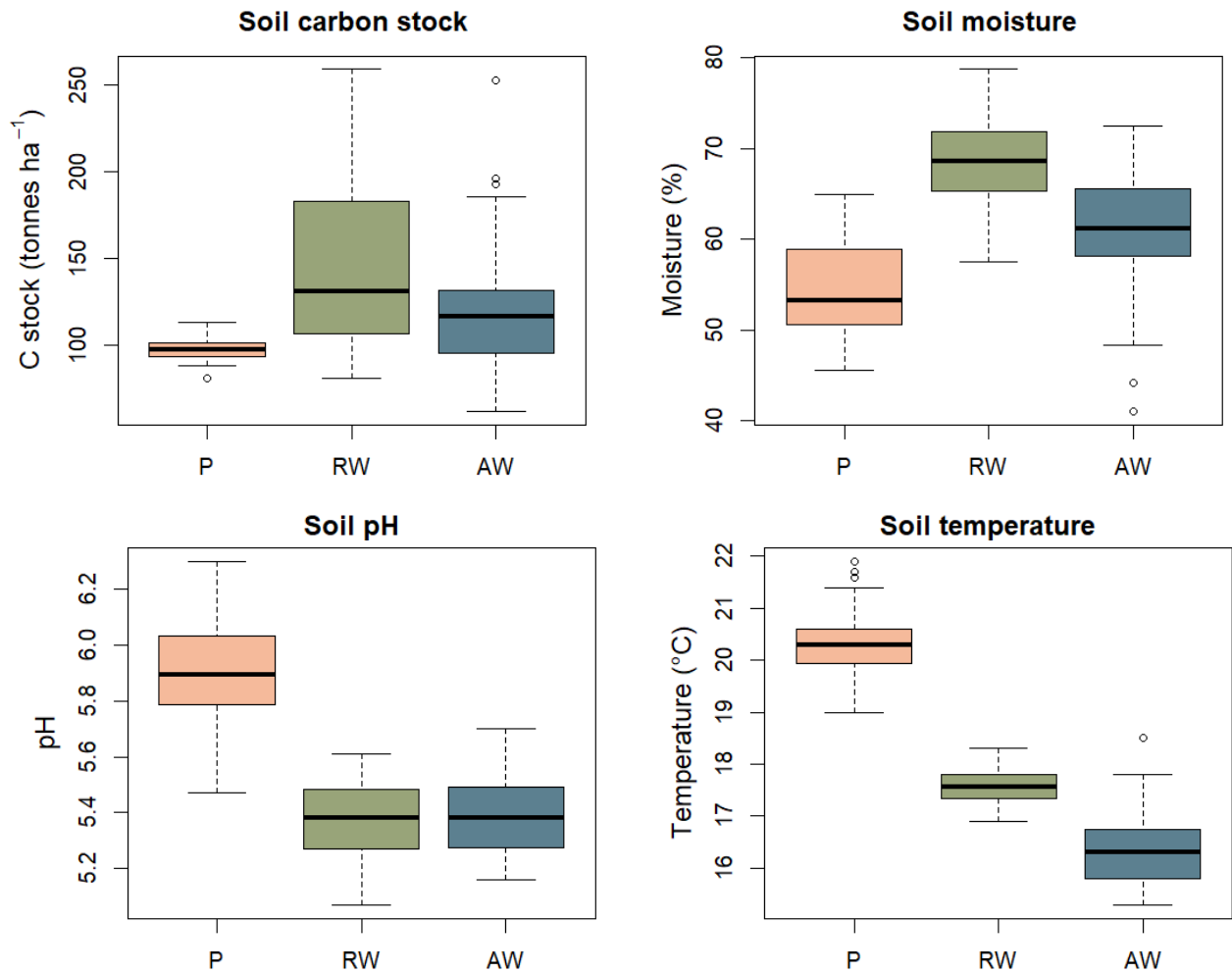


Figure 6: Boxplots showing differences between variables. Solid black bar within box body represents median value. Each measurement is significantly different with the exception of pH between woodlands.

3.2 Relationships between SOC and other soil properties

While no significant relationships were found between the measured soil properties at the site-level, some were observed at the plot-level. Simple linear analysis revealed that soil moisture was a significant predictor of SOC in the P ($R^2 = 0.53$, $p = <0.001$), indicating that for every 1% increase in soil moisture, SOC is expected to increase by approximately 0.064% (**Figure 7**). Similarly, soil moisture was a significant predictor of log SOC in the RW ($R^2 = 0.61$, $p = <0.001$) and AW ($R^2 = 0.68$, $p = <0.001$), with a 1% increase in moisture causing an expected 5.106% and 3.854% increase in SOC, respectively.

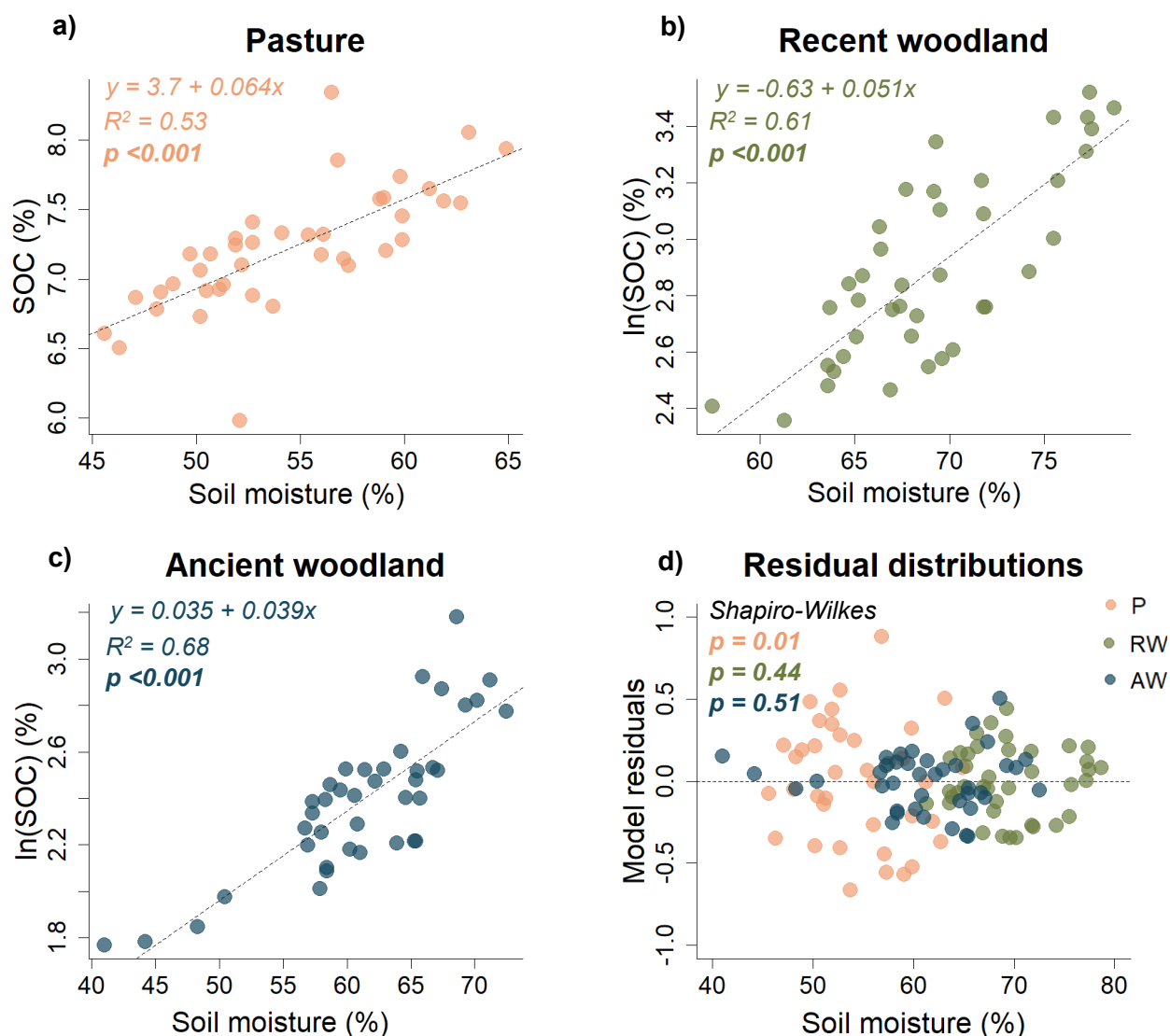


Figure 7: Relationships between soil moisture and SOC for each plot. Note the log-linear relationships in plot (b) RW, and (c) AW. Dashed lines in plots a, b, and c represent the model fit. Plot d shows residual distributions for each regression line. Despite the Shapiro-Wilk test showing a p-value <0.05 for the 'P' residuals, normality is supported by visual inspection of the QQ plot and histogram.

Additionally, analysis revealed that slope gradient explains about 34% of the variation in SOC in P ($R^2=0.34$, $p < 0.001$), although no such relationship is observed in the woodlands (**Figure 8**). In line with this, no clear spatial distribution in SOC stock is observed in either plot (**Figure 9**). Within the pasture, multivariate regression revealed SOC was most strongly dependent on soil moisture and slope gradient ($R^2=0.60$, $p < 0.001$). Moisture alone explained a similar amount of variation in SOC ($R^2=0.53$, $p < 0.001$), though the overall model was stronger with slope gradient included.

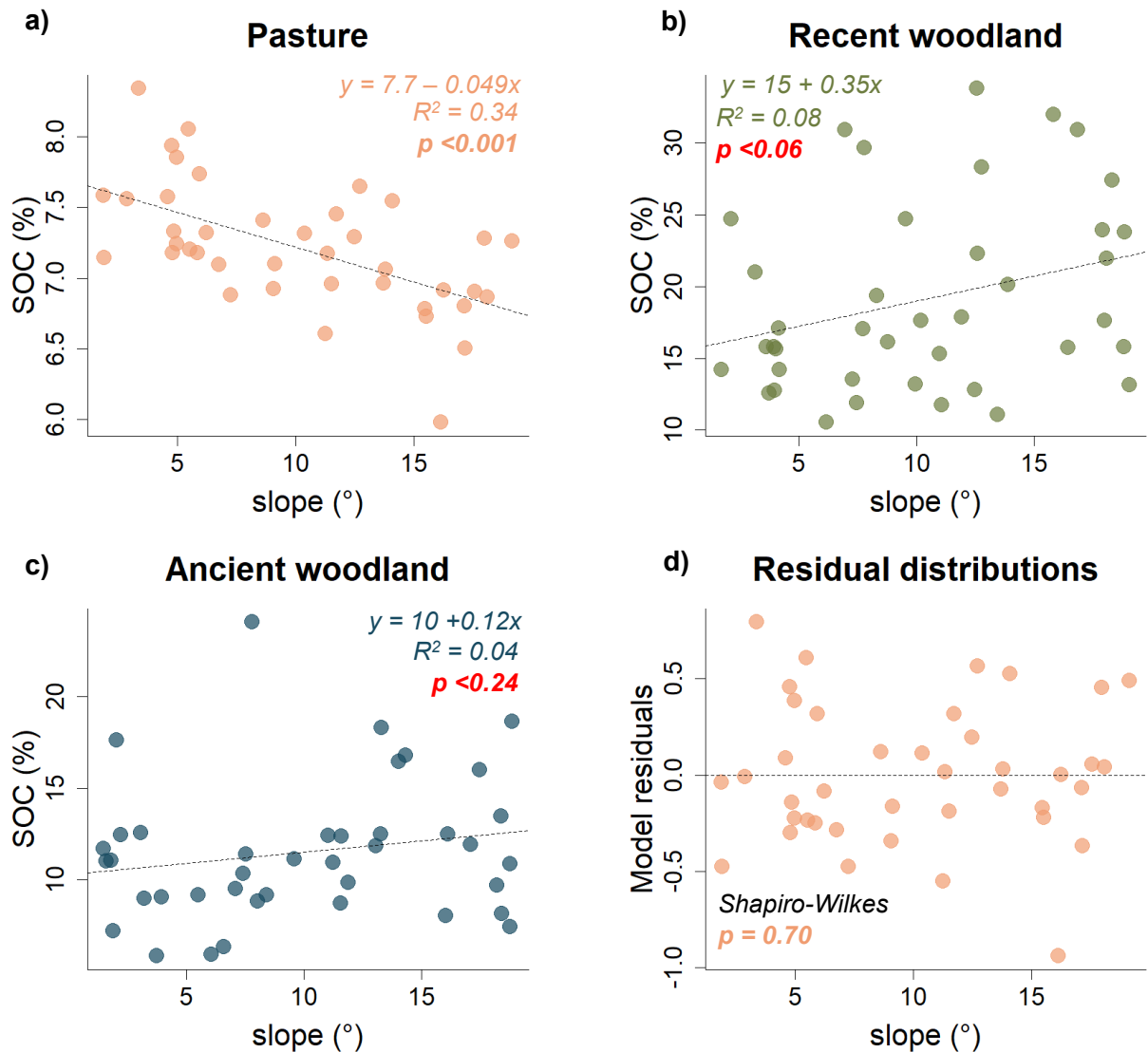


Figure 8: Relationships between slope gradient and SOC in the P (a), RW (b), and AW (c). The only significant relationship observed is in P. Dashed lines in plots a, b, and c represent the model fit. Plot d shows residual distributions for P regression line.

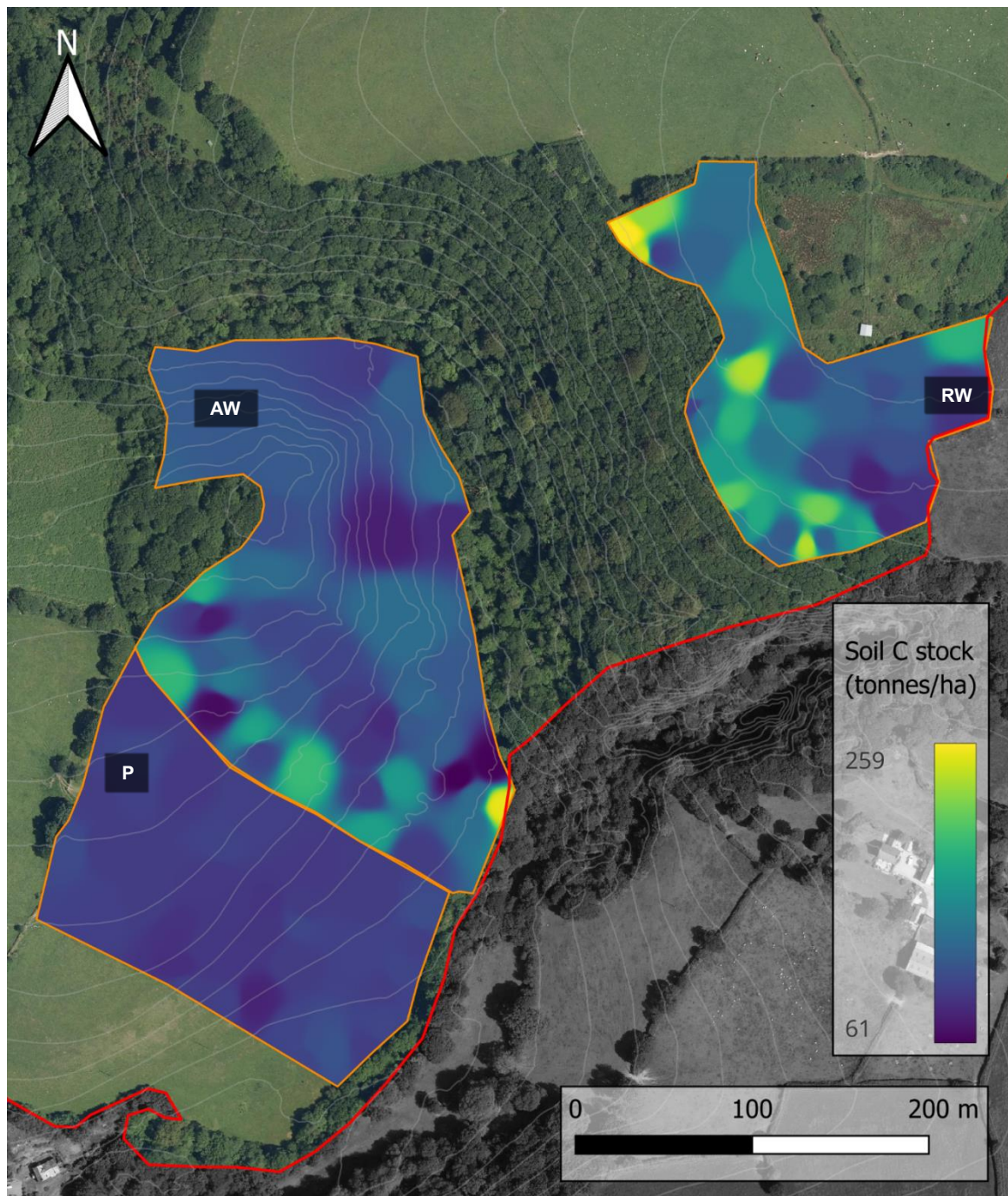


Figure 9: 2D interpolation map of SOC stocks in each plot. No clear topographic trend in SOC stock is observed. P clearly presenting with lowest averages SOC stock.

3.3 Woodland tree species, density, canopy cover, and height comparison

PCQ data revealed that tree density was slightly lower in the AW compared to RW (561 and 660 trees ha⁻¹, respectively), but not significantly different ($t = -0.83$, $p = 0.45$). In both woodlands, only pedunculate oak (*Quercus robur*) and hazel (*Corylus avellana*) were recorded, oak being the dominant tree in both cases (AW = 98.77%, RW = 94.52%). Importance values for these species were almost indistinguishable between woodlands (AW oak = 224.66, hazel = 75.34; RW oak = 226.67, hazel = 73.33). Comparing tree size

between woodlands, the diameter at breast height (DBH) of oaks were significantly larger in the AW (44.73 cm) than the RW (25.17 cm; **Figure 10**). Average tree height was likewise greater in the AW (12.83 m) compared to the RW (6.68 m), although overall canopy cover was similar (AW = 98.05%, RW = 95.87%).

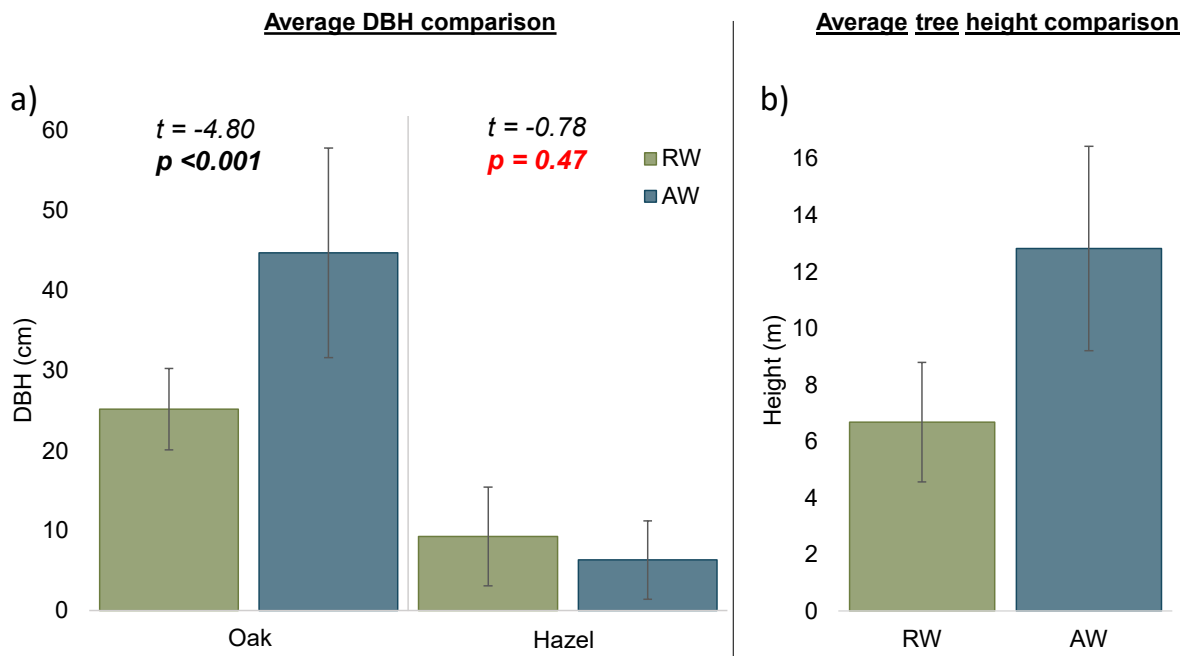


Figure 10: Bar chart (a) comparing the average DBH of tree species in the AW and RW. P-values represent results from t-tests and show the DBH of oak but not hazel to be significantly different between woodlands. Bar chart b presents the canopy height model derived average tree height of both woodlands. Error bars represent standard deviation. No overlap suggests a significant difference in tree height.

4. DISCUSSION

4.1 How SOC stocks compare to other studies

The SOC stocks (0-15 cm) reported here were found to be significantly higher in both the AW (120.93 tonnes C ha⁻¹) and RW (146.17 tonnes C ha⁻¹) compared to pasture (97.42 tonnes C ha⁻¹). This represents around a 33% increase from the P to the RW, and a 19% increase from P to AW. While these SOC stocks are relatively high, they are within the range of expected values for these land covers in Britain. For example, a survey of upland grasslands (including permanent pasture) found SOC stocks of 74 tonnes C ha⁻¹ (59 to 101; Eze et al., 2018). Using the soil health benchmarks created by Feeney et al., (2023) to calculate SOC stocks for modified grassland yields a similar result of 68 tonnes C ha⁻¹ (46 to 123). This range is particularly relevant, being calculated for modified grassland on light coarse textured soils in a high rainfall environment (>1000 mm yr⁻¹) i.e., soil and climate conditions similar to the study site. Using the same methodology to calculate SOC stocks for broadleaved woodlands returns 74 tonnes C ha⁻¹ (32 to 162; Feeney et al., 2023). SOC stocks (0-15 cm) for oak woodlands specifically were only available from a single study of urban woodlands by Edmondson et al., (2014), who found SOC stocks of 65 tonnes ha⁻¹ (20 to 90).

4.2 Explaining the variations in SOC between pasture and woodlands

Considering the findings of earlier studies, it was expected that SOC stocks in the P would be either statistically indistinguishable or slightly higher than those in the RW and AW (Guo and Gifford, 2002; Upson *et al.*, 2016; Wellock *et al.*, 2011). This is particularly true given that afforestation of pasture in high rainfall areas almost always leads to SOC losses (Kirschbaum *et al.*, 2008). However, the findings from this study largely contradict these expectations, with SOC stocks being greatest in the RW, followed by AW, and lowest in the P. This may be explained by differences in the other measured variables: Soil temperature, pH, and moisture. Firstly, the relationship between soil temperature and soil respiration (decomposition) is well established, with greater temperatures leading to accelerated rates of SOM decomposition and thus SOC loss (Davidson and Janssens, 2006; Yvon-Durocher *et al.*, 2012). With this in mind, the greater soil temperature recorded in the P (20.35 °C) compared to the RW (17.55 °C) and AW (16.33 °C) could be inhibiting SOC accumulation. However, in this instance, temperature is unlikely to be exerting the greatest control on SOC stocks, as no significant relationship between temperature and SOC was observed in any plot.

Soil pH is also known to have a meaningful impact on SOC through acting as a control on microbial communities and enzyme activity. Indeed, soil microbial efficiency is demonstrated to be maximal around pH 6 to 7 (Liao *et al.*, 2016), with a pH outside this

range reducing rates of decomposition and enhancing SOC accumulation (Malik *et al.*, 2018). Significant associations between soil pH and SOC have been reported by field studies though the relationship varies substantially between land uses. For example, in forest soils pH explains only 4.3% of the variation in SOC, whereas for grasslands the figure is 59.4% (Liao *et al.*, 2016). Nevertheless, in the current study there were no significant associations between soil pH and SOC in any plot. Thus, while soil pH was closer to optimal in the P (5.9) compared to the RW (5.38) or AW (5.38), this is unlikely to explain the observed differences in SOC. However, this study does relate to the broader afforestation literature in that pH was greatest in the P (Mayer *et al.*, 2020). This is likely explained by the high acidity of oak leaves and the organic acids produced during litter decomposition (Yuan *et al.*, 2023).

Differences in soil moisture between plots are the most likely explanation for the variation in SOC, being lowest in the P (54.44%), followed by the AW (61.23%), then RW (69.16%). Much like temperature and pH, moisture affects SOC by governing SOM decomposition rates and nutrient availability (Larson *et al.*, 2023). Soil respiration rates, which are linked to decomposition and thus carbon loss, tend to increase as the soil dries in temperate hardwood forests (Davidson *et al.*, 1998). Comparing woodlands and grasslands in the Eastern USA, Smith and Johnson (2004) also found decreased moisture in woodlands to be associated with lower rates of soil respiration. In fact, under many land-uses soil moisture is often the single most influential predictor variable for SOC (Kerr and Ochsner, 2020). This was also found to be the case in the current study, with moisture being the only soil property to exhibit a significant association with SOC in each plot, explaining 53%, 61%, and 68% of the variability in SOC in the P, RW, and AW respectively. Where this study diverges from previous findings is that soil moisture increased from P to woodlands. Typically, afforestation of pasture has been associated with a decrease in soil moisture, as rates of evapotranspiration increase (Nosetto *et al.*, 2012; Upson *et al.*, 2016).

Slope gradient was also shown to be a significant predictor of SOC in the pasture, though multivariate regression confirmed the strength of the relationship was weaker than between moisture and SOC. Still, the negative relationship between slope gradient found here is contrary to expectations, with prior grassland studies reporting either no relationship (Zhang, Xueyao *et al.*, 2018), or a slight positive relationship (Fissore *et al.*, 2017). It is possible that the P is losing soil on steeper slopes through the same mechanism as croplands (Borrelli *et al.*, 2017), though this is unlikely as slope gradient is shown to have no effect on soil loss in grasslands (Zhang, Xuexian *et al.*, 2021). Nevertheless, that the relationship observed in the P is not present in either woodland highlights the capacity of woodlands to disrupt soil movement and SOC loss (Hou *et al.*,

2020). A global meta-analysis found this woodland effect to be greatest on slopes of 10-20° (Yang *et al.*, 2023). Generally, then, the findings from this present study contradict prior research on the relationship between slope gradient, land cover, and SOC. This underscores the importance of considering slope gradient in any future studies that examine SOC.

4.3 Explaining the difference in woodland SOC stocks

As mineral SOC is shown to increase over time with oak woodland age (Benham *et al.*, 2012), it was expected that the AW would have greater SOC stocks than RW. This was shown to not be the case here, and while soil moisture is likely the major driver of SOC variation between woodlands (being greater in the RW), it is unlikely to be the sole explanation. Results from the PCQ analysis revealed that both the RW and AW had effectively the same tree species composition, density, and canopy cover. Thus, while tree density is sometimes shown to have a significant positive association with SOC stocks (Mayer *et al.*, 2020), this factor is unlikely to be at play here. Rather, other factors like management history (e.g., timber extraction and coppicing) likely explain the lower SOC stocks in AW compared to RW. For example, coppicing is demonstrated to lead to higher rates of CO₂ efflux and a short-term decline in SOC stocks (Darenova *et al.*, 2016). Although other studies have demonstrated long-term soil (and thus SOC) losses following coppicing (Maciej Serda *et al.*, 2019). The discrepancy between observed and expected SOC values in the RW and AW also highlights the limitations of chronosequence studies. For example, the exact pre-afforestation habitat is unknown and these initial SOC stocks may have been significantly higher than those in the current P.

4.4 Implications for upland management and future research

Although the afforestation of upland pasture may have increased SOC stocks in this study, the exact reason for this increase is yet to be confirmed. As prior studies more often demonstrated losses of SOC, if the gains observed in this study are related to soil moisture, then future afforestation projects may benefit from monitoring soil moisture and soil respiration where possible. Still, confirming this assumption should be a priority for future studies. Several methods may be used for this though a simplified litter bag experiment using tea bags can be a cheap and effective approach (Keuskamp *et al.*, 2013). Alternatively, estimating soil respiration through measurements of soil CO₂ efflux either in situ using a static chamber technique (Dou *et al.*, 2016) or in the lab from collected soil samples (Ortiz *et al.*, 2016) is an option.

Additionally, while sampling the upper 15 cm of soil was the chosen method in this study for its capacity to best reflect recent management changes (Vanguelova *et al.*, 2016),

around 50% of total SOC can be stored below 30 cm (Laganière *et al.*, 2010; Rumpel and Kögel-Knabner, 2010). Indeed, older stands are shown to contain a greater proportion of SOC below 20 cm: 30% compared to 20% in younger stands (Hale *et al.*, 2019). Given this, it is recommended that future studies consider sampling at greater depths, particularly when studying older woodlands. This would provide a more comprehensive understanding of SOC distribution and the impact of management changes on C sequestration in these ecosystems. Moreover, in the instances where the afforestation of pasture has been shown to increase SOC stocks, almost all of this gain has been attributed to the forest floor (Laganière *et al.*, 2010). This forest floor may contain >16% of the total SOC down to 1 m (De Vos *et al.*, 2015). Therefore, future research may also benefit from studying the role of the forest floor in enhancing SOC sequestration following the afforestation of pasture.

5. CONCLUSION

In the context of climate change mitigation, this study set out to find whether the afforestation of upland pastures with oak woodland can be an effective strategy for enhancing SOC stocks. While the SOC stocks reported in this study were relatively high for each plot, they were all within the range previously reported for these land covers in the UK. However, while previous studies often found negligible to negative rates of SOC sequestration following the afforestation of pasture, this study found SOC stocks to be lowest in the pasture, followed by the ancient oak woodland, and highest in the 30–40-year-old naturally regenerated oak woodland. Lower SOC stocks in the P compared to woodlands are likely the result of a combination of factors: (1) greater soil moisture content in the woodlands, and (2) the significant negative relationship between slope gradient and SOC in the P. This present study also contradicted past studies which demonstrate that ancient woodlands have greater SOC stocks than younger stands. As these woodlands had virtually indistinguishable species composition, tree density, and canopy cover, the observed difference in SOC is also likely explained by variations in soil moisture. Management history is also likely to have a small impact, with the AW being extensively managed in the past for oak timber and coppice. This is known to deplete SOC stocks through increasing rates of soil erosion soil respiration. Ultimately, because of the uncertainty surrounding the impact of pasture afforestation on SOC stocks, the woodland carbon code currently assumes no change. Instead, the results presented here demonstrate that the afforestation of upland pastures with oak woodlands can increase SOC stocks in the 0-15 cm layer by up to 33.35% over the course of a 40-year period.

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