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Long-Term Impact of Management on Deep Soil Carbon and Soil Health in Mediterranean
Agroecosystems

By

JESSICA LEIGH CHIARTAS
DISSERTATION

Submitted in partial satisfaction of the requirements for the degree of

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Abstract

Agriculture is under increasing pressure to produce more food with less environmental impacts and in the face of a changing climate. Management practices capable of sequestering soil carbon (C) and improving overall soil health hold promise for sustainable intensification, as well as climate change mitigation and adaptation. As market and policy-based incentives develop to support these practices, however, it is critical that adequate sampling protocols, minimum viable data sets, and thresholds of management responses to soil health indicators are identified across the diversity of cropping systems and edaphoclimatic conditions.

Much of the research into the impacts of agricultural management on soil C and soil health have been conducted in the Midwest, over the short-term, and to a shallow depth. Soil C dynamics and other soil health indicators are strongly influenced by climate and mineralogy, necessitating more research across a range of edaphoclimatic conditions. Further, detectable changes in soil C take decades to accrue, requiring long-term research. Proper accounting of changes in C stocks on a given acreage for climate mitigation strategies and economic incentive programs also necessitates sampling to a sufficient depth (minimum 1 meter or a root-limiting layer).

Using long-term, on-farm interventions, controlling for cropping system, climate and soil type, this work investigates the impact of soil health practices on soil C in surface and subsurface soils, as well as on a suite of physical, chemical, and biological soil properties commonly used to assess soil health. Deep soil cores at a long-term, industrial scale, agricultural research station in a Mediterranean-type climate indicated that 19 years of cover cropping with annual composted poultry manure applications (4t ha^{-1}) increased soil C to a depth of 200 cm by $+21.8\text{ Mg ha}^{-1}$ relative to a -4.8 Mg ha^{-1} loss under conventional management (Chapter 1). Trends also indicated

potential losses of -13.4 Mg ha^{-1} under conventional management with cover cropping, despite increases of $+1.4 \text{ Mg ha}^{-1}$ in the surface 0-30 cm, stressing the importance of deep soil sampling for greenhouse gas accounting purposes.

Continuing the theme of deep soil C, a nearby regional survey of 10+ yr old hedgerows and adjacent cultivated fields across four soil types showed a strong impact of hedgerows on soil C to a depth of 100 cm, with an average difference of 3.85 kg C m^{-2} (0-100 cm) and few differences across the four soil types (Chapter 2). Most differences occurred in the surface 0-10 cm and the subsoil at 50-100 cm, indicating a dual role of surface management (litter accumulation, reduced disturbance) and deep, woody perennial roots. Soil type differences were only apparent in one of the four soil types, which differed substantially in parent material, mineralogy, and degree of weathering. Soil type did not influence the management effect and may indicate broad potential for hedgerows as a climate mitigation strategy. The magnitude of this strategy is limited, however, by the extent of hedgerows on a given farm/ranch.

Revegetation of field margins with hedgerows also had a positive impact on a broad suite of physical, chemical, and biological parameters (0-20 cm) commonly associated with soil health (Chapter 3). Hedgerow values were greater than cultivated fields for nearly every indicator in the surface 0-10 cm, commonly 2-3 times greater. Fewer, smaller differences were observed at 10-20 cm. Total soil C and N, available C, microbial biomass C, aggregate stability, and surface hardness were some of the most sensitive and least variable indicators of management type. Texture, pH, and bulk density were more indicative of soil type. A composite of variables was necessary to explain most of the variation in the data, indicating the complexity of soil health.

Introduction

In an increasingly polarizing time, farmers/ranchers, scientists, policymakers, and the general public are finding common ground around agriculture and improved soil health as a solution to global challenges. Whereas agriculture is one of the most vulnerable sectors to the impacts of climate change, it also holds great potential for both climate change mitigation and adaptation through the sequestration of soil carbon (C) and overall improvement of soil health (Paustian et al. 2016; Amelung et al. 2020; Bossio et al. 2020). Market and policy-based programs are emerging to incentivize soil C sequestration and soil health management practices, with a growing interest in a broader suite of ecosystem services (Minasny et al. 2017; Vermeulen et al. 2019; Norris et al. 2020).

Soil C in the form of organic matter is central to soil health and sustainability in agricultural systems (Weil & Magdoff 2004; Lal et al. 2016), but a reductionist approach by markets and policy-based incentive programs threatens to undermine attempts to address climate change. There is a paucity of data across the range of cropping systems and edaphoclimatic conditions (Devine et al. 2021; Kögel-Knabner & Amelung 2021) and to a sufficient sampling depth (Harrison et al. 2011; Olson et al. 2014) to adequately predict/model soil C stock changes. There is also a strong need for standardized sampling protocols, minimum viable data sets, and indicator thresholds across the diversity of contexts to support on-the-ground sampling efforts (Fine et al. 2017; Nunes et al. 2020; Norris et al. 2020). Further, soil and other earth scientists are increasingly tempering the carbon exuberance, citing limitations due to carbon saturation, stoichiometric constraints, and socio-cultural barriers (Amundson & Biardeau, 2018; Poulton et al. 2018; van Groenigen 2019).

Many of the practices touted for their carbon sequestration potential, however, have other well-documented benefits that provide climate adaptation and/or resilience by promoting soil health and overall ecosystem function (Lal et al. 2016; Vermeulen et al. 2019). Fixating on soil health as a means for sequestering soil C -- a silver bullet solution for climate change --

overshadows other deeply related existential crises including soil erosion, water availability, and biodiversity loss. By shifting our perspective to soil health as an agronomic and environmental bundle, we can diversify our societal and economic risks, so that in the event we do not sequester 4 kg C per 1000 kg of soil each year (Minasny et al. 2017), our investments in agriculture will still provide valuable returns in the form of reduced CH₄/N₂O emissions, improved water quality and use efficiency, increased biodiversity, and other ecosystem services.

Compost applications and cover cropping have been well documented to increase surface soil C in California and other Mediterranean agroecosystems (Poudel et al. 2001; Kong et al. 2005; Veneestra et al. 2007; Aguilera et al. 2013; Mitchell et al. 2015), as well as to improve soil function including improved aggregate stability (Kong et al. 2005), increased water holding capacity (Brown & Cotton 2011), reduced nitrate leaching (Poudel et al. 2001), and greater yield stability (Li et al. 2020). Less is known, however, about the long-term impacts on deep soil C and how that might affect overall greenhouse gas (GHG) accounting. Likewise, much is known about the positive impacts of hedgerows on ecosystem function, including increased infiltration and water storage (Marshall and Moonen 2002; Ghazavi et al. 2008; Holden et al. 2019); reduced nitrate leaching and runoff (Long et al. 2010; van Vooren et al. 2017; Thomas and Abbot 2018); increased pollinators, birds, and beneficial insects; and reduced pest pressure overall (Morandin et al. 2016; Heath et al. 2017; Long et al. 2017). Far less is known, however, about the impact on deep soil C or soil health (Thiel et al. 2015; Cardinael et al. 2017).

Chapter 1 capitalizes off a long-term, industrial scale agricultural research station (The Century Experiment at Russell Ranch) to assess the impacts of 19 years of various cropping rotations, nutrient management systems, and irrigation strategies on soil organic carbon to a depth of 200 cm. Chapter 2 utilizes a historic, multi-stakeholder campaign in Yolo County, California, to “Bring Farm Edges Back to Life,” to continue the theme of assessing the long-term impacts of management on soil carbon to a minimum recommended depth of 100 cm. By conducting a regional survey, we had the opportunity to explore the impact of revegetation with hedgerows across soil types, while controlling for variations in climate and cropping system.

Chapter 3 expands on the investigation of long-term hedgerow plantings to understand the impacts of revegetation of field margins on physical, chemical, and biological parameters of soil health in surface soils, as well as which indicators may be most effective in intensive row crop systems in a Mediterranean-type climate.

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Chapter 1: Deep soil inventories reveal that impacts of cover crops and compost on soil carbon sequestration differ in surface and sub-surface soils¹

1.1 Abstract

Getting more carbon (C) into soil via organic inputs is a key strategy for increasing long-term soil C storage and improving the climate change mitigation and adaptation potential of agricultural systems. Traditionally, research has focused on the top ~30 cm of soil assuming that management does not impact the deeper soil profile. A long-term experiment in California's Mediterranean climate measured changes in both surface and subsurface soil C in maize-tomato and wheat-fallow cropping systems. Soil C concentrations and stocks were measured at the initiation of the experiment and year 19, at five depth increments down to 2 meters, accounting for changes in bulk density. In maize-tomato rotations, addition of winter cover crops (WCC) + composted poultry manure in an organic system increased soil C by 7.9 Mg C ha⁻¹ (21%) in the top 30 cm of soil, by 13.9 Mg C ha⁻¹ from 30-200 cm, resulting in total gains of 21.8 Mg C ha⁻¹ (12.6%). When WCC was added to a conventionally managed system, soil C stocks increased by 1.44 Mg C ha⁻¹ (+3.5%) in the top 30 cm but decreased by 14.86 Mg C ha⁻¹ (-10.8%) in the 30 to 200 cm layer, resulting in an overall loss of 13.4 Mg C ha⁻¹. There was slight decline in C in the conventional system (no compost or WCC). Soil C did not change substantially across the 2 m profile in wheat-fallow systems with N fertilizer, winter cover crops, or irrigation alone, but decreased by 5.6% with no inputs. If only the surface soil had been measured, we would have falsely concluded that adding WCC to conventionally managed crops increases soil C and dramatically underestimated the capacity of the ORG to sequester C. Our results provide concrete examples of the importance, particularly for carbon markets, of full accounting of soil C throughout the entire soil profile as management practices do have large impacts on subsoil C.

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1.2 Introduction

Soil organic carbon (SOC) is a cornerstone of agroecosystem sustainability, as a driver of soil structure, nutrient cycling, water dynamics, microbial activity, and biodiversity. Increasing soil organic C reallocates atmospheric CO₂ to long term organic pools, offsetting greenhouse gas emissions of CO₂, and increasing the resilience of agroecosystems. Soil organic C is also a very common indicator of soil health, receiving considerable attention from growers, environmental advocates, and policymakers alike (Lal 2010; Lehman et al. 2015; California Department of Food and Agriculture 2017). Recent international policy to mitigate global impacts aims to sequester 0.4% C/yr in agricultural soils (French Ministry of Agriculture and Food 2015), attracting widespread investment, scrutiny, and criticism (Chabbi et al. 2017; Minasny et al. 2017; Amundson and Biardeau 2018). Given the likelihood of future incentives to build soil C in agricultural soils, it is essential to understand and accurately estimate potential gains and losses associated with different management practices. Gaining this knowledge for rainfed and irrigated systems in a semi-arid climate, and across a diversity of inputs, will be key for prioritizing management strategies that sequester soil C.

Crop management practices that increase long-term C include cultivation of perennial crops and/or cultivation of pastures (Armstrong et al. 2003; Lal 2004; Follett and Reed 2010; Sanford et al. 2012) and applications of organic amendments (e.g., manure, compost) (Zhang et al. 2012; Brar et al. 2013; Poulton et al. 2018). Net losses, on the other hand, can result from excessive tillage, overgrazing and fallowing (Hernanz et al. 2009; Maia et al. 2009). Increases of 0.3 to 4.0 Mg C ha⁻¹ yr⁻¹ (Smith et al. 1997; Su et al. 2006; Lee et al. 2007) have been observed widely in manured systems. In semiarid rainfed systems, frequent fallowing resulted in no net

soil C change, whereas systems cropped annually resulted in gains of 0.44 to 1.32 Mg C ha⁻¹ yr⁻¹ (Peterson et al. 1998; Curtin et al. 2000). Cropland soils in semiarid regions were identified by Lal (2004) to have high C sequestration potential if irrigation efficiency is maximized and tillage minimized (West and Marland 2002).

Winter cover crops (WCC) increase biomass production of cropping systems, often increasing soil organic C. A global meta-analysis of 30 studies found that cover crops increase soil C stocks by 0.32 Mg C ha⁻¹ yr⁻¹ but most studies only analyzed the top 30 cm of soil (Poeplau and Don 2015). In contrast, Poulton et al. (2018) observed losses of 0.55 Mg C ha⁻¹ yr⁻¹ in temperate annual cropping systems with WCC. Studies more commonly report increases rather than decreases in soil C with WCC. How and to what extent WCC influences C, especially throughout the soil profile, needs more study along a co-management gradient, considering different WCC species and climates.

While Mediterranean agroecosystems represent some of the most diverse, productive, and economically valuable systems in the world, we know surprisingly little about how management affects SOC in these systems (DeGryze et al. 2009; Suddick et al. 2011; Aguilera et al. 2013). These systems tend to be under-saturated in SOC and have potential for sequestering additional C if water and nutrient constraints can be overcome (Jones et al., 2005; West and Six, 2007; Romanya and Rovira, 2011; Munoz-Rojas et al., 2012). An estimated 75% of Mediterranean agroecosystems contain less than 2% soil organic matter (SOM) (Van-Camp et al., 2004). Increasing SOC could increase the adaptive capacity of these regions, as they are particularly susceptible to rising temperatures and drought expected with climate change (Romanya et al. 2007; Munoz-Rojas et al. 2012).

Few studies have examined soil C below 30 cm (Poeplau and Don 2015; Govaerts et al. 2009; Follett et al. 2012; Kirkby et al. 2013). The reported average sampling depth is only 25.7 cm (Aguilera et al. 2013) and overlooks much of soil's potential to sequester C, as soil below 30 cm can hold between 30 to 75% of its total C stocks (Jobbagy and Jackson 2000; Rumpel and Kögel-Knabner 2010; Harrison et al. 2011; Chaopricha and Marin-Spiotta, 2013). Radiocarbon dating showing increased mean residence times with depth suggests that deep soil C is inherently more stable and resistant to decomposition (Paul et al. 1997; Kaiser and Guggenberger 2003; Rumpel et al. 2004; Chabbi et al. 2009). The subsoil generally contains greater reactive surface areas (von Lutzow 2008) and SOM exists there predominately in organo-mineral complexes, which are considered a key mechanism for long-term stabilization of SOM (Kögel-Knabner et al., 2008; Rumpel 2015). Moreover, deeper layers are not subjected to tillage, a physical disturbance that increases oxidation of SOC.

Long-term experiments provide unique opportunities for understanding C dynamics. Outcomes can be linked to well-documented management practices and evaluated for how they impact overall sustainability. The Century Experiment is a cropping systems trial initiated at the University of California, Davis, in 1993. The experiment examines the long-term sustainability of soil-health building practices (such as WCC and compost), frequent fallow, and irrigation in maize-tomato and wheat-fallow crop rotations on 0.4 ha plots. This experiment is one of few long-term studies in irrigated Mediterranean agroecosystems (Davidson and Janssens 2006; Potter et al. 2012). Previous research at the Century Experiment found that after 10 years of consistent management, soil C stocks were greater in organic than conventional tomato–maize systems, with and without WCC (Kong et al., 2005). However, analyses were restricted to the

surface 0-15 cm of soil and evaluated only a small subset of the cropping systems under comparison.

Here we describe changes in soil C concentrations and stocks to a 2-meter depth, and across 9 farming systems, after 19 years of management at the Century Experiment. Our principal questions were: (1) How do long-term inputs of different carbon sources and management affect soil C sequestration in row crops? (2) Do patterns of C sequestration across different depths vary between crops and management practices, and (3) Can patterns of C sequestration observed in the top 30 cm of soil predict C sequestration throughout the deeper soil profile. We predicted that 1) intensive annual vegetable/grain systems with the highest organic C inputs—e.g., from WCC and/or compost—will show the greatest soil C gains; 2) low-intensity systems with fallow will lose soil C throughout the soil profile; and 3) the direction of soil C change will differ among soil layers throughout the 2 m profile, particularly between the disturbed cultivated layer (0 to 30 cm) and the undisturbed subsoil (60 to 200 cm).

1.3 Materials and Methods

1.3.1 Experimental site and cropping systems design

The Century Experiment (previously known as Long-Term Research on Agricultural Systems, LTRAS), is located at the Russell Ranch Sustainable Agriculture Facility near University of California, Davis (38°32'24" N, 121°52'12" W), with an elevation of 16 m. The site is located in California's northern Central Valley in an alluvial plain of the Putah Creek watershed, which contains soil deposited from what is now the Berryessa Reservoir and includes the Great Valley Complex, Sonoma Volcanics, and Quaternary surface deposits (Wolf et al. 2018; Shlemon et al. 2000; Wagner et al. 2011). The area was originally oak savannah and

perennial grassland, ecotypes which have been mostly replaced by annual row crop agriculture. The climate is semi-arid, Mediterranean, characterized by wet winters and hot, dry summers.

The Century Experiment was established in 1993 to test the long-term impacts of wheat (*Triticum aestivum* L.) or maize (*Lycopersicum esculentum* Mill.) based cash crop rotations common to northern California on productivity, profitability, resource-use efficiency, environmental impacts, and ecosystem services. The site has two soil types: (i) Yolo silt loam (Fine-silty, mixed, superactive, nonacid, thermic Mollic Xerofluvents) and (ii) Rincon silty clay loam (fine, smectitic, thermic Mollic Haploxeralfs). Detailed soil horizon information (classification and depths) can be found in the Century Experiment published dataset in Wolf et al. (2018). Prior to layout of the Century plots, the site was surveyed for soil characteristics and laid out in a randomized complete block design with three blocks. Two blocks are placed on the Rincon silty clay loam, and the third block is located on the Yolo silt loam. The experiment includes 9 cropping systems in 2-yr rotations (Table 1.1), on 0.4-ha (64 x 64 m) replicate plots. Each cropping system is replicated 6 times (2 plots per block), with both crops present within a block every year (3 crop within system replicates, 1 plot per block). Disking operations are restricted to 15 to 20 cm depths, and tillage to a maximum depth of 25 cm.

1.3.2 Maize-based systems management

Maize-based systems compare conventional vs. organic crop and soil management, and consist of 1) conventional maize–tomato with synthetic fertilizer, pesticides, and winter fallow (CONV); 2) certified organic maize–tomato with composted poultry manure and WCC (ORG), and 3) a hybrid system with synthetic fertilizer, pesticides, and WCC (CONV+WCC; Table 1.1).

In ORG, composted poultry manure was broadcast and incorporated in March at an average rate of 4 t ha⁻¹. Beds were rolled to prepare the seedbed. Maize was planted in two rows per bed in all maize–tomato systems in early April with 56 kg N ha⁻¹ 8-24-6 starter fertilizer. Maize in the CONV and CONV+WCC systems was fertilized via side-dressing in one application, or two split applications, with ammonium sulfate at a total rate of 180 kg N ha⁻¹. Maize in all systems was furrow irrigated with an average of 33.6 mm per year, a minimum of 17.6 mm in 1995 and a maximum of 43.5 mm in 2004. Maize was harvested with a full-scale combine in late September or early October. Stalks were chopped and disked to incorporate residues. In CONV, maize was followed by winter fallow, whereas in CONV+WCC and ORG systems, a WCC mix of field pea (*Pisum sativum* L.) and hairy vetch (*Vicia villosa* Roth) was planted from 1994 through 2001, and in 2002 through 2012, field pea was replaced with faba bean (*Vicia faba* L.) and cereal oat (*Avena sativa* L.). WCC were planted in November on the top of the beds and terminated by mowing and incorporated with 2 to 3 disking operations in March.

Maize was followed by tomato in all rotations. Tomatoes were started in a commercial greenhouse and transplanted in April or May into 150 cm beds prepared by listing and rolling. A pre-plant herbicide was applied and incorporated in CONV and CONV+WCC systems (Table 1.1) and tomatoes were planted with 56 kg N ha⁻¹ 8-24-6 starter fertilizer. CONV and CONV+WCC tomatoes were side-dressed in one or two split applications with ammonium sulfate at a total rate of 112 kg N ha⁻¹. In ORG, composted poultry manure at an average rate of 4 t ha⁻¹ was broadcasted, incorporated and rolled prior to tomato transplanting or maize seeding. Tomatoes in all systems were furrow irrigated as described for maize. Tomatoes were mechanically harvested in August and green fruits and vine residues incorporated by shallow

disking. Tomatoes in CONV were followed by winter fallow. In CONV+WCC and ORG systems, a WCC mix followed tomatoes as previously described.

No synthetic biocides were applied in ORG. One cultivation was performed between beds in each crop phase of conventionally managed systems and three to four cultivations in ORG to control weeds. In CONV and CONV+WCC systems, metribuzin and glyphosate in maize and trifluralin in tomato were applied prior to planting.

1.3.3 Wheat-based systems management

Wheat cropping systems were designed to represent dryland wheat–fallow systems on semi-marginal lands in the foothills of California. The wheat systems compare the effect of N fertilizer, supplemental winter irrigation, and leguminous N via WCC and include: 1) rainfed wheat–fallow control with no inputs (RWF), 2) rainfed wheat–fallow + N fertilizer (RWF+N), 3) rainfed wheat–fallow with WCC (RWF+WCC), 4) irrigated wheat–fallow with winter supplemental irrigation and no fertilizer (IWF), and 5) irrigated wheat–fallow with supplemental irrigation and N fertilizer (IWF+N).

Winter wheat was planted in November, harvested by combine in July, straw incorporated by two shallow disking operations, then fallowed from August to the following November with the exception of RWF+WCC (Table 1.1). Fertilized wheat received 56 kg ha⁻¹ 15-15-15 fertilizer at planting, and an additional 90 and 112 kg ha⁻¹ urea in March in the rainfed and irrigated systems, respectively. Rainfed systems received an average 366.1 mm precipitation from 1993 to 2012 (minimum of 101.6 and maximum of 615.7 mm) (SI Table 1.1). Irrigated wheat systems received supplemental irrigation of 80 mm per year except in 1995, 1998, 2000, 2005, and 2008-2012 due to sufficient precipitation. During fallow, weeds were managed with

one herbicide application and four disking operations, beginning after wheat harvest. RWF+WCC (Table 1.1) only received fertility from WCC, planted in November following wheat harvest. The WCC mix included hairy vetch (*Vicia villosa* Roth.) and ‘Magnus’ pea (*Pisum sativum* L.) from 1993 to 2006, and faba bean (*Vicia faba* L.), hairy vetch, and ‘Montezuma’ oat (*Avena sativa* L.) from 2007 to 2012. In March or April, WCC were terminated with 2 to 3 diskings, as necessary. Soils remained fallow until planting of wheat in November.

1.3.4 Plant and compost sampling and analysis

After machine harvest, aboveground plant biomass was measured by cutting crop residues at the soil surface at two locations per plot (1.5 m² in maize and tomato and 1.0 m² in wheat). WCC incorporation was measured by cutting aboveground biomass at the soil surface in a 4.5 m² area. Root biomass was not measured during the study period. Crop residues and WCC biomass were dried for 4 days at 60°C and ground to 2 mm. Total C and N of incorporated aboveground biomass and composted manure were determined each year, using dry combustion on an ECS 4010 Costech Elemental Analyzer (Costech Analytical Technologies, Valencia, CA) multiplying percent C and N by total biomass. Total aboveground C inputs were calculated by summing crop residue C, WCC C, and compost C incorporated per plot per year.

1.3.5 Soil sampling

At the onset of the experiment in September 1993, 3-cm inner diameter soil cores were collected from all 6 replicates in all nine cropping systems. Samples were composited from 10 random locations within plots in depth increments of 0 to 15 cm, 15 to 30 cm, 30 to 60 cm, 60 to 100 cm layer, and 100 to 200 cm. Sampling by depth layers, rather than soil horizons, was chosen because soils at this site are very young (< 6,000 y), and horizon are relatively homogeneous compared to more highly weathered soils. Horizon boundaries are gradual and

Table 1.1. Maize- and wheat-based cropping systems in the Century Experiment, and inputs, from which soil C was measured 1993 and 2012. “Supplemental flood” irrigation refers to the application of irrigation water to wheat when winter rainfall was insufficient to meet wheat water needs. CONV = conventional; WCC = winter legume cover crop mix; ORG = organic; RWF = rainfed wheat–fallow; N = nitrogen fertilizer; IWF = irrigated wheat–fallow

Cash Crop	Abbreviation	Crop Rotation	Irrigation	Fertilizer Source	Annual N Rate kg ha ⁻¹
	<i>CONV</i>	Maize - Tomato	Furrow	Synthetic N Fertilizer	168
Maize	<i>CONV+WCC</i>	WCC/Maize - WCC/Tomato	Furrow	Synthetic N Fertilizer + WCC	168
	<i>ORG</i>	WCC/Maize - WCC/Tomato	Furrow	Poultry Manure Compost + WCC	150-200 [†]
	<i>RWF</i>	Wheat - Fallow	None	None	0
	<i>RWF+N</i>	Wheat - Fallow	None	Synthetic N Fertilizer	146
Wheat	<i>RWF+WCC</i>	Wheat - WCC/Fallow	None	WCC	0
	<i>IWF</i>	Wheat - Fallow	Supplemental Sprinkler	None	168
	<i>IWF+N</i>	Wheat - Fallow	Supplemental Sprinkler	Synthetic N Fertilizer	168

[†] Depending on N composition of poultry manure compost

diffuse, changing over vertical distances >15 cm. In September 2012, 3-cm-diameter soil cores were collected from all 6 replicates of the 9 cropping systems. Samples were composited from 6 random locations per plot in similar depth increments, then air-dried, sieved to <2mm, and archived in glass vials at room temperature.

Bulk density samples were collected with a Giddings hydraulic probe in both the 1993 and 2012 soil samplings. In 1993, bulk density was collected in 0 to 25 cm, 25 to 50 cm depth layers with an 8.25-cm diameter probe. In 2012, bulk density was collected in 0 to 15 cm, 15 to

30 cm, 30 to 60 cm, 60 to 100 cm, and 100 to 200 cm depth layers, with a 4.7-cm diameter probe. In both 1993 and 2012, cores were collected from four random locations within each plot. Bulk densities were determined using mass of oven-dried soil (105 °C, 24 h) and total volume of the core averaged for each depth increment (Blake and Hartge 1986). Soils were void of rock fragments (Batjes, 2014). Bulk density depths from 1993 for 0-25 and 25-50 cm were adjusted to 2012 depths through the calculation of weighted averages using the adjacent 1993 and 2012 depth layers. Bulk density from 50-100 and 100-200 cm was assumed to not have changed between 1993 and 2012 and measured data from 2012 was used for both years.

1.3.6 Soil total C and N analysis

In 2015, subsamples were collected from well-homogenized archived soils from 1993 and 2012. All visible plant material was removed and samples were oven-dried at 60°C for 72 h and ground via ball mill for 12 h. Total C and N were determined by dry combustion (ECS 4010 Costech Elemental Analyzer). The pH of all plots and depths, was measured prior to C/N analysis. When pH measured above 7.4 (SI Table 1.2), suggesting the presence of inorganic carbon, samples were leached out using 2 M HCl until no effervescence was observed, as described in Carnell et al. (2018). Total soil C and N at each depth were calculated on both a percent and mass basis, converting concentrations to stocks, by the depth weighted sum (Eq. 1),

$$C_i = BD_i * d_i * [\%]_i \quad [1]$$

where C_i is the total mass of soil C (Mg ha^{-1}) for depth increment i , BD is bulk density of the soil (Mg m^{-3}), d indicates the length of depth increment i (m), and $[\%]$ indicates the percent C in the sample. Change in total soil C from 1993 to 2012 (ΔC concentration and ΔC stock) was calculated by subtracting C_{1993i} from C_{2012i} , for each depth increment i , for each plot. Positive

values indicate a gain in soil C, whereas negative values indicate loss. Total C to N ratios were calculated for each plot in 1993 and 2012 by dividing total soil C concentration by total N concentration for each depth increment i , for each plot. Change in soil C:N ratio (Δ C:N) was calculated by subtracting C:N_{1993*i*} from C:N_{2012*i*}.

1.3.7 Statistical analysis

Maize- and wheat-based systems (Table 1.1) were analyzed separately. Both linear and quadratic regression curves were fitted to cumulative C inputs across 19 years within each system using mixed effects models in the R statistical package *nlme* (Pinheiro et al. 2018) with cropping system and year as fixed effects and replicate as a random effect. Regressions were compared using AIC values to indicate the best model for each cropping system. The linear regression model provided the best fit in all cases and was used to compare the rate of cumulative C inputs across systems. Statistical significance was determined using $\alpha = 0.05$.

Change in soil bulk density was analyzed using mixed effects models in the R package *nlme*. Cropping system was treated as a fixed effect and replicate was treated as a random effect. Treatment by block interaction effects on change in soil C concentrations were examined to check for differences among soil types. Change in soil C concentration and stocks data met assumptions of normality and homoscedasticity. Statistically significant change in soil C concentrations and stocks was determined using t-tests, with the null hypothesis that soil C change = 0 from year 0 to year 19. T-tests were performed for each independent cropping system, within each depth layer, and 95% confidence intervals were computed for C change variables. Significant change was determined with a t-test and confirmed using examination of 95% confidence intervals, where the intervals did not overlap with zero. Differences in change in

soil C among cropping systems were determined using 95% confidence intervals, according to the visual inference methods described in Cumming (2009) and Brennan et al. (2017). Linear regression models were used to analyze change in soil C concentration and cumulative C inputs and evaluated using Pearson's correlation coefficients (r) and P values using the R package *Hmisc* (Harrell and Dupont, 2018). Change in soil C:N ratio (Δ C:N) from 1993 to 2012 among cropping systems was analyzed similarly. Linear regression curves were fitted to soil Δ C concentration vs. Δ C:N from 1993 to 2012 across all cropping systems in the R package *nlme*.

Results

1.4.1 Baseline soils

At the start of the experiment, average soil C content was 9.46 g kg^{-1} in the surface 0 to 15 cm and decreased in concentration moving down the soil profile (Table 1.2). Compared to the surface layer, soil C content was 34% and 60% lower at 60 to 100 and 100 to 200 cm, respectively (Table 1.2). Bulk density was similar between 0 and 60 cm and greater by 0.1 Mg m^{-3} on average in the 60 to 200 cm layers (Table 1.2). Clay content was similar among the depth layers in the top 0 to 60 cm and 10% greater in the 60 to 100 and 100 to 200 cm layers (Table 1.2). Clay content was not correlated with soil C in 1993 or 2012.

Table 1.2. Initial soil carbon concentrations, soil C to N ratios, bulk densities, and clay content among depth increments in 1993 at Russell Ranch, at the initiation of the Century Experiment. Different letters within a column represent statistically significant differences at $\alpha = 0.05$.

Depth Increment	Soil Total C Concentration	Soil C:N	Bulk Density	Clay Content	pH
cm	g kg ⁻¹	-	Mg m ⁻³	%	-
0-15	9.46 a	10.1 a	1.49 a	18.1 a	7.17 a
15-30	8.56 b	10.3 a	1.48 a	18.2 a	7.14 a
30-60	7.27 c	10.3 a	1.49 a	18.6 a	7.18 a
60-100	6.24 d	10.2 a	1.59 b	20.0 b	7.22 a
100-200	3.87 e	9.1 b	1.57 b	20.2 b	7.49 b

1.4.2 Bulk density

Bulk density in the maize-based systems declined from 1993 to 2012 ($P < 0.001$; SI Table 1.3). There was no interaction between cropping system and year ($P = 0.179$) or cropping system, year and depth ($P = 0.816$); however, there was an interaction between year and depth ($P < 0.001$). Bulk density declined on average by 0.31 Mg m⁻³ and 0.032 Mg m⁻³ in the 0-15 and 15-30 cm layers, respectively, and did not change in the 30-60, 60-100, and 100-200 cm layers (SI Table 1.3).

In the wheat-based systems, bulk density declined from 1993 to 2012 ($P < 0.001$; SI Table 1.3), with no interaction between cropping system and year ($P = 0.179$); year and depth ($P = 0.165$); or year, cropping system and depth ($P = 0.912$). There was an interaction between cropping system and depth ($P < 0.0001$). Soil bulk density on average declined by 0.24 Mg m⁻³ and 0.35 Mg m⁻³ in 0 to 15 and 15 to 30 cm, respectively but increased by 0.39 Mg m⁻³ in the 30 to 60 cm layer (SI Table 1.3).

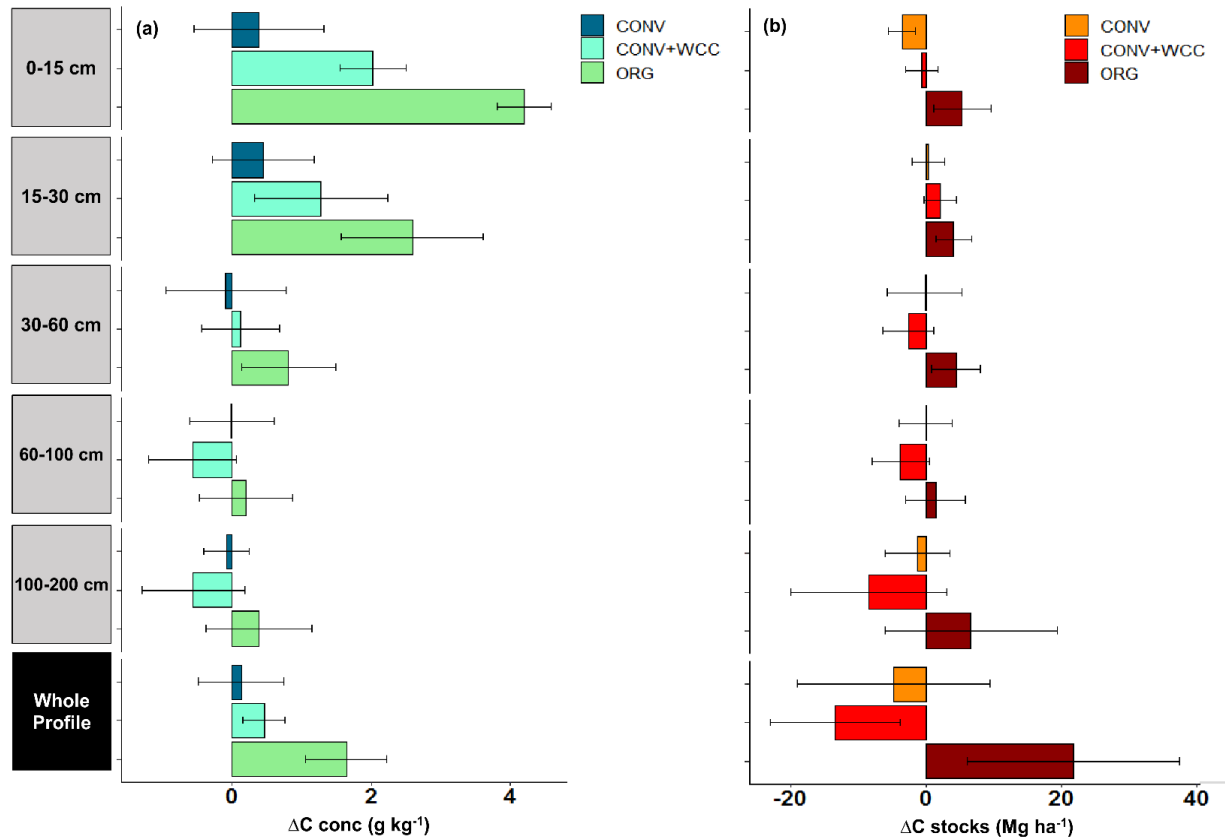


Figure 1.1. Soil C changes in maize-based systems from 1993 to 2012, expressed as a) change in C concentration (ΔC conc) and b) change in C stocks (ΔC stocks). Whole profile data indicate the averages of soil C concentrations, and the sums of soil C stocks, across all five depths. Error bars indicate 95% confidence intervals.

1.4.3 Aboveground cumulative C inputs

Of the maize-based systems, ORG had the greatest aboveground C input ($P < 0.001$), with an average C input of $7.27 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ (Table 1.3). CONV+WCC had lower C inputs than ORG but greater C inputs than CONV ($P = 0.024$), with an average of $5.05 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$. The C inputs from WCC and crop residues were similar between CONV+WCC and ORG systems ($P = 0.696$), but poultry manure compost in ORG added in an additional $40.4 \text{ Mg C ha}^{-1}$ over 19 years (Figure 1.3a). Of the cumulative aboveground C inputs in CONV+WCC, 30% was

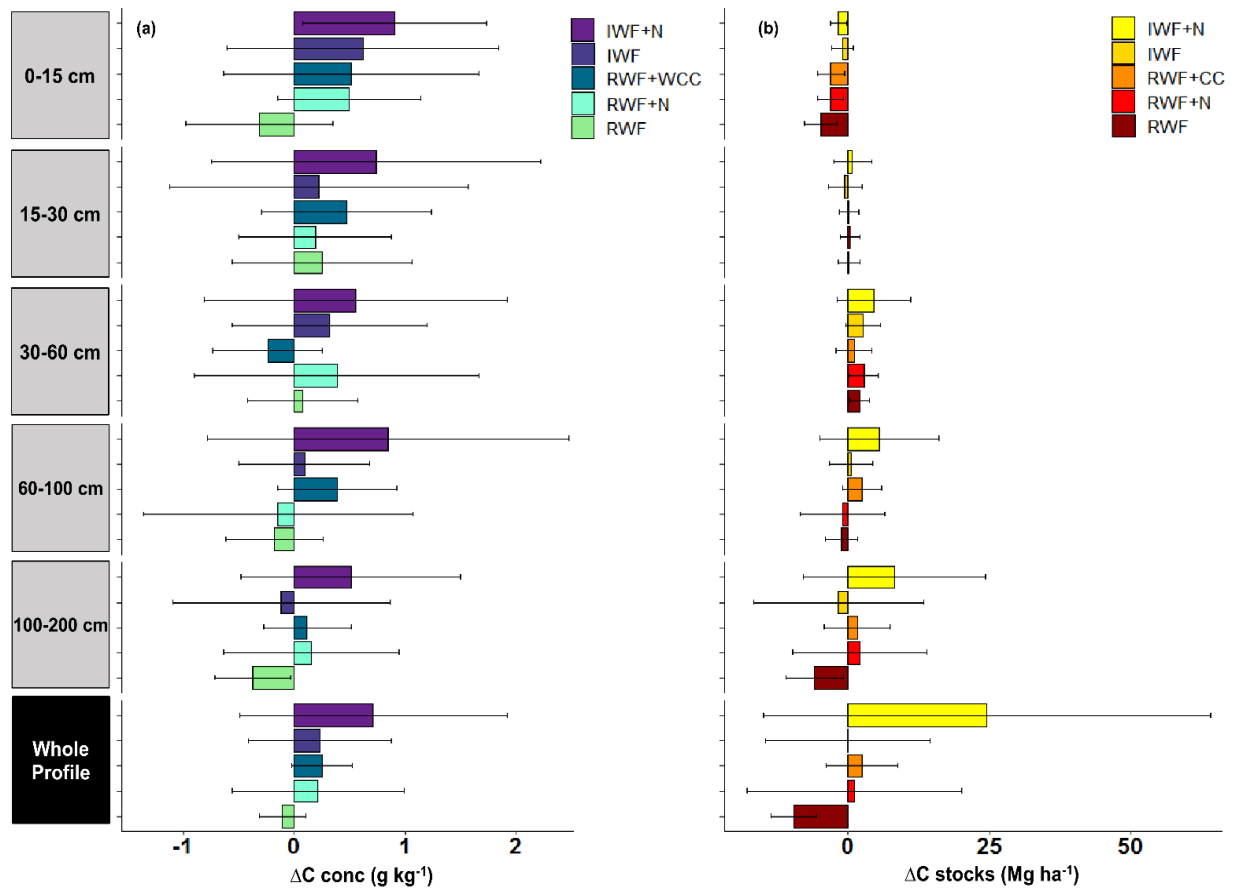


Figure 1.2. Soil C changes in wheat-based systems from 1993 to 2012, expressed as a) change in C concentration ($\Delta C \text{ conc}$) and b) change in C stocks ($\Delta C \text{ stocks}$). Whole profile data indicate the averages of soil C concentrations, and the sums of soil C stocks, across all five depths. Error bars indicate 95% confidence intervals.

from WCC, 24% from tomato residues and 46% from maize residues. Without a WCC, CONV received $0.75 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ less aboveground inputs (33% from tomato residues, 67% from maize stover) (SI Figure 1.1a). While tomato C inputs were similar in the conventional systems ($P = 0.115$), maize stover yields were generally greater following winter fallow than WCC ($P = 0.027$), leading to a cumulative stover-C input that was 7.1 Mg C ha^{-1} greater in CONV than CONV+WCC (SI Figure 1.1a).

In wheat-based systems, the greatest cumulative aboveground C inputs were in the RWF+WCC ($P < 0.001$), followed by IWF+N ($P = 0.047$), and were similar and lowest among the RWC, RWF+N, and IWC (Figure 1.3b). The RWF+WCC had the greatest cumulative C inputs (22.3 Mg C ha⁻¹ from straw, 15.5 Mg C ha⁻¹ from cover crops), despite higher crop yields and wheat straw C inputs (37.8 Mg C ha⁻¹; $P = 0.043$) in the IWF+N. Supplemental irrigation without N fertilizer did not increase cumulative C inputs (Figure 1.3b).

System	Intercept	Annual C Input Mg C ha ⁻¹ yr ⁻¹	2.5%	97.5%	R ²
<i>Maize-based</i>					
CONV	-1.9766	4.3042	4.1649	4.4435	0.9713
CONV+WCC	0.1590	5.0540	4.9084	5.1995	0.9771
ORG	4.4155	7.2736	7.1682	7.3790	0.9941
<i>Wheat-based</i>					
RWF	1.0250	1.1986	1.1357	1.2614	0.9610
RWF+N	-2.8546	1.4784	1.4117	1.5508	0.9659
RWF+WCC	-0.1663	1.9934	1.9106	2.0348	0.9535
IWF	0.9669	1.3377	1.2578	1.4177	0.9499
IWF+N	-2.2878	1.6219	1.5355	1.7084	0.9591

Table 1.3. Average annual aboveground C input linear model parameters, and 95% confidence intervals, derived from regressing cumulative aboveground C inputs vs. management year, for maize- and wheat-based systems, from 1993 to 2012. CONV = conventional; WCC = winter legume cover crop mix; ORG = organic; RWF = rainfed wheat–fallow; N = nitrogen fertilizer; IWF = irrigated wheat–fallow

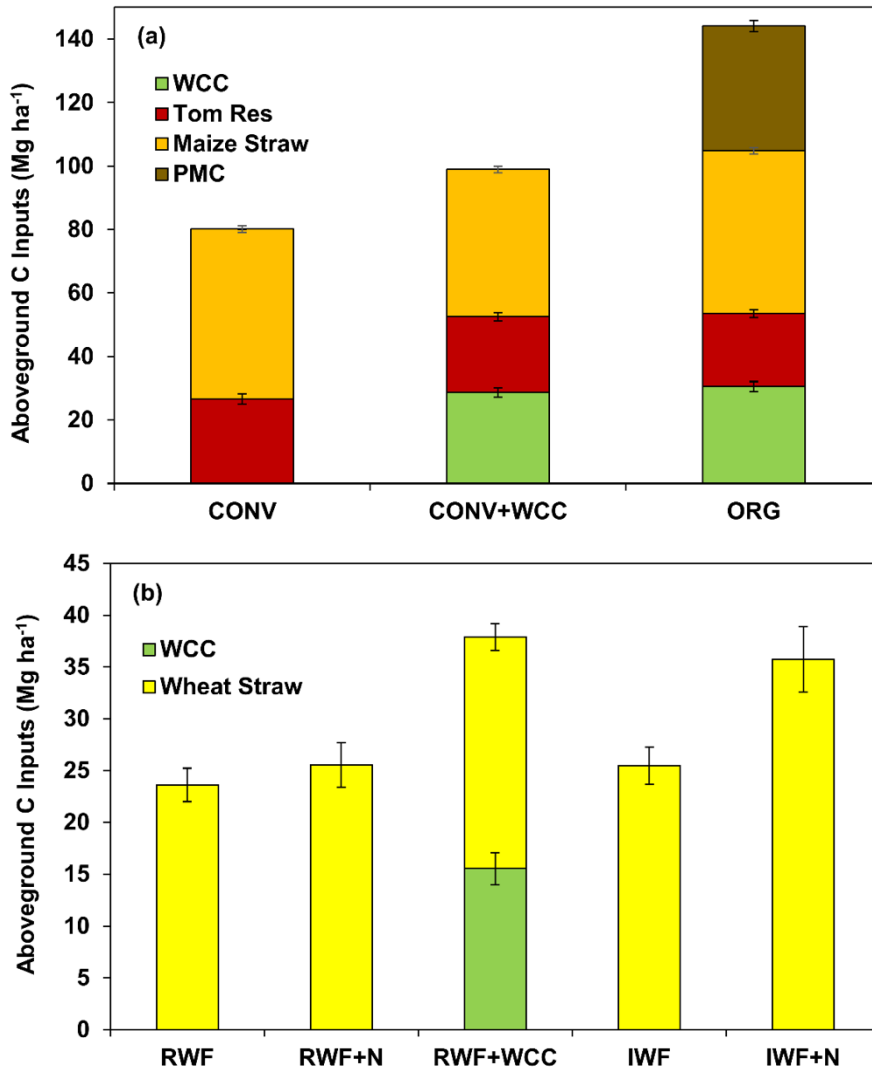


Figure 1.3. Source of cumulative aboveground C inputs incorporated into the soil in maize-based systems (a), and in wheat-based systems (b), with 95% confidence interval bars, over nineteen years of cropping system management.

1.4.4 Soil C changes: maize-based systems

There was no interaction between treatment and block effects ($P = 0.537$), indicating soil C changes among treatments were not significantly different among Yolo and Rincon soil types. The greatest increases in soil C were observed in ORG where soil C increased at all depths. Soil

C concentrations increased by 4.20 g kg^{-1} ($P < 0.001$) and 2.59 g kg^{-1} ($P = 0.006$; Figure 1.1a) at 0 to 15 and 15 to 30 cm, respectively. Converting to stocks, soil C increased by $5.31 \text{ Mg C ha}^{-1}$ ($0.266 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$; $P = 0.015$) in the 0 to 15 cm layer and by $2.59 \text{ Mg C ha}^{-1}$ in the 15 to 30 cm layer ($P = 0.010$; Figure 1.1b). From 30 to 60 cm, soil C increased by 0.81 g kg^{-1} ($4.41 \text{ Mg C ha}^{-1}$; $P = 0.026$).

In CONV+WCC, soil C concentrations increased by 2.03 g kg^{-1} in the top 15 cm ($P < 0.001$), and by 1.28 g kg^{-1} in the 15 to 30 cm layer ($P = 0.018$; Figure 1.1a). C stocks did not significantly change in 0 to 15 cm ($P = 0.556$) or 15 to 30 cm ($P = 0.082$; Figure 1.1b) layers. No changes in soil C concentration were observed in CONV in 0 to 15 cm ($P = 0.380$) or 15 to 30 cm ($P = 0.231$) layers. However, decreases in bulk density resulted in stock declines of $3.57 \text{ Mg C ha}^{-1}$ ($-0.179 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$; $P = 0.003$) in the 0 to 15 cm layer (Figure 1.1b,c).

No changes in soil C concentrations or stocks were observed in the 30 to 60 cm layer except in the ORG. In the 60 to 100 cm layer, there was no change in soil C concentration or stocks in CONV ($P = 0.975$) or ORG ($P = 0.454$; Figure 1.1 a,b) but CONV+WCC trended towards declines in concentration (-0.57 g kg^{-1}) and stocks ($-3.80 \text{ Mg C ha}^{-1}$). Negative changes were not significant ($P = 0.067$ and $P = 0.070$, respectively; Figure 1.1a,b).

In the 100 to 200 cm layer, significant changes in soil C concentrations were not observed in any system. Changes in concentrations and stocks trended negative in CONV+WCC ($P = 109$ and $P = 0116$, respectively; Figure 1.1a,b) and positive in ORG. No net change in soil C was observed throughout the 2 m profile in CONV ($P = 0.424$). In CONV+WCC, soil C concentration increased on average by 0.46 g kg^{-1} ($P = 0.012$) across the soil profile, while stocks decreased by $13.4 \text{ Mg C ha}^{-1}$ ($-0.670 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$; $P = 0.016$). In ORG, however,

average soil C concentrations and total C stocks increased across the entire soil profile by 1.64 g kg⁻¹ ($P < 0.001$) and 21.8 Mg C ha⁻¹ (1.09 Mg C ha⁻¹ yr⁻¹; $P = 0.016$), respectively (Figure 1.1 a,b).

There was no relationship between the change in soil C concentration from 1993 to 2012 and the cumulative maize and tomato aboveground residue C, nor WCC C inputs, at any depth (SI Figure 1.1). However, in ORG changes in soil C were positively correlated with cumulative poultry manure compost C inputs at 15 to 30 cm ($r = 0.88$; $P = 0.019$), 30 to 60 cm ($r = 0.84$; $P = 0.038$), and 100 to 200 cm ($r = 0.80$; $P = 0.047$) (SI Figure 1.1).

1.4.5 Soil C changes: wheat-based systems

In the surface 15 cm, soil C concentration did not change in RWC ($P = 0.275$), RWF+N ($P = 0.105$), RWF+WCC ($P = 0.304$), or IWF ($P = 0.251$), and increased in IWF+N (0.91 g kg⁻¹; $P = 0.038$; Figure 1.2a,b). Soil C stocks did not change in IWF ($P = 0.265$), and declined by 4.82 Mg C ha⁻¹ ($P = 0.007$) in RWF, by 3.09 Mg C ha⁻¹ ($P = 0.020$) in RWF+N, by 3.02 Mg C ha⁻¹ ($P = 0.021$) in RWF+WCC, and by 1.66 Mg C ha⁻¹ ($P = 0.032$; Figure 1.2a,b). In the 15 to 30 cm layer, no changes were observed in soil C concentrations or stocks in any of the systems (Figure 1.2a,b).

In the 30 to 60 and 60 to 100 cm layers, neither soil C concentrations nor stocks changed significantly in any of the wheat systems (Figure 1.2a,b). In the 100 to 200 cm layer, both soil C concentration (-0.037 g kg⁻¹; $P = 0.036$) and stocks (-5.85 Mg C ha⁻¹, $P = 0.032$) decreased significantly in the RWF and did not change significantly in the other four systems (Figure 1.2a,b). Soil C concentration and stocks trended towards increases in IWF+N (Figure 1.2a,b), but changes were not significant due to high variation among replicates (e.g., 95% confidence

interval for IWF+N stocks ranged from $-7.89 \text{ Mg C ha}^{-1}$ to $24.2 \text{ Mg C ha}^{-1}$).

Across the entire soil profile (0 to 200 cm), soil C concentration increased by 3.5% (0.25 g kg^{-1} ; $P = 0.048$) in RWF+WCC and did not change in the other systems (Figure 1.2a). Soil C stocks declined by $9.52 \text{ Mg C ha}^{-1}$ ($-0.476 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$) in RWF ($P = 0.002$; Figure 1.2b). Soil C stocks across the entire profile of IWF+N increased by $17.5 \text{ Mg C ha}^{-1}$ on average, but the change was not statistically significant ($P = 0.680$) due to high variation among plots, with soil C stock changes ranging from -4.74 to $59.0 \text{ Mg C ha}^{-1}$. Soil C stocks did not change in RWF+N, IWF, and RWF+WCC (Figure 1.2). There was no relationship between soil C concentration change and cumulative wheat C inputs ($P = 0.453$), nor with cumulative WCC C inputs ($P = 0.899$), throughout the soil profile.

1.4.6 Soil C:N

In 1993, soil C:N ratios ranged from 9.0 to 11.3 in the top 100 cm, and 6.3 to 11.5 in the 100 to 200 cm layer (data not shown). These ratios generally increased across plots after 19 years of management. Between 1993 and 2012, change in soil C:N ratio varied substantially among maize-based systems. In the CONV, soil C:N increased in 0 to 15, 15 to 30 cm, and 100 to 200 cm layers. In contrast, C:N declined in the 30 to 60 cm layer and showed no change in the 60 to 100 cm layer. In ORG, soil C:N showed trends that were opposite of those observed in CONV: C:N in ORG decreased in the surface layers but increased in the lower two depths (Figure 1.4a). In CONV+WCC, C:N only decreased in the 30 to 60 and 60 to 100 cm depths, where N increased 2 to 2.5 times relative to C (data not shown). Averaged across the entire 200 cm profile, soil C:N increased in CONV and ORG, and decreased in the CONV + WCC (Figure 1.4a).

In the rainfed wheat-based systems, addition of synthetic N had no impact on soil C:N at most depths, or across the entire soil profile. The only exception was the 100 to 200 cm layer, where soil C increased relative to N in RWF, while N increased relative to C in RWF+N (Figure 1.4b). Irrigating wheat did not substantially alter soil C:N compared to the RWF; however, N fertilizer inputs combined with supplemental irrigation generally increased Δ C:N across the 0 to 200 cm soil profile (Figure 1.4b). Inclusion of WCC increased soil C:N in the top (0 to 15 cm) layer but decreased C:N in 30 to 60 and 100 to 200 cm layers. RWF+WCC was the only wheat-based cropping system that exhibited enrichment of soil N relative to soil C across the soil profile (Figure 1.4b). No consistent relationships were observed between changes in soil C and changes in soil C:N ratios (SI Figure 1.2).

1.5 Discussion

Our study, one of a few long-term efforts to track soil C changes in surface and subsoil layers in an agricultural system, highlights the importance of including deep soil measurements in soil C accounting. Of the 9 cropping systems observed in our 19-year study, only one system (ORG) showed increases in soil C stocks throughout the entire 0 to 200 cm soil profile. While three other systems displayed an increase in soil C concentration overall, these gains did not translate into changes in stocks either due to declines in bulk density offsetting gains in soil C concentration and/or declines in some layers offsetting gains in others. Bulk density likely declined in the surface layers of most systems due to the cumulative build-up of organic matter. Others have observed an inverse relationship between SOM and bulk density, as well as declines in bulk density in long-term cropping experiments (Périé and Ouimet 2008; Poulton et al. (2018).

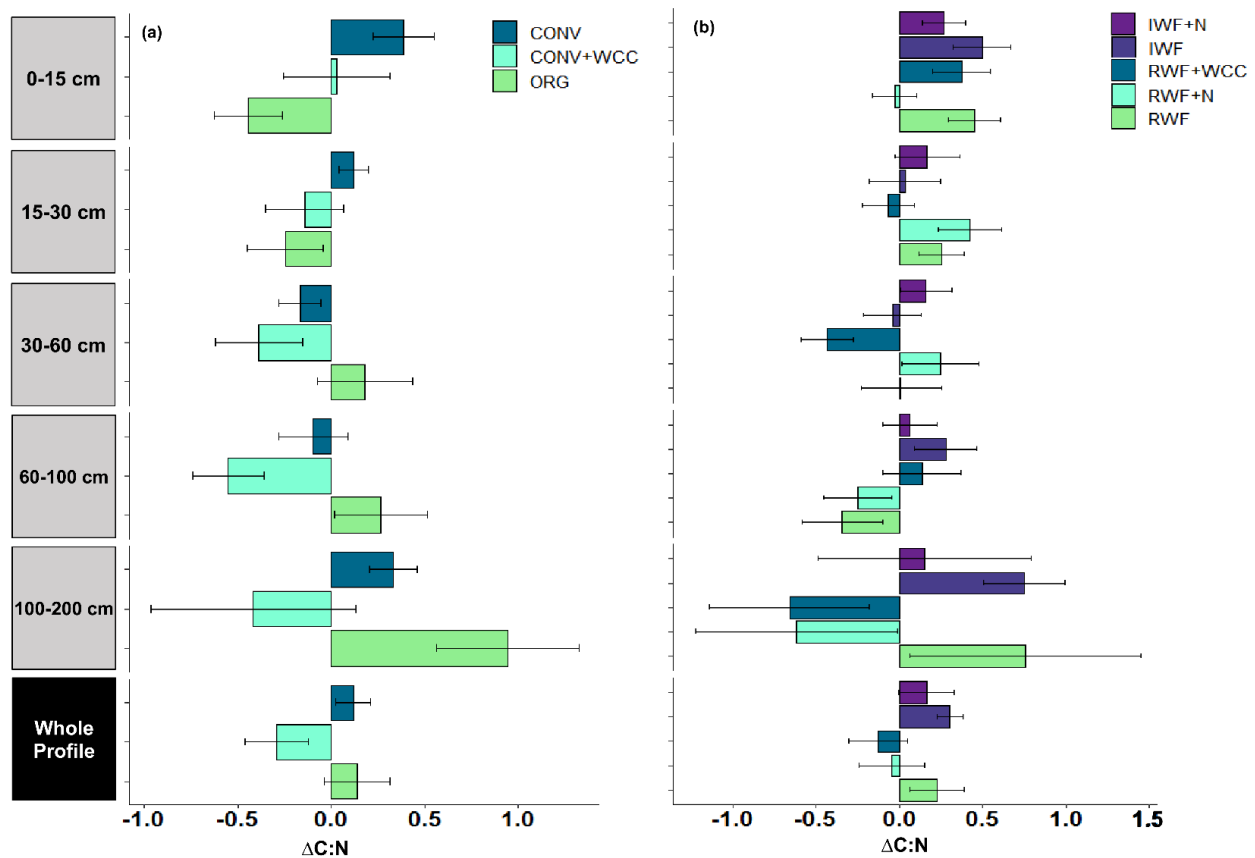


Figure 1.4. Change in soil C:N ratio from 1993 to 2012, and 95% confidence intervals, in maize-based (a) and wheat-based (b) cropping systems.

In maize-based systems, annual inputs of 9 t ha^{-1} of composted poultry manure contributed $2.22 \text{ Mg ha}^{-1} \text{ yr}^{-1}$ more C to ORG than CONV+WCC systems, and were associated with a 3.5 times greater soil concentration in ORG. The $21.8 \text{ Mg C ha}^{-1}$ gain in soil C stocks in ORG translated to an annual increase of 6.6 % in soil C per year, exceeding the benchmark of 4 % targeted by the 4 per 1000 initiative (French Ministry of Agriculture and Food 2015). Relying primarily on poultry manure compost as an input, either statewide or globally, may be limited by supply as well as economic and environmental costs of transportation. Potential C feedstocks

should be evaluated individually for their efficacy in C sequestration along with a life cycle assessment to estimate their total footprint. However, replacing synthetic fertilizers with compost can reduce greenhouse gas emissions. Alluvione et al. (2010) observed a 49% reduction in CO₂ emissions from soils amended with compost rather than synthetic urea. A global meta-analysis of Mediterranean croplands found emissions of N₂O (300x radiative forcing of CO₂) to be lower in organic systems fertilized with compost than in conventional systems with synthetic fertilizers (Aguilera et al. 2013). In another global meta-analysis of greenhouse gas emissions from organic and synthetic soil amendments, Charles et al. (2017) calculated an N₂O emissions factor of 0.27% of total N applied in compost-fertilized systems, compared to 1.34% of total N applied in synthetic fertilized systems. Our results demonstrated that substantial increases in soil C are achievable even in semiarid climates and compost-C may be effective in increasing soil C and decreasing greenhouse gas emissions on decadal time scales. More research is needed to correlate compost characteristics (e.g., C to nutrient ratios) with soil C sequestration potentials.

Soil C is only rarely measured at depths below 30 cm (Poeplau and Don 2005), despite the likelihood that carbon there is more protected from biotic and abiotic losses (Jobbágy and Jackson 2000; Hicks Pries et al. 2018). Not considering the potential for C storage throughout the soil profile may overlook considerable opportunities for sequestration. Had our study only measured carbon in the top 30 cm, we would have missed the gains of 12.39 Mg C ha⁻¹ observed in deeper layers (30 to 200 cm) of the ORG system, grossly underestimating soil C sequestration by 57%.

In contrast, focusing on only the surface layer of soil can greatly overestimate C gains. In CONV+WCC, the increases in soil C in the top 30 cm were offset by substantial losses that occurred below 30 cm. Follow-up sampling of the CONV+WCC system has indicated loss of C

at soil depths near the bottom of and below the rooting zone of the cover crops (~60 to 80 cm; N. Tautges, unpublished data). This phenomenon may be due to priming of SOM (Dignac et al. 2017) by resource-limited deep microbial communities and/or low soil moisture conditions decreasing occlusion and adsorption mechanisms that might have helped retain soil C (Jardine et al. 1989; Blankenship and Schimel 2018; Jones et al. 2018). More research is needed to elucidate C dynamics in this understudied zone.

The comparatively lower rates of soil C sequestration we observed with cover crops may have been due to lower cover crop-C inputs in our study ($1.43 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$) relative to the average input of $1.87 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$ estimated by Poeplau and Don (2015). The WCC mix had a higher proportion of legumes in the first eight years of our study, which may have decreased biomass production and C inputs relative to global estimates from higher biomass cover crops (i.e., grasses). Despite this, WCC did increase soil C in the top 30 cm layer of our highly tilled system, as many other studies report when sampling the top layer of soil alone.

Microbial utilization of cover crop-C is an important pathway for increasing SOC (Kallenbach et al. 2015). We observed this in the 0 to 30 cm layer where both soil C and microbial biomass (K. Scow, unpublished data) increased in cover cropped systems compared to CONV. Increased available N provided by legumes in the WCC may have increased the carbon use efficiency of WCC-C inputs, with greater incorporation of WCC-C into microbial bodies and ultimately greater soil organic C (Lange et al. 2015). Most research on microbial C cycling has focused on surface soil (Kallenbach and Grandy 2011; Poeplau and Don 2015, Tiemann et al. 2015). Relationships between microbial biomass and soil C are less clear in the subsurface where C inputs and microbial biomass are much lower and impacts of physical processes, such as occlusion and sorption, may predominate.

Nutrient stoichiometry is an important consideration for SOM dynamics, as it is key for microbial growth and turnover in soil (Kirkby et al., 2013; Kirkby et al., 2016). Ensuring that microbial nutrient requirements are met-- adding nitrogen, phosphorus and sulfur at time of residue C incorporation--increased soil C throughout a 1.6 m soil profile (Frossard et al. 2016; Kirkby et al. 2016). The increases of soil C we observed may have been similarly facilitated by the relatively large amounts of N, P, S (>25 kg/t) added alongside the C inputs of the poultry manure compost. WCC alone has been observed to immobilize soil P and K levels compared to winter fallow at our site (N. Tautges, unpublished data).

Significant mass loss of soil C was observed in only one system (unfertilized RWF), likely due to water and nutrient (i.e. N) limitations, resulting in low productivity and biomass-C inputs. Wheat systems not receiving N fertilizer (IWF, RWF and RWF+WCC) produced the least amount of wheat straw and those not receiving any N fertility (IWF and RWF) showed no change or lost C. C input and storage is often higher in fertilized, irrigated systems (Haynes and Naidu 1998; Lal 2002).

Given the large variance in changes in soil C concentration and stock in IWF+N, particularly from 60 to 200 cm, it was not possible to demonstrate significant change in soil C stocks. The high observed variance may be the result of subsoil C occurring in rhizosphere “hot spots.” While some samples may have represented bulk soil under no root influence, others may have been taken from the rhizosphere, impacted by wheat roots which can reach to 2 m deep. In these layers, soil C trended upwards in IWF+N and decreased in RWF. There was no evidence of subsoil C gains in IWF alone (>1 m), suggesting that addition of N fertilizer, and not irrigation, was driving C gains.

Surprisingly, in the IWF+N fertilizer plots with higher levels of soil C ($>50 \text{ Mg C ha}^{-1}$), increases in subsoil C were greater than in maize-based systems, supporting Jobbágy and Jackson's (2000) observation that grasses tend to distribute soil C deeper in the soil profile than does maize. As carbon appears to be better protected from degradation in the subsoil than surface soil, maximizing wheat productivity (e.g. with adequate fertilizer) is a potential strategy for increasing C. Intensifying cropping cycles and using mineral fertilizer inputs, when combined with high rates of C inputs to soils, have substantially increased soil organic C in other systems (Sanderman 2017).

Considering the entire 2 m soil profile, WCC incorporated without additional nutrient application may have decreased soil C at depths $>60 \text{ cm}$, resulting in net declines in C across the profile. By comparison, application of $700 \text{ to } 800 \text{ kg C ha}^{-1} \text{ yr}^{-1}$ via compost in the ORG drove soil C gains of 12% over 19 years in maize–tomato systems. C loss in the WCC system was unexpected and research is ongoing on-site to understand mechanisms involved, as well as interactions of cover crops and compost in stabilizing soil C.

To conclude, if only the surface soil (0 to 30 cm) had been analyzed—the typical practice in monitoring soil C—we would have falsely concluded that adding WCC to conventionally managed row crops increases soil C sequestration. Similarly, measuring soil C to a 2 m deep indicated the organic system had substantially greater capacity to sequester C than surface sampling would reveal. Our results provide concrete examples of the importance, particularly for carbon markets, of full accounting of soil C throughout the entire soil profile when recommending management practices to optimize soil C sequestration.

1.6 Acknowledgements

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1.8 Supplemental Information

SI Table 1.1. Precipitation at Russell Ranch from 1993 through 2012 in the winter (October–March) and summer (April–September) cropping seasons.

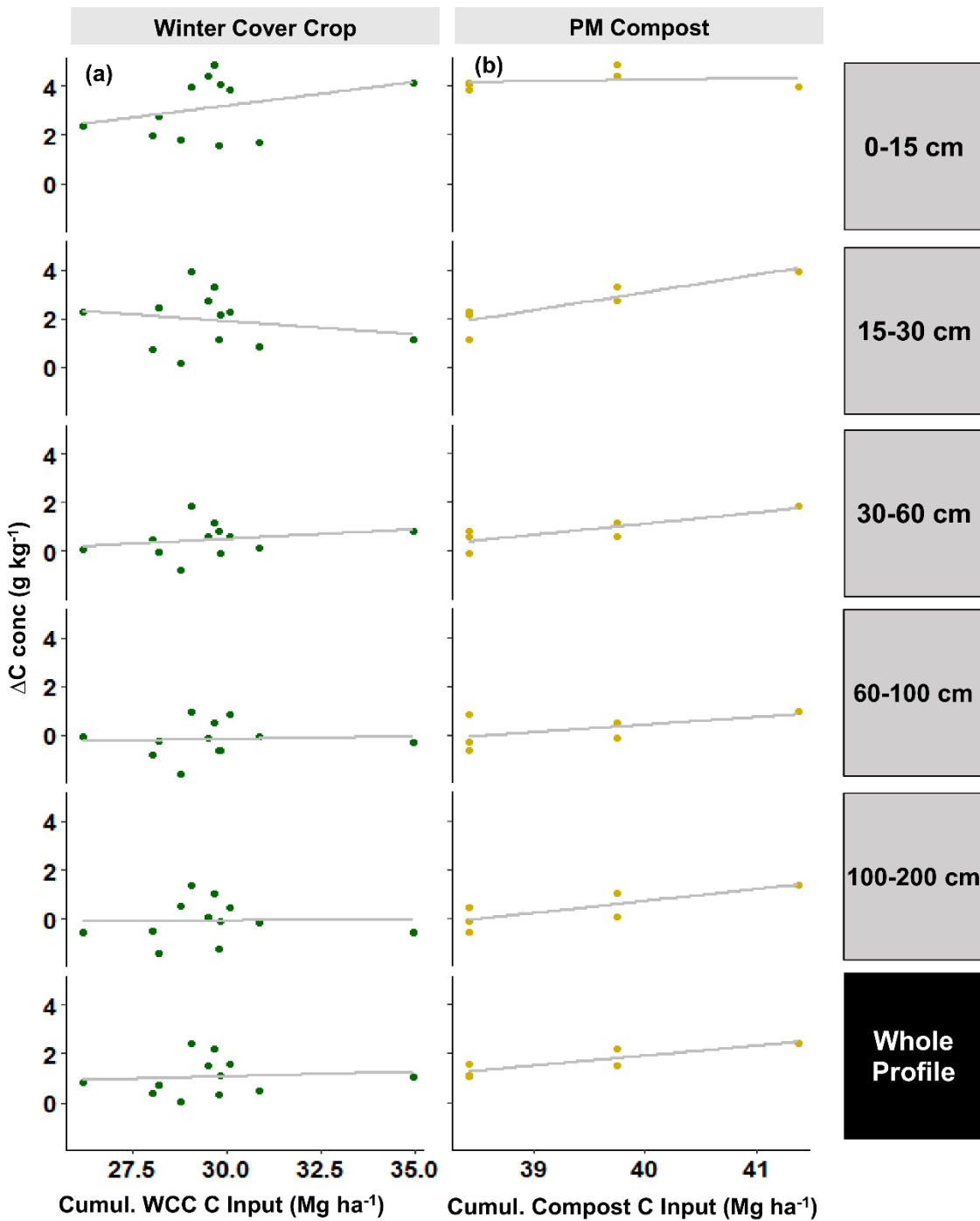
	Precipitation	
	Winter	Summer
	----- mm -----	
1993 - 1994	101.6	50.3
1994 – 1995	318.3	71.3
1995 – 1996	615.7	104.1
1996 – 1997	528.1	22.4
1997 – 1998	384.3	106.2
1998 – 1999	565.7	32.3
1999 – 2000	231.9	46.5
2000 – 2001	417.6	35.6
2001 – 2002	492.3	16.3
2002 – 2003	389.4	99.3
2003 – 2004	303.5	5.08
2004 – 2005	448.1	49.8
2005 – 2006	500.1	93.7
2006 – 2007	305.8	49.8
2007 – 2008	210.8	0.0
2008 – 2009	336.0	34.5
2009 – 2010	292.6	88.1
2010 – 2011	462.0	64.5
2011 – 2012	324.1	51.1

SI Table 1.2. Soil pH at year 19 (2012), at five depth layers in maize- and wheat-based systems. Bolded values indicate samples with pH values above the 7.4 threshold, which were treated with HCl prior to C/N analysis.

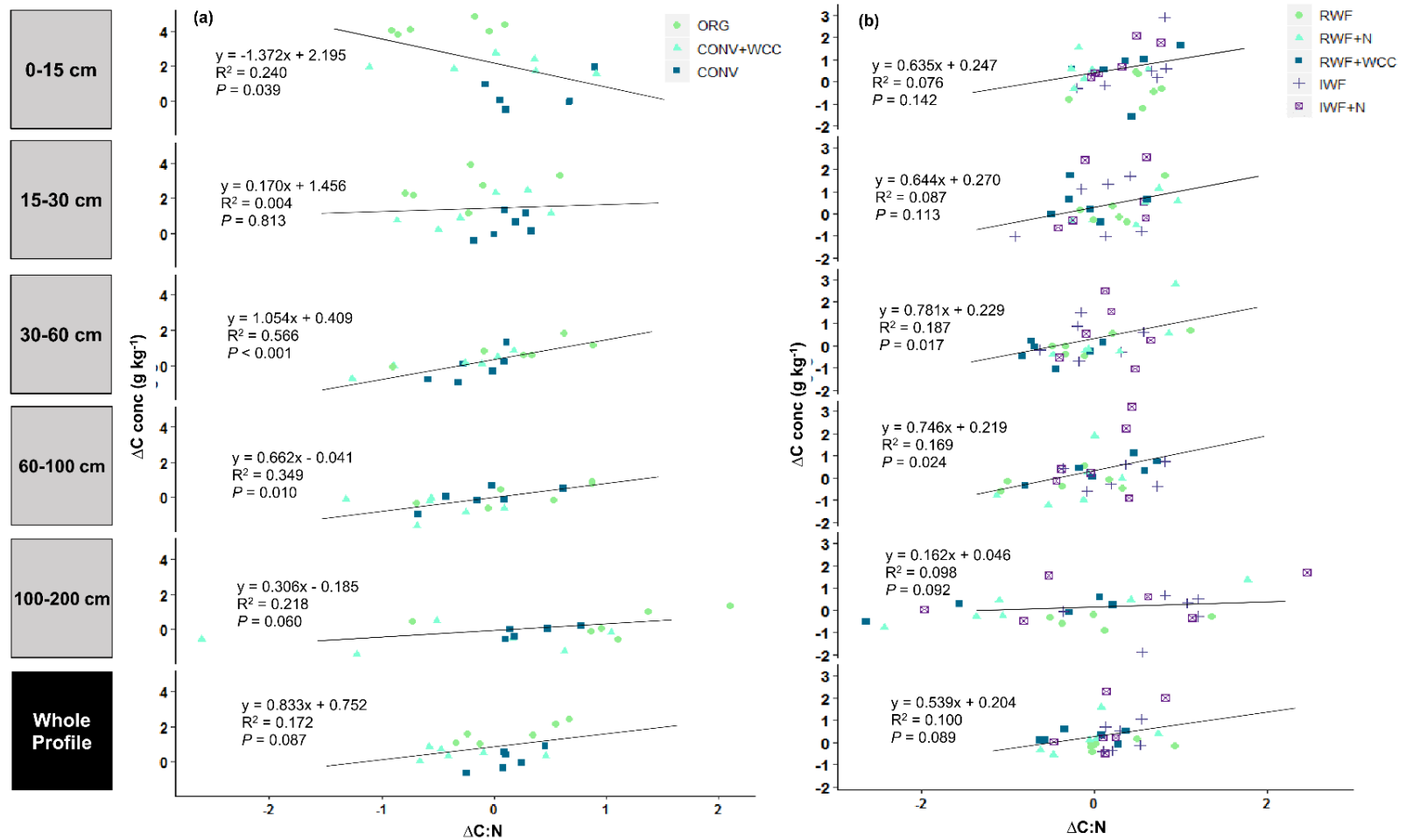
System	Depth	pH	se
<i>Maize-based</i>			
CONV	0-15 cm	7.18	0.06
	15-30 cm	7.28	0.04
	30-60 cm	7.44	0.05
	60-100 cm	7.55	0.05
	100-200 cm	7.69	0.04
CONV+WCC	0-15 cm	7.14	0.05
	15-30 cm	7.28	0.03
	30-60 cm	7.38	0.04
	60-100 cm	7.49	0.03
	100-200 cm	7.66	0.06
ORG	0-15 cm	7.19	0.03
	15-30 cm	7.25	0.05
	30-60 cm	7.40	0.02
	60-100 cm	7.50	0.04
	100-200 cm	7.59	0.05
<i>Wheat-based</i>			
RWF	0-15 cm	7.08	0.07
	15-30 cm	7.19	0.05
	30-60 cm	7.32	0.06
	60-100 cm	7.45	0.06
	100-200 cm	7.65	0.04
RWF+N	0-15 cm	6.93	0.07
	15-30 cm	7.04	0.03
	30-60 cm	7.24	0.06
	60-100 cm	7.35	0.05
	100-200 cm	7.58	0.07
RWF+WCC	0-15 cm	6.92	0.03
	15-30 cm	7.03	0.02
	30-60 cm	7.25	0.06
	60-100 cm	7.42	0.07
	100-200 cm	7.60	0.10
IWF	0-15 cm	7.11	0.04
	15-30 cm	7.26	0.04
	30-60 cm	7.36	0.04
	60-100 cm	7.47	0.04
	100-200 cm	7.63	0.05
IWF+N	0-15 cm	6.93	0.10
	15-30 cm	7.21	0.04
	30-60 cm	7.42	0.06
	60-100 cm	7.49	0.04
	100-200 cm	7.73	0.06

SI Table 1.3. Soil bulk density at baseline (1993) and at year 19 (2012), and change in soil bulk density (Δ BD; soil BD₂₀₁₂ – soil BD₁₉₉₃), at five depth layers in maize- and wheat-based systems.

System	Depth	Soil Bulk Density				Δ BD
		1993	se	2012	se	
<i>Maize-based</i>						
		----- g kg ⁻¹ -----				
CONV	0-15 cm	1.51	0.01	1.22	0.05	-0.29
	15-30 cm	1.49	0.01	1.32	0.09	-0.17
	30-60 cm	1.51	0.02	1.52	0.05	-0.01
	60-100 cm	1.58	0.03	1.58	0.04	0.00
	100-200 cm	1.52	0.03	1.52	0.04	0.00
CONV+WCC	0-15 cm	1.51	0.03	1.21	0.03	-0.30
	15-30 cm	1.50	0.03	1.24	0.06	-0.26
	30-60 cm	1.53	0.04	1.39	0.03	-0.14
	60-100 cm	1.62	0.03	1.62	0.05	0.00
	100-200 cm	1.57	0.03	1.58	0.04	0.01
ORG	0-15 cm	1.47	0.03	1.28	0.07	-0.19
	15-30 cm	1.46	0.02	1.27	0.03	-0.19
	30-60 cm	1.48	0.05	1.51	0.05	0.03
	60-100 cm	1.64	0.02	1.63	0.03	-0.01
	100-200 cm	1.61	0.03	1.62	0.04	0.01
<i>Wheat-based</i>						
RWF	0-15 cm	1.51	0.03	1.21	0.03	-0.30
	15-30 cm	1.49	0.02	1.44	0.03	-0.05
	30-60 cm	1.49	0.02	1.57	0.02	0.08
	60-100 cm	1.53	0.06	1.55	0.06	0.02
	100-200 cm	1.56	0.03	1.57	0.02	0.01
RWF+N	0-15 cm	1.48	0.02	1.21	0.05	-0.27
	15-30 cm	1.47	0.01	1.50	0.05	0.03
	30-60 cm	1.51	0.04	1.56	0.04	0.05
	60-100 cm	1.53	0.04	1.54	0.05	0.01
	100-200 cm	1.58	0.04	1.60	0.05	0.02
RWF+WCC	0-15 cm	1.45	0.03	1.18	0.03	-0.27
	15-30 cm	1.44	0.02	1.34	0.05	-0.10
	30-60 cm	1.45	0.04	1.54	0.03	0.09
	60-100 cm	1.61	0.05	1.61	0.05	0.00
	100-200 cm	1.53	0.05	1.54	0.05	0.01
IWF	0-15 cm	1.47	0.01	1.32	0.05	-0.15
	15-30 cm	1.47	0.01	1.32	0.06	-0.15
	30-60 cm	1.51	0.02	1.57	0.05	0.06
	60-100 cm	1.61	0.05	1.63	0.06	0.02
	100-200 cm	1.57	0.03	1.57	0.03	0.00
IWF+N	0-15 cm	1.48	0.03	1.24	0.07	-0.24
	15-30 cm	1.46	0.02	1.38	0.07	-0.08
	30-60 cm	1.47	0.04	1.58	0.04	0.11
	60-100 cm	1.51	0.06	1.53	0.06	0.02
	100-200 cm	1.56	0.04	1.57	0.05	0.01



SI Figure 1.1. Soil C change versus cumulative C inputs from winter cover crops (WCC) in conventional maize–tomato with cover crops and organic maize–tomato systems (a) and soil C change versus cumulative C inputs from poultry manure compost in the organic maize–tomato system (b), from 1993 to 2012



SI Figure 1.2. Change in soil C concentration vs. change in soil C:N ratio ($\Delta C:N$) from 1993 to 2012 in maize-based (a) and wheat-based (b) rotations at five depth layers, and average change across the whole profile, depicted with fitted linear regression models and coefficients.

Chapter 2: Hedgerows on margins of row crop fields increase soil carbon across the soil profile

2.1 Abstract

Emerging markets and policies to incentivize storage of soil carbon (C) as a climate mitigation strategy necessitate an improved understanding of potential gains and losses across a range of edaphic factors. To properly account for total stocks and determine if a given intervention is an overall source or sink, samples must be collected to a sufficient depth. Here, the difference in soil C concentrations and stocks (0-100 cm) was assessed between long-term hedgerow plantings (10+years) and adjacent cultivated fields, at 21 paired sites representing four soil types (Mollic Xerofluvent, Typic Haploxerept, Typic Haploxerert, and Typic Palexeralf) common in Yolo County, CA. Total soil C was significantly higher at all depths (0-100 cm) under hedgerows relative to cultivated fields. While the difference in total C between hedgerows and cultivated fields did not vary by soil type at any depth, soil C was significantly different under both hedgerows and cultivated fields at all depths from 20-100 cm in one of the four soil types. Considering a 1-m soil profile inventory, the average difference in soil C between hedgerows and cultivated fields was 3.85 kg m^{-2} , indicating that farm hedgerows can contribute in part to greenhouse gas reduction strategies, while providing a suite of field and landscape scale co-benefits.

2.2 Introduction

Conversion of native ecosystems to agriculture and continued intensification has resulted in substantial losses of carbon (C) from the top 100 cm of soils worldwide (Amundson et al. 2001; Sanderman et al. 2017). Net global greenhouse gas emissions continue to rise by approximately 4.9 Pg C/yr (Amelung et al. 2020) and recent projections indicate negative emissions of 150 Pg C are needed to avoid a concomitant rise in global temperatures (UNFCCC, 2015; Hansen et al. 2017). There is a growing interest in soil C storage to simultaneously mitigate and adapt to climate change (Lal et al. 2016; Paustian et al. 2016; Bossio et al. 2020). Although agriculture only accounts for 9-14 % of the global GHG budget (Mbow et al. 2020; [EPA](#) 2019), it is one of the most vulnerable sectors to climate change and strategies to increase soil organic carbon (OC) have been documented to improve soil hydrologic function (Franzlubbers et al. 2002; Libohova et al. 2018) and increase resilience in the face of extreme hydrometeorological events (Williams et al. 2016; Bowles et al. 2020; Kane et al. 2021; Renwick et al. 2021).

Although soil C saturation (West & Six 2007; Stewart et al. 2009; Dignac et al. 2017), stoichiometric constraints (Kirkby et al. 2013; van Groenigen et al. 2017), and socioeconomic barriers (Poulton et al. 2018; Amundson & Biardeau, 2018; Rumpel et al. 2020) may limit actual sequestration, it has been estimated that globally, soils can sequester 0.7-1.85 Pg C yr⁻¹ for up to 20 years (Smith et al. 2016; Amelung et al. 2020). There are an increasing number of federal, state, and market-based initiatives emerging to incentivize soil C storage on natural and working lands. It is widely documented that conservation practices, in particular cover cropping, reduction of fallow, residue management, and conservation tillage have the capacity to mitigate emissions by storing soil C (Smith et al. 2016; Paustian et al. 2020, Bossio et al. 2020).

Reforestation or the integration of perennial vegetation in the form of hedgerows, windbreaks, and/or riparian corridors provides an additional form of C in the form of woody biomass (e.g., trees, shrubs and vines) (Schoeneberger 2009, 2012; Wang et al. 2013; Thiel et al 2015). They are typically planted on marginal lands and bare, field edges and waterways; infringing little on production agriculture (Schoeneberger et al. 2009; Brodt et al. 2020), although this may not be the case as prices go up in some low value commodities. They also provide valuable ecosystem services at the field and landscape scale including buffering of wind (Bentrup 2008, Marshall and Moonen 2002); increased infiltration and interception of nutrients (Ghazavi et al. 2008; Long et al 2010; Smukler et al.2012; Thomas and Abbot 2018); increased pollination and pest control (Morandin et al. 2011, 2014, 2016); and the promotion of habitat and biodiversity in increasingly fragmented landscapes (del Barrio et al. 2006; Smukler et al. 2010; Ponisio et al. 2015; Long et al. 2017).

There is a body of literature showing the potential for hedgerows to sequester C (Falloon et al. 2004; Follain et al. 2007; Schoeneberger et al. 2009; Smukler et al. 2010; Thiel et al. 2015; Drexler et al. 2020) through litter deposition, extensive root systems, root exudation, reduction of disturbance, and erosion control (Walter et al. 2003; Montagnini and Nair 2004; Lenka et al. 2012; Pardon et al 2017; Cardineal et al. 2018; Zheng et al. 2020). Quantitative investigations of soil C stocks in deeper soil layers under hedgerows and other potentially deep-rooted, woody plants, however, remain scarce (Aguilera et al. 2013; Upson and Burgess 2013; Cardinael et al. 2015). Recent meta-analyses found average sampling depths of 28.4 cm (83 sites) when comparing hedgerows to adjacent cropland (Drexler et al. 2021); 25.7 cm (174 datasets) in Mediterranean cropping systems at large (Aguilera et al. 2013 This neglects mechanisms that contribute to SOC at depth (Jobbagy & Jackson, 2000; Rumpel & Kogel-Knabner, 2011; Batjes,

2014; Torres-Sallan et al. 2017), and can lead to misestimation of soil C change (West & Post 2004; Baker et al. 2007; Poeplau & Don 2015).

While concentrations of C (g kg^{-1}) are commonly higher near the surface, soil layers below 30 cm may hold greater proportions of total stocks (Jobbagy & Jackson 2000; Harrison et al., 2011; Zabowski et al. 2011). Global estimates range from 755-863 Pg C in the upper 30 cm, but increase to 1,408-1,824 Pg when considering the upper 100 cm (Jobbagy & Jackson 2000; Batjes 2016; Sanderman et al., 2017). The subsoil may provide greater potential for soil OC stabilization, as it often contains greater reactive surface areas (von Lutzow 2006), is commonly undersaturated with respect to OC (Scharlemann et al. 2014), and has been found, through radiocarbon dating, to cycle over greater mean residence times (Kaiser et al. 2002; Rumpel et al. 2004; Chabbi et al. 2009). There is increasing evidence, however, that subsoil OC is still thermodynamically labile (Schmidt et al. 2011; Lehmann et al. 201; Kogel-Knabner & Amelung 2021) and susceptible to management (Strahm et al. 2009; Follett et al. 2009; Devine et al., 2011; Harrison et al., 2011; Stewart et al. 2017; Dal Ferro et al. 2020). Management-induced reductions in subsoil OC stocks have been shown to offset gains in the surface (Don et al., 2009; Syswerda et al. 2011; Mobley et al. 2015; Veneestra et al., 2015; Tautges et al. 2019); threatening to undermine climate mitigation efforts (James et al. 2014). While Kyoto protocol and the scientific literature now recommend sampling depths of 1-2 m, or the maximum rooting depth (Harrison 2011; Suddick et al. 2013; Olson et al 2014); many incentive and accounting programs (CDFA, ESMC, IPCC, Soil Carbon Index, Indigo Ag, US Forest Service) still only require sampling to 20-30 cm (O'Neill et al. 2005; Aalde et al. 2006; CDFFA 2018; Jackson et al. 2021).

Understanding the potential of management/land use change to affect soil C across a range of edaphoclimatic conditions is needed to determine appropriate interventions and thresholds for policy and incentive programs (Morari et al. 2019; Devine & O’Geen 2021; Amelung et al. 2020). Much of soil C research, however, has been focused on Midwestern agroecosystems and there is a paucity of data in the semi-arid Western US (DeGryze et al., 2009; Suddick et al., 2010; Aguilera et al, 2013). Climate is a major driver of soil C dynamics (von Lutzow et al., 2009, Carvhalais et al., 2014) and may present challenges to storing C in semi-arid regions (Zhou et al. 2009; Doetterl et al 2015) where models project increases in temperature and severity/ frequency of drought (Hayhoe et al. 2004; Cayan et al. 2006; Romanya et al., 2007; Munoz-Rojas et al., 2012). Geochemical drivers (texture, mineralogy, pH), however, may dominate below 20 cm (Jobbagy & Jackson 2000; Hobley et al. 2015; Mattieu et al. 2015).

Here, we address California agroecosystems, which span a diversity of climate zones, soil types, and cropping systems characterized by intensive, irrigated cropping systems with frequent bare fallow, low diversification, and relatively heavy tillage use (Culman et al. 2010; Suddick et al. 2010), presenting both challenges and opportunities for C storage. Previous studies have shown C sequestration benefits of reforestation with woody species in Yolo County landscapes (Young-Mathews et al. 2010; Smukler et al. 2010). The benefits of hedgerow plantings have also been investigated extensively in the study region (Long et al. 2017), but the impact of hedgerows on soil C storage is poorly understood. The objectives of the study were to: 1) compare soil C and physiochemical properties in cultivated fields and adjacent hedgerows (10+ years) to a depth of 1 meter, using a regional survey; 2) evaluate the difference in soil C between management types, across a range of soil types; 3) compare a typical sampling method (concentrations at 0-20 cm) with whole profile soil C stocks (0-100 cm); and 4) identify the

factors that contribute most to the accrual of soil C in the surface (0-20 cm) and subsurface (20-100 cm) in cultivated fields and adjacent hedgerows.

2.3 Materials and Methods

2.3.1 Site Description

This regional survey was conducted across the nearly level, lowland alluvial plains, fans, and terraces of Yolo County (Figure 2.1), situated in the southern Sacramento Valley, California, USA. Sites ranged in elevation from 16 to 140 m above sea level. The region is characterized by cool, wet winters and hot, dry summers; a xeric soil moisture regime (annual precipitation from 40-56 cm) and a thermic soil temperature regime (average annual temperature of 10–17°C) (Andrews 1972). Soils are developed largely from materials deposited from the Coast Range to the west.

The study area was historically dominated by oak woodlands, savannas, and wetlands, but is characterized today by intensive irrigated agriculture with dominant crop rotations including wheat, processing tomato, alfalfa hay, sunflower, safflower, wine grapes, almonds, and rice. Since the mid-1990's multi-stakeholder collaborations have helped farmers establish hedgerows, filter strips, and vegetated riparian corridors on agricultural lands, (Earnshaw et al. 2004; Brodt et al. 2009), resulting in approximately 175 acres of hedgerows in Yolo Co. with goals to establish an additional 100 miles by 2030 (Climate Action Plan).

2.3.2 Site Selection

Criterion for site selection included hedgerows that were 1) greater than or equal to 10 years in age; 2) greater than or equal to 5 feet in height; 3) contiguously planted without

contiguous grass cover; 4) immediately adjacent to a cultivated row or field crop; 5) not situated in or along a waterway or irrigation canal; and, 6) where soil had not been reworked, moved, or made into a berm.

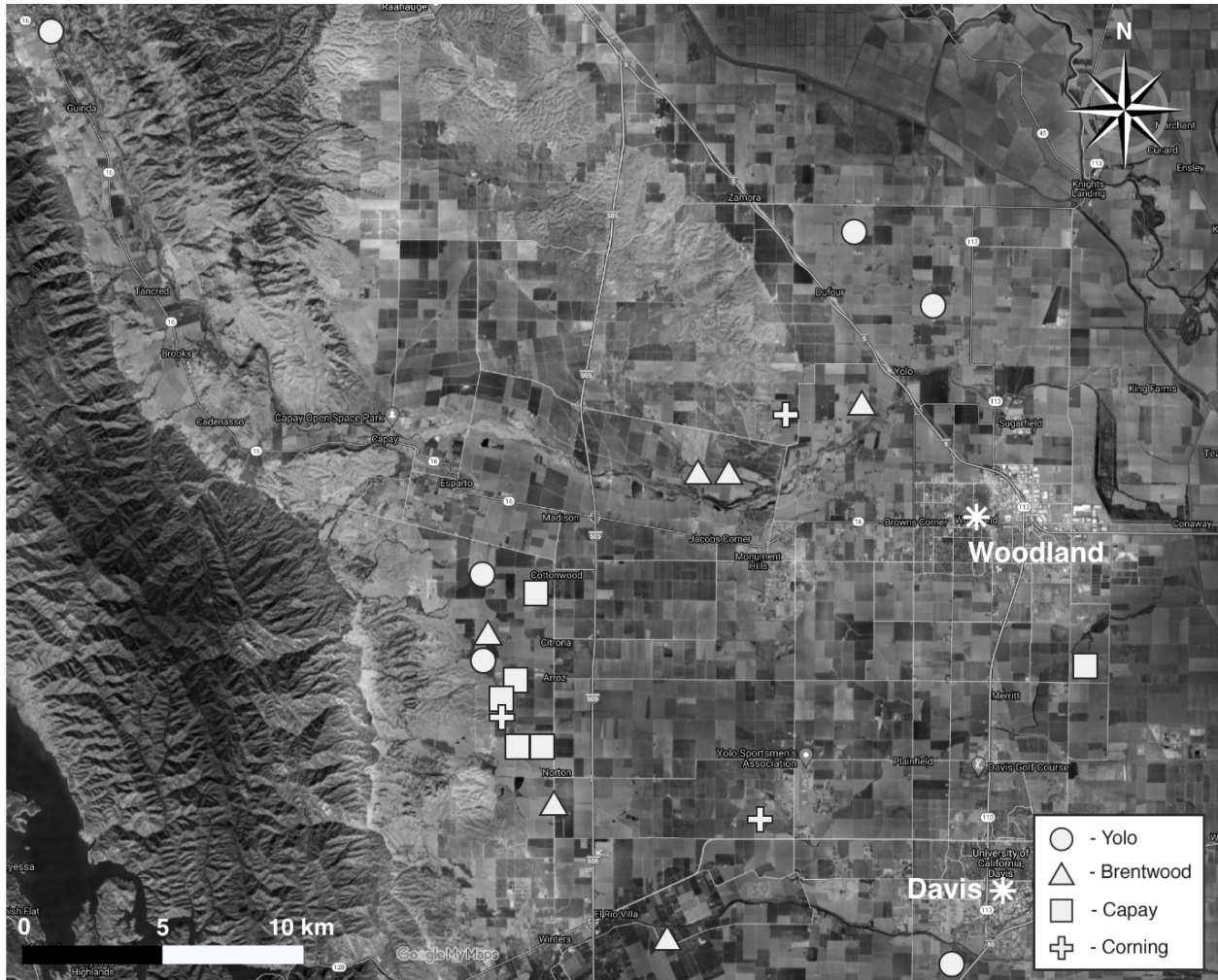


Figure 2.1. Map of 21 sampling locations across Yolo County, California; designated by soil type, including Yolo silt loam (n=6), Brentwood clay loam (n=6), Capay silty clay (n=6), and Corning loam (n=3).

We chose to study four agriculturally representative soil types spanning a range in soil development. Twenty-one paired sites were identified (Figure 2.1), which include: six on Yolo

silt loam (*Fine-silty, mixed, superactive, nonacid, thermic Mollic Xerofluvents*), six on Brentwood clay loam (*Fine, smectitic, thermic Typic Haploxerepts*); six on Capay silt clay (*Fine, smectitic, thermic Typic Haploxererts*); and three on Corning loam (*Fine, mixed, semiactive, thermic Typic Palexeralfs*) (Table 2.1). Yolo soils represent 601 km² (or 148,463 acres) in the state, Brentwood soils occupy 206 km² (or 50,940 acres); Capay soils are found on 1216 km² (or 300,576 acres); and Corning soils on 559 km² (or 138,240 acres). Field sizes ranged from 25,010 m² to 995,931 m² (or 6.18 to 246.1 acres), occupying an average of 259,768 m² (or 64.2 acres). Hedgerows covered an area ranging from 461 m² to 12,262 m² and constituting 0.33-9.43% of total field area, or an average 3,521 m² and 1.89%, respectively (O'Geen et al., 2017).

Cultivated fields represented similar cropping systems (furrow irrigated tomato/wheat or tomato/corn rotations), despite variability in management and current crop in rotation at time of sampling (Table 2.1). One perennial system was sampled to achieve a sufficient sample size on the Corning series. Corning soils are less commonly found in the county, especially under irrigated agriculture (Andrews 1972), necessitating the inclusion of a vineyard. One-on-one interviews were conducted in May 2019 to characterize management practices in both hedgerows and cultivated fields.

Hedgerows consisted of predominantly shrubs with occasional tree species. Commonly occurring species included Willow (*Salix spp.*), Ceanothus (*Ceanothus spp.*), Elderberry (*Sambucus exicana*), California coffeeberry (*Rhamnus californica tomentella*), Toyon (*Heteromeles arbutifolia*), Saltbush (*Atriplex lentiformis*), Coyote Brush (*Baccharis pilularis*), Western Redbud (*Cercis occidentalis*), and Milkweed (*Asclepias spp.*). Hedgerows ranged in age from 10-25 years (mean = 17 years) and were all established with irrigation and amendments

(compost and/or mineral fertilizer) in the first three years, although levels of maintenance (i.e. pruning and weeding) may have varied over their lifetime.

Table 2.1. Site information for 21 hedgerows and adjacent cultivated fields in Yolo County, California. Management practices represent the typical management for the past 5 years, while crop refers to the current crop in the rotation.

Site	Soil Type	Soil Textural Class	Hedgerow Age (yrs)	Compost (tons ha ⁻¹ yr ⁻¹)	Crop	Cover Crop (months)	Fallow Crop (months)
1	Yolo	Silt loam	20	0	wheat	N	6
2	Yolo	Silt loam	23	0	tomato	N	4.5
3	Yolo	Silt loam	11	0	tomato	N	7
4	Yolo	Silt loam	10	24	diverse	Y	2
5	Yolo	Silt loam	19	0	tomato	N	7
6	Yolo	Loam	10	4	tomato	N	7
7	Brentwood	Clay loam	13	12	diverse	Y	2
8	Brentwood	Clay loam	10	6	tomato	Y	2
9	Brentwood	Clay loam	10	6	tomato	Y	2
10	Brentwood	Clay loam	14	0	tomato	N	4.5
11	Brentwood	Clay loam	16	4	wheat	N	6
12	Brentwood	Clay loam	23	0	tomato	N	4.5
13	Capay	Silty clay	20	0	tomato	N	4.5
14	Capay	Silty clay	20	0	tomato	N	4.5
15	Capay	Silty clay	25	0	rye	N	3
16	Capay	Silty clay	25	0	rye	N	3
17	Capay	Silty clay	20	0	wheat	N	4.5
18	Capay	Silty clay	15	0	wheat	N	4.5
19	Corning	Loam	25	0	poppies	N	3
20	Corning	Loam	10	0	grapes	N	4
21	Corning	Loam	11	0	oat hay	N	2

Yolo = Mollic Xerofluvent, Brentwood = Typic Haploxerept, Capay = Typic Haploxerert, Corning = Typic Palexeralf

¹ diverse = cultivation of more than one species at the same time

2.3.3 Soil Sampling and Profile Descriptions

In April 2019, prior to spring irrigation, soil samples were collected from each site. Three locations were selected along a 100 m transect within the hedgerow, using a random number generator (Figure 2.2). To avoid an edge effect (impact of traffic/equipment), but minimize variability in inherent soil properties, three locations were selected 50-m directly parallel to hedgerow locations, within the cultivated field. Soil pits were dug by hand or with an excavator, where possible, in the central sampling location of both the hedgerow and the cultivated field, while 10-cm diameter augers were used to collect samples on the flanks of each transect.

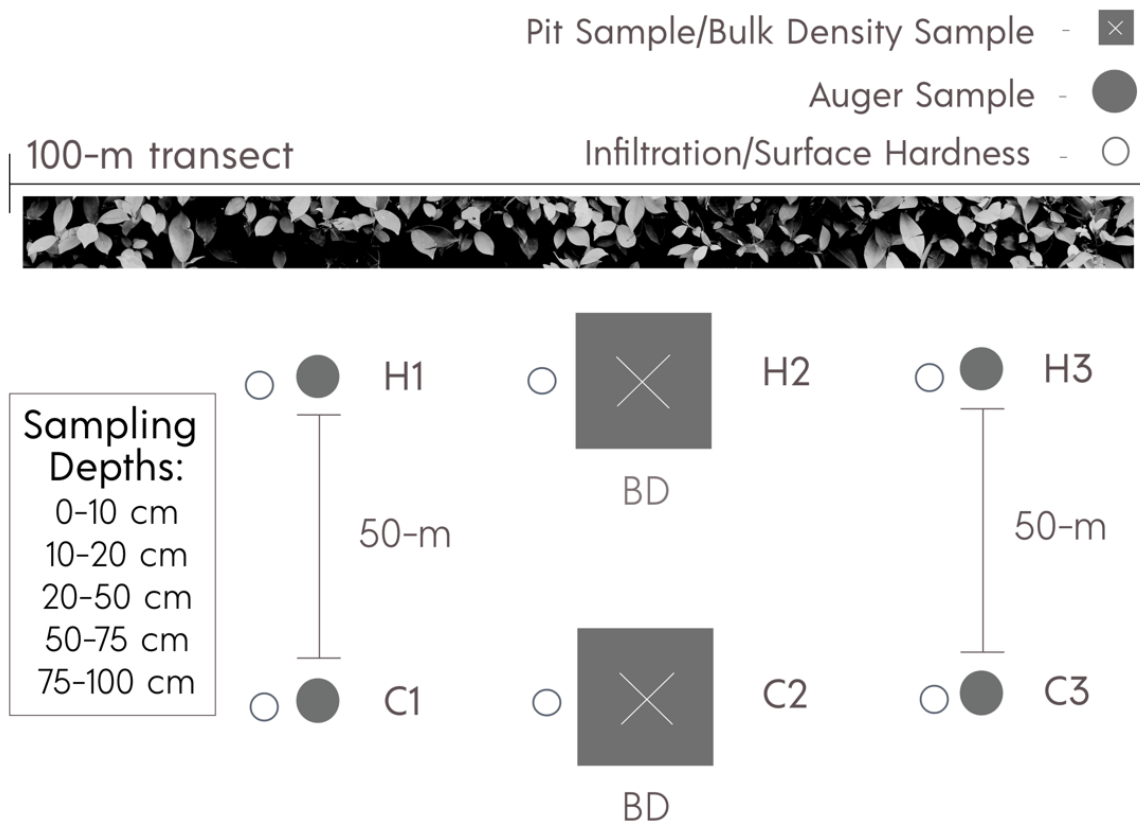


Figure 2.2. Sampling design for measuring soil properties and conducting in-field monitoring tests across a 100-m transect in hedgerows and 50-m away in adjacent cultivated fields.

Samples were collected from depth increments of 0-10, 10-20, 20-50, 50-75, and 75-100 cm, representing agriculturally relevant surface horizons and similar genetic horizons across soil types. Soils from each sampling location were kept separate, but were thoroughly homogenized before bagging for subsequent analysis of total soil carbon, total nitrogen, texture, and pH. Soils were stored at 4°C until field sampling was complete (no more than 12 days).

Soil profile descriptions were conducted using standard soil survey techniques (Schoeneberger et al. 2002). The following morphologic indicators were characterized: (1) A-horizon thickness; (2) depth to redoximorphic features; (3) maximum rooting depth, 4) root size and quantity; and (5) type, size, and grade of soil structure. Redoximorphic features were not encountered at any of the sites. Rooting intensity was calculated by assigning values of 1-5 for very fine, fine, medium, coarse, and very coarse root sizes and values of 1-3 for few, common, and many quantities; multiplying for all sizes present in each sampling depth; summing up the total for each depth; and calculating a weighted average for 0-20 cm and 20-100 cm depths. The quantity of earthworms present was also documented and converted to an index of 1-3 for few (1-2), common (3-4), and many (5+).

2.3.4 Bulk Density

At each pit (Figure 2.2), bulk density samples were collected from the center of each sampling depth using the core method (8.25 cm diameter, 7.5 cm length) (Blake & Hartge, 1986). For depths greater than 10 cm, 2 cores were collected in sequence (i.e. 27.5 – 35 cm and 35 – 42.5 cm for 20-50 cm depth); not directly on top of one another to avoid potential compaction. Rock fragments (>2mm) present in Corning soils were weighed, wrapped in

paraffin wax and submerged in water to determine volume. Mass and volume of rock fragments, where present, were removed from the total mass and volume prior to calculating bulk density.

2.3.5 Soil Properties

Soil samples were air-dried at 25°C and sieved to <2mm. All visible plant materials, including fine roots, were removed and subsamples oven-dried at 60°C for 72 hours before grinding in metal cylinders for 12-24 hours, or until a fine powder was achieved. Soil pH was measured on <2mm sieved samples in a 1:2 solution with 0.01M CaCl₂ using a pH electrode (Miller & Kissel, 2010). Soil texture was measured by hydrometer (Gavlack et al. 2005). Total C and N were determined on ground/ball milled samples by dry combustion using an ECS 4010 Costech Elemental Analyzer and a LECO soil standard (Blair et al., 1995). Samples with a pH over 7.4 were treated with 1N HCl to remove carbonates (Hedges & Stern 1984).

Total soil C and N were calculated on a mass basis for each depth, in order to convert concentrations to stocks (Equation 1):

$$C_i = BD_i \times d_i \times (c_i/100) \quad (1)$$

Where C_i is the total mass of soil C (kg m^{-2}) for sampling depth i , BD is bulk density (kg m^{-3}) of sampling depth i , d_i is the length (m) of sampling depth i , and c is the concentration of soil carbon (g kg^{-1} soil) for sampling depth i . Profile C stocks (0-100 cm) were calculated by summing total C (kg m^{-2}) from each individual soil depth (Batjes, 2014). Weighted averages of measured C data collected at 0-10 cm and 10-20 cm depths were used for 0-20 cm values and from samples collected at 20-50 cm, 50-75 cm, and 75-100 cm for 20-100 cm values.

$$S_i = (\sum_i^n (s_i \times d_i))/l_t \quad (2)$$

Where S_i is the total mass of soil C (kg m^{-2}) for the total aggregated depth (0-20 or 20-100 cm), s_i is the stock of soil carbon (kg m^{-2}) for sampling depth i , d_i is the length (m) of sampling depth i , and l_t is the total length of the aggregated depth.

2.3.6 Statistical Analyses

Data for total C and N, BD, pH, sand and clay were tested for normality and homogeneity of variance and normalized using $\log(x + 1)$ transformations when necessary to meet ANOVA assumptions. Effects of management (hedgerow vs cropped) and soil type on each variable were analyzed using a mixed-effects model using the R statistical package nlme (Pinheiro et al. 2018). Management (within-subject factor) and soil type (between-subject factor) were considered fixed effects, while site was considered a random effect (based on repeated measures). Data was analyzed separately for each individual sampling depth, as well as for the entire 0–100 cm. Differences between means were calculated using Tukey's Honestly Significant Difference (HSD) tests. Statistical significance was evaluated at $P < 0.05$ unless otherwise stated.

Box plots (SI Figure 2.2) were graphed using the *ggplot* package in R (Wickham, 2009). Linear regression models were used to evaluate the relationship between C concentrations and silt + clay content (Figure 2.6), as well as the ability of C concentrations in the surface 0-20 cm to predict whole profile C stocks (kg m^{-2}) (Figure 2.6) or C concentrations in the subsurface 20-100 cm (SI Figure 2.2). Residuals were tested for normality and homogeneity of variance, and analysis of covariance (ANCOVA) was used to determine if relationships were different across management types.

Sources of variability in the dataset were characterized by Principal Component Analysis (PCA) (Figure 2.7) on a standardized correlation matrix using the *vegan* package in R (Oksanen

et al., 2012). Loadings and proportions of variance, as well as the raw data for included variables are presented in SI Table 2.3 and 2.4. The first three components were selected based on visual interpretation of the scree plots and criteria of having eigenvalues >1 and a cumulative variance of at least 70% (Jolliffe 2002). Spearman's correlation coefficients and significance of correlations at $P < 0.05$ were calculated for measured soil properties using the Hmisc package in R (Harrell & Dupont, 2018).

2.3.7 Scenario Estimates

To evaluate the viability of hedgerow plantings in achieving state and county-wide emissions reductions goals, the potential for C sequestration was assumed to be equivalent to the difference between measured C stocks (0-100 cm) in hedgerows and cultivated fields. A total of 949 and 76,500 farms and an average farm size of 1.96 and 1.26 km² (or 484 and 311 acres) were used for Yolo County and the state of CA, respectively. Farms were assumed to be square to estimate the perimeter of an average farm in each region. Based on recommendations for implementation and a literature review of 60 studies, hedgerows were assumed to be an average of 5-m wide (Earnshaw et al. 2004; Long et al. 2010; van Vooren et al. 2017) to calculate the total area of hedgerows if the entire perimeter of each farm were re-vegetated in hedgerows. The total potential hedgerow area was then multiplied by the total number of farms and either 0.5 or 0.8 to represent a 50% or 80% adoption scenario. The final area was back converted to hectares for both Yolo County (1,295 or 2,073 ha) and the state of CA (77,167 or 123,467 ha) and multiplied by 34 Mg/ha, the average difference in C stocks under hedgerows relative to adjacent cultivated fields. Finally, the total Mg of C was converted to Mg of CO₂ (or CO₂e) using a conversion factor of 3.67 (44g CO₂/12g C).

Table 2.2. Soil properties to a depth of 1 meter according to soil type and management type (hedgerows and cultivated fields) for 21 sites in Yolo County, California. Cultivated fields were sampled 50 m away from hedgerows, which were located along field margins.

Soil Type	Texture Class	Sand Content (g 100 g ⁻¹ soil)				Clay Content (g 100 g ⁻¹ soil)				pH									
		Hedgerow		Cultivated		Hedgerow		Cultivated		Hedgerow		Cultivated							
0-10 cm depth																			
Yolo	SiCL	17.4	(1.08)	18.3	(1.29)	ns	c	26.8	(0.65)	27.6	(0.68)	ns	b	6.8	(0.07)	6.8	(0.05)	ns	a
Brentwood	Cl	25.6	(1.03)	25.2	(0.90)	ns	b	30.9	(0.50)	31.8	(0.63)	ns	b	6.5	(0.05)	6.5	(0.04)	ns	b
Capay	Clay	16.0	(1.22)	15.2	(1.06)	ns	c	49.6	(1.07)	49.6	(1.15)	ns	a	6.6	(0.10)	6.6	(0.10)	ns	bc
Corning	Loam	36.0	(2.71)	38.3	(1.44)	ns	a	17.2	(1.58)	16.5	(1.64)	ns	c	5.5	(0.04)	5.2	(0.07)	ns	c
10-20 cm depth																			
Yolo	SiCL	17.0	(0.98)	18.0	(1.34)	ns	c	26.9	(0.63)	27.9	(0.89)	ns	b	6.8	(0.07)	6.7	(0.06)	ns	a
Brentwood	Cl	24.2	(1.06)	23.3	(1.05)	ns	b	31.4	(0.78)	32.1	(0.84)	ns	b	6.5	(0.04)	6.6	(0.03)	ns	b
Capay	Clay	16.7	(0.85)	14.6	(0.94)	ns	c	50.8	(1.04)	50.9	(1.20)	ns	a	6.6	(0.10)	6.6	(0.10)	ns	ab
Corning	Loam	36.6	(1.71)	36.3	(2.14)	ns	a	17.6	(1.35)	16.1	(1.70)	ns	c	5.4	(0.04)	5.3	(0.07)	ns	c
20-50 cm depth																			
Yolo	SiCL	17.3	(0.73)	17.8	(1.60)	ns	c	26.8	(0.81)	26.5	(1.27)	ns	c	6.6	(0.07)	6.5	(0.04)	ns	b
Brentwood	Cl	23.2	(1.11)	21.5	(0.97)	ns	b	35.2	(1.06)	36.0	(0.86)	ns	b	6.3	(0.05)	6.3	(0.03)	ns	b
Capay	Clay	16.8	(1.15)	15.3	(0.62)	ns	c	48.9	(0.97)	49.9	(0.81)	ns	a	7.1	(0.07)	7.0	(0.05)	ns	a
Corning	Loam	43.3	(1.30)	39.1	(2.32)	ns	a	24.1	(1.11)	22.9	(0.84)	ns	c	5.5	(0.07)	5.2	(0.10)	*	c
50-75 cm depth																			
Yolo	SiL	19.5	(0.77)	17.5	(1.76)	ns	c	25.1	(0.80)	25.0	(1.50)	ns	c	6.6	(0.07)	6.6	(0.04)	ns	b
Brentwood	CL	23.5	(1.36)	23.3	(1.01)	ns	b	31.6	(0.90)	31.5	(0.80)	ns	b	6.3	(0.05)	6.4	(0.04)	ns	b
Capay	Clay	16.1	(1.38)	15.0	(0.95)	ns	c	50.5	(1.13)	50.8	(1.06)	ns	a	7.1	(0.06)	7.0	(0.05)	ns	a
Corning	CL	45.3	(0.94)	43.3	(1.16)	ns	a	30.3	(1.68)	32.3	(1.52)	ns	bc	5.5	(0.10)	5.2	(0.06)	*	c
75-100 cm depth																			
Yolo	SiL	25.6	(1.10)	26.0	(1.26)	ns	b	21.7	(1.05)	22.6	(1.00)	ns	b	6.6	(0.06)	6.6	(0.06)	ns	b
Brentwood	Loam	28.3	(1.43)	27.9	(1.14)	ns	b	23.8	(1.10)	23.1	(1.01)	ns	b	6.4	(0.04)	6.4	(0.03)	ns	b
Capay	Clay	18.1	(1.78)	17.9	(1.05)	ns	c	49.4	(1.32)	49.7	(1.43)	ns	a	7.1	(0.06)	7.1	(0.06)	ns	a
Corning	SL	51.2	(1.60)	49.0	(0.32)	ns	a	25.5	(0.60)	27.5	(1.23)	ns	b	5.6	(0.13)	5.1	(0.04)	ns	c

Within a column, for each depth, values for soil type followed by the same letter are not significantly different at $P < 0.05$. Soil properties were not significantly different by management ($P < 0.05$) at any soil depth, except pH from 20-75 cm in Corning soils. Numbers in parentheses indicate standard error ($n = 18$); for Corning ($n=9$). SiCL = Silty clay loam; CL = Clay loam; SiL = Silt Loam; SL = Sandy Loam

2.4 Results and Discussion

2.4.1 Whole Profile Stocks

Whole profile (0-100 cm) soil C (kg m^{-2}) stocks across all sites differed by land-use and soil type ($P < 0.001$) (Figure 2.3). Whole profile stocks were on average 36% greater in hedgerows, (mean = 14.4 kg m^{-2} , range = $7\text{-}26 \text{ kg m}^{-2}$) compared to cultivated fields (mean = 10.6 kg m^{-2} ; range = $4\text{-}19 \text{ kg m}^{-2}$) (Figure 2.3). Similar magnitudes of soil C stock increases (32-34%) have been found in agroforestry systems relative to adjacent croplands to depths of 75-100 cm (DeStefano & Jacobson et al. 2018; Lim et al. 2018). Whole profile stocks were 24%, 27%, 49% and 74% greater under hedgerows on Yolo, Brentwood, Capay, and Corning soils, respectively, with absolute differences ranging from 2.6 kg m^{-2} for Yolo to 5.8 kg m^{-2} for Capay. Whole-profile stocks for cultivated fields were similar to those found at a nearby long-term research station with similar soil types (Yolo and Rincon soil series), where conventionally managed fields averaged 10.7 kg m^{-2} and organically managed fields (receiving annual compost applications and cover crops (CC)) resulted in soil C stocks of 13.1 kg m^{-2} to a depth of 100 cm (Tautges et al. 2019). In this study, cultivated fields that were managed organically and received CC and annual compost applications had mean soil C stocks (0-100 cm) of 13.0 kg m^{-2} , while all other cultivated fields stored 9.54 kg m^{-2} , indicating that organic management can help close the gap between cultivated fields and hedgerows.

Other studies across a diversity of climates and soil types have reported a range of 3-30 kg m^{-2} (0-100 cm) under trees/shrubs in agroforestry systems (Cardinael et al. 2015). Thiel et al. 2015 found that planted hedgerows adjacent to cropland on Inceptisols in British Columbia stored a mean of 17.6 kg m^{-2} from 0-100 cm; 40% greater than adjacent cultivated fields. In

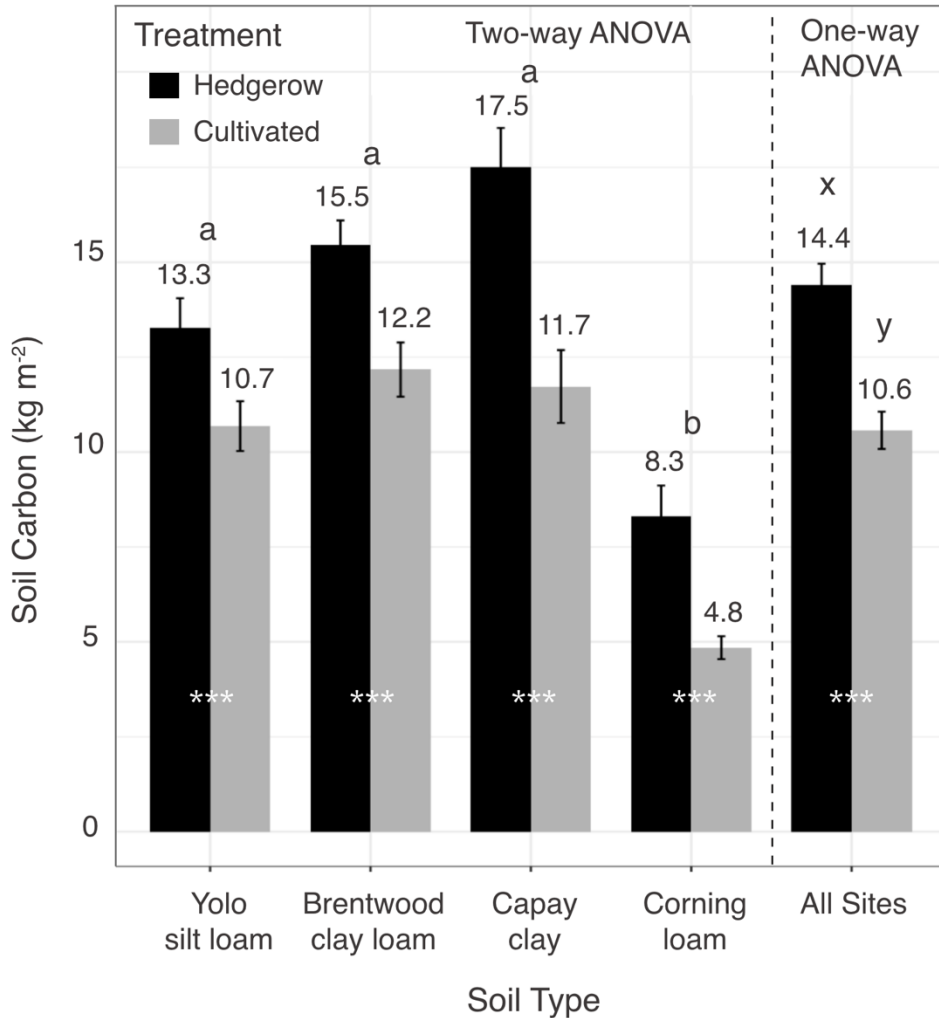


Figure 2.3. Mean soil carbon stocks by management and soil type calculated using soil carbon concentration and bulk density from 5 distinct depths to 1-m in hedgerows and adjacent cultivated fields (Two-way ANOVA). One-way ANOVA refers to soil carbon stocks across sites by management type. Number at the top of each column indicates the sample mean, bars indicate standard error. Letters indicate significant differences between soil types and asterisks indicate significant differences by management type in Tukey means comparisons at $P < 0.05$ ($n=21$ for hedgerow, $n=21$ for cultivated fields, $n=6$ for each soil type, except Corning $n=3$).

western France, hedgerows on Inceptisols and Alfisols were found to contribute 15.5 kg m^{-2} to a depth of 90 cm; 42% greater than adjacent cultivated fields (10.9 kg m^{-2}) (Viaud & Kunnemann 2021). Within California, across a range of soil types, soil C (0-100 cm) was approximately 9.0 kg C m^{-2} higher in reforested riparian corridors relative to adjacent cropland (Young-Mathews et

al. 2010) and 11-20 kg m⁻² higher in woodland ecosystems relative to adjacent vineyards (Williams et al. 2011).

2.4.2 Soil C Concentrations and Stocks by Depth

When analyzed by depth increments, soil C concentrations (g kg⁻¹) and soil C stocks (kg m⁻²), were significantly higher under hedgerows at all depths and across all soil types, except at 10-20 cm and 20-50 cm in Yolo (concentrations only) and Brentwood, although trends still showed higher soil C under hedgerows (Table 2.3, SI Figure 2.1). At 0-10 cm, soil C in hedgerows was nearly double that of cultivated fields across soil types. This is likely due to increased litter deposition (Chander et al. 1998; Lenka et al. 2012; Cardinael et al. 2017; Ramos et al. 2018), prevalence and turnover of fine roots (Lehmann and Zech 1998; Nair et al. 2010), as well as, the lack of tillage under hedgerows. For example, a comparison of agroforestry systems relative to adjacent wheat fields measured 40% greater organic inputs under agroforestry systems (Cardinael et al. 2017).

Under perennial woody shrubs, the physical environment may be altered in ways that impact microbial activity and overall C-dynamics. Hedgerows have been found to create more favorable microclimates (Sanchez et al. 2010; Kanzler et al. 2019; Veste et al. 2020), resulting in lower air and surface soil temperatures (Clinch et al., 2009; Dubbert et al., 2014). This may contribute to increased carbon use efficiency or lower specific respiration rates in the surface 0-10 cm (Allison et al. 2010; Frey et al. 2013; Doetterl et al. 2015). Moisture content may be lower or not significantly different, as year-round vegetative cover increases transpiration, some of which may be offset by reduced evaporation and increased infiltration (Merot et al. 1999; Ilstedt et al., 2016; Kanzler et al. 2019). Lower moisture content slows microbial activity.

Table 2.3. Bulk density, soil carbon concentrations and soil carbon stocks by soil type and management type at 5 distinct depths from 0-100 cm for 21 sites in Yolo County, CA.

Soil Type	Bulk Density (g cm ⁻³)						Total Soil Carbon (g kg ⁻¹)						Soil Carbon Stocks (kg m ⁻²)					
	Hedgerow		Cultivated				Hedgerow		Cultivated				Hedgerow		Cultivated			
	0-10 cm depth																	
Yolo	1.29	(0.06)	1.29	(0.03)	ns	b	2.62	(0.27)	1.36	(0.14)	***	a	3.27	(0.22)	1.74	(0.17)	***	a
Brentwood	1.32	(0.05)	1.23	(0.05)	ns	b	2.52	(0.16)	1.30	(0.08)	***	a	3.26	(0.17)	1.57	(0.08)	***	a
Capay	1.46	(0.06)	1.40	(0.05)	ns	a	2.51	(0.16)	1.03	(0.09)	***	a	3.63	(0.21)	1.47	(0.15)	***	a
Corning	1.24	(0.09)	1.46	(0.07)	***	b	2.47	(0.30)	1.01	(0.12)	***	a	3.01	(0.35)	1.48	(0.20)	***	a
	10-20 cm depth																	
Yolo	1.49	(0.06)	1.44	(0.03)	ns	b	1.42	(0.16)	1.15	(0.13)	ns	a	2.05	(0.19)	1.66	(0.19)	*	a
Brentwood	1.40	(0.06)	1.39	(0.05)	ns	b	1.29	(0.08)	1.20	(0.10)	ns	a	1.80	(0.11)	1.66	(0.13)	ns	a
Capay	1.69	(0.04)	1.61	(0.05)	ns	a	1.29	(0.1)	0.90	(0.07)	***	a	2.18	(0.16)	1.45	(0.11)	***	a
Corning	1.49	(0.09)	1.47	(0.08)	ns	b	1.01	(0.13)	0.70	(0.06)	*	a	1.53	(0.23)	1.03	(0.08)	*	a
	20-50 cm depth																	
Yolo	1.48	(0.08)	1.52	(0.05)	ns	b	0.82	(0.08)	0.77	(0.05)	ns	a	3.53	(0.27)	3.53	(0.29)	ns	a
Brentwood	1.53	(0.04)	1.56	(0.04)	ns	b	0.99	(0.07)	0.90	(0.06)	ns	a	4.48	(0.29)	4.17	(0.27)	ns	a
Capay	1.72	(0.03)	1.75	(0.05)	ns	a	1.03	(0.07)	0.75	(0.07)	***	a	5.32	(0.38)	3.94	(0.37)	***	a
Corning	1.67	(0.09)	1.59	(0.08)	ns	ab	0.43	(0.02)	0.33	(0.02)	***	b	2.17	(0.14)	1.58	(0.13)	***	b
	50-75 cm depth																	
Yolo	1.47	(0.02)	1.49	(0.03)	ns	c	0.64	(0.03)	0.54	(0.03)	*	a	2.34	(0.12)	2.01	(0.12)	*	a
Brentwood	1.50	(0.06)	1.43	(0.05)	ns	c	0.88	(0.05)	0.72	(0.05)	***	a	3.25	(0.17)	2.53	(0.17)	***	a
Capay	1.78	(0.02)	1.80	(0.04)	ns	a	0.77	(0.07)	0.60	(0.05)	***	a	3.44	(0.31)	2.68	(0.25)	***	a
Corning	1.67	(0.06)	1.68	(0.09)	ns	b	0.24	(0.03)	0.14	(0.02)	***	b	1.01	(0.13)	0.56	(0.08)	***	b
	75-100 cm depth																	
Yolo	1.39	(0.06)	1.48	(0.06)	ns	c	0.60	(0.04)	0.47	(0.03)	***	a	2.09	(0.15)	1.75	(0.14)	*	a
Brentwood	1.40	(0.05)	1.42	(0.05)	ns	c	0.76	(0.04)	0.63	(0.04)	***	a	2.68	(0.16)	2.25	(0.17)	***	a
Capay	1.76	(0.03)	1.77	(0.01)	ns	a	0.67	(0.05)	0.49	(0.04)	***	a	2.93	(0.23)	2.19	(0.17)	*	a
Corning	1.6	(0.07)	1.57	(0.12)	ns	b	0.21	(0.06)	0.08	(0.01)	***	b	0.89	(0.27)	0.29	(0.02)	***	b

Within a column, for each depth, values for soil type followed by the same letter are not significantly different at $p < 0.05$.

Asterisks indicate significant difference between management treatments (* = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$).

Numbers in parentheses indicate standard error (n = 18); for Corning (n=9).

The greater levels of soil C at 50-75 and 75-100 cm under hedgerows could be due to multiple mechanisms, including deep roots and their exudates (Upson and Burgess, 2013; Germon et al., 2016), increased dissolved organic carbon that accompanies greater surface inputs (Kaiser & Kalbitz 2012; Toosi et al. 2012), and/or increased bioturbation (Wilkinson et al. 2009). Cessation of tillage can lead to increased prevalence of earthworms (House & Parmelee 1985; Rovira et al. 1987; Briones & Schmidt 2017) and other invertebrates (Stinner & House 1990; Neave and Fox 1998; Errouissi et al. 2011), which can redistribute organic materials throughout the profile.

Hedgerow soil C concentrations were consistently higher at paired sites, with 94% of sampling locations higher at 0-10 cm, 70% from 10-20 and 20-50 cm, 78% from 50-75 cm, and 83% from 75-100 cm (Table 2.3). At 0-10 cm, all paired sampling locations with higher soil C in cultivated fields, were under diversified cropping systems receiving compost + CC. At 10-20, 20-50 cm, and 75-100 cm, 2/3 of sampling locations with higher soil C in cultivated fields were under field crops (wheat, rye) or diverse crops (with compost + CC). At 50-75 cm, 71% were under field crops. Several studies in California and other semi-arid environments have shown higher soil C with compost applications, (Poudel et al. 2001; Brown & Cotton et al. 2011; Aguilera et al. 2013; Tautges et al. 2019), while field crops like wheat and rye are known to have deep fibrous roots (Baveye & Laba 2015; Thorup-Kristensen et al. 2020), which when irrigated and fertilized (as in our study sites) have been shown to increase soil C (Gan et al. 2012; Novara et al. 2016; Tautges et al. 2019).

When comparing hedgerow soils only, soil C concentrations increased with hedgerow area (m²) at 10-20 cm (P=0.015) and 20-50 cm (P=0.029). Arrouays et al. (2002) also found that soil C varied by the size of the stand, in addition to height and location in the landscape. Age of

stand, however, did not significantly impact soil C concentrations or stocks at any depth. This may be due to the narrow age range (10-25 years) of hedgerows in this study. It has been found that soil C eventually reaches a steady-state equilibrium after a change in management with sequestration rates peaking at ~10 years (21 with cessation of tillage), achieving maximum sequestration by year 7 (West & Six 2007). As such, models frequently assume a default period of 20 years for C accrual (Houghton et al. 1997; Arrouays et al. 2002; Eggleston et al. 2006; Stewart et al. 2007). A study of hedgerows greater than 20 years old found that age of stand did not significantly impact soil C (Viaud & Kunnemann 2021), while others have found a weak negative correlation between soil C and age (9-45 years) (Thiel et al. 2015).

When comparing cultivated soils only, soil C concentrations were significantly higher with compost use at 0-10 cm ($P=2.09e-6$), 10-20 cm ($P=7.17e-7$), and 20-50 cm ($P=0.0033$). At 50-75 and 75-100 cm, cropping system had a significant impact on soil C concentrations ($P=4.03e-4$ and $P=6.48e-4$, respectively) with diversified, wheat and rye systems exhibiting stronger positive relationships with soil C than tomato rotations. When examining the differential between hedgerows and cultivated fields, compost use was the only factor to significantly impact the difference in soil C stocks with effects at 0-10 cm ($P=0.01$), 10-20 cm ($P=0.005$), and 20-50cm ($P=0.001$).

Depth distributions of soil C were significantly different between management types for most soil types (except Corning) with a greater proportion of C stored at 0-10 in hedgerows and a greater proportion in cultivated fields from 20-50 cm (Figure 2.4). Under hedgerows, Yolo, Brentwood, and Capay stored an average 22% of C (kg m^{-2}) in the 0-10 cm depth, 13% at 10-20 cm, 29% at 20-50 cm, and 36% at 50-100 cm. Under cultivated fields, these soils stored an average of 14% at the 0-10 and 10-20 cm depths, 33% at 20-50 cm, and 39% at 50-100 cm.

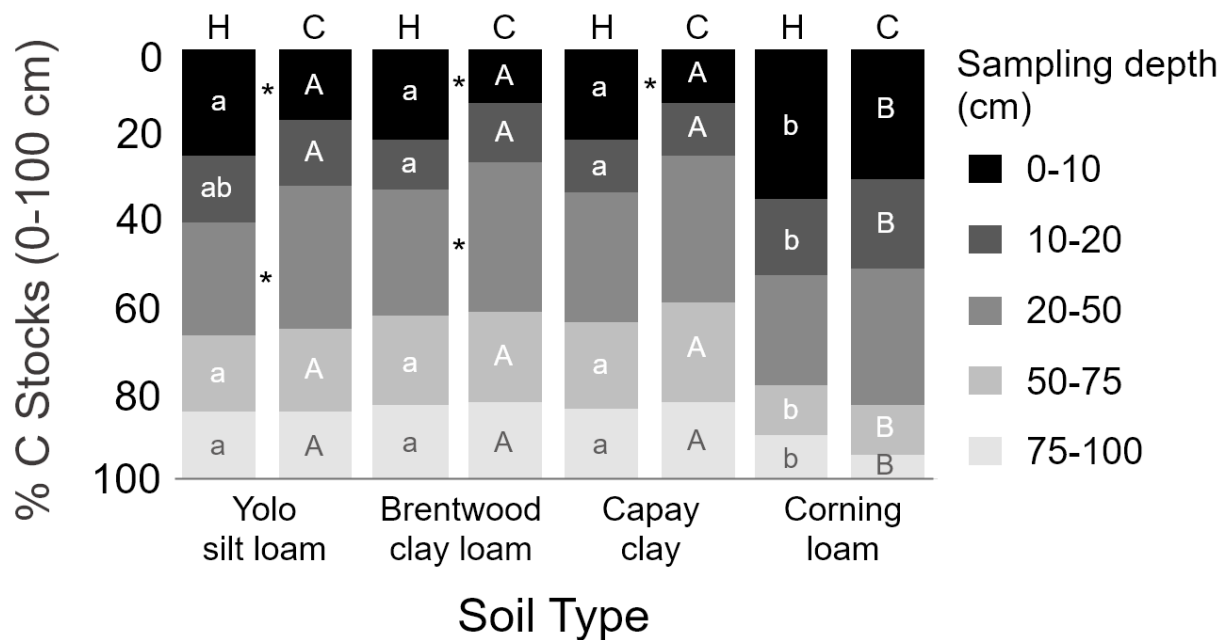


Figure 2.4. Proportion of whole profile C stocks (0-100 cm) situated in each sampling depth by management and soil type. For a given sampling depth, asterisks represent significant differences between hedgerows and cultivated fields within a given soil type, while letters show significant differences across soil types within a given management type (lowercase letters for H = hedgerows; uppercase letters for C = cultivated fields) at $P < 0.05$.

Corning soils differed from all other soils at nearly all depths with a greater proportion of C at 0-10 and 10-20 cm and a lesser proportion at 50-100 cm, and no difference at 20-50 cm (Figure 2.4). Under hedgerows, Corning soils held 35%, 18%, 25%, and 22% of C at 0-10, 10-20, 20-50, 50-100 cm, respectively. Corning cultivated fields contained 20%, 21%, 32%, 17% at 0-10, 10-20, 20-50, 50-100 cm. Global estimates suggest that 30-63% of soil C stocks (0-100 cm) are situated at 30-100 cm (Batjes et al. 1996; Jobaggy & Jackson, 2000; Harrison et al. 2011; Kogel-Knabner & Amelung 2021). Under hedgerows, specifically, studies have found 66% of SOC below 20 cm (Thiel et al. 2015).

While the proportion of total C stocks was higher in the subsoil of cultivated soils, the absolute amount was lower, indicating that the difference is attributed to lower relative C in the surface of cultivated fields, rather than mechanisms contributing additional C to depth. Long fallow without adequate C inputs has been shown to decrease SOC in Mediterranean agroecosystems (Rasmussen et al 1998, Guo & Gifford 2002; Machado et al 2011; Ghimire et al 2015). Heavy tillage also contributes to loss of soil C in the topsoil, as it disrupts aggregates, exposing SOM that was previously physically protected from microbes and their enzymes (Balesdent et al. 2000; Kladivko, 2001; Six et al. 2004; Williams and Hedlund 2013; Zakharova et al. 2014) and increases oxygen content, which in turn drives a pulse in microbial activity, increasing decomposition and overall loss of soil C (Calderon et al. 2000; Calderon and Jackson, 2002; Jackson et al. 2003; Reicosky et al. 2003).

The combination of climate and management may be limiting the potential for C storage in the surface 0-10 cm of cultivated fields, as bare fallow leaves fields exposed for several months of the year and can increase soil temperatures in semi-arid environments by 5-10°C relative to vegetative cover (Akinremi et al. 1999; Herrero et al. 2001; Fernandez et al. 2008; Mitchell et al. 2015). Although sensitivity of decomposition varies by substrate/SOM quality (Davidson & Janssens 2006), increased temperature is known to result in increased microbial metabolism and has been shown to result in a decrease in carbon use efficiency (Allison et al. 2010; Manzoni et al. 2012; Frey et al. 2013), which is increasingly understood to contribute to an overall loss of soil C (Kallenbach et al. 2019; Wang et al. 2021). The greater proportion of soil C at 20-50 cm in cultivated fields may be attributed to redistribution of C through tillage (Baker et al. 2007; Veneestra et al. 2007) and/or translocation of dissolved organic carbon associated with fresh residue inputs (Kaiser & Kalbitz 2012; Toosi et al. 2012).

Root exudates introduce labile forms of C, which can contribute to stable SOC formation (Schmidt et al. 2011; Cotrufo et al. 2013) and fine roots and exudates may be more readily stabilized by physical mechanisms in the subsoil (Rasse et al. 2005). Many agricultural crops have rooting depths of 2.1 ± 0.2 m and woody shrubs, such as those found in our hedgerows, commonly have rooting depths of 5.1 ± 0.8 m (Canadell et al. 1996; Baveye and Loba, 2015; Throrup-Kristensen et al. 2020) and although DOC is known to translocate C to the subsoil (Kaiser & Kalbitz et al. 2001), these dynamics are still poorly understood (Schmidt et al. 2011; Rumpel et al. 2011, 2012).

2.4.3 Soil C Concentrations and Stocks by Soil Type

Soil C concentrations (g kg^{-1}) and stocks (kg m^{-2}) were not significantly different by soil type at 0-10 or 10-20 cm, but Corning soil C concentrations and stocks were significantly different ($P < 0.05$) from all other soil types at each depth from 20-100 cm (Table 2.3, SI Figure 2.2). The lack of significance by soil type in the surface 0-20 cm corroborates several findings that climate is the main driver of surface soil C dynamics (Jobbágy and Jackson, 2000; Gray et al., 2009; Doetterl et al. 2015), while soil type is thought to predominate in the subsoil (Hobley et al. 2015; Mathieu et al. 2015; Torres Sallan et al. 2018; Mayer et al. 2019; Vos et al. 2019). The surprising lack of significant differences between Yolo, Brentwood, and Capay soils in the subsoil, despite a wide range of clay concentrations (21-51% in Yolo and Capay, respectively) is corroborated by Rasmussen et al. 2018, which found that clay was not an effective predictor of C storage, but rather, other physicochemical properties, such as dominant mineralogy and degree of weathering. It has been further postulated that mineral reactivity, rather than particle size, drives soil C stabilization or turnover and is a better predictor of overall C storage; with climate subsequently modulating these dynamics (Kogel-Knabner & Amelung 2021).

Yolo, Brentwood, and Capay soils are all derived from the same parent material (mixed alluvium from the Coast Range), are comprised of similar mineralogy (dominated by 2:1 smectitic clays), and exhibit a similar degree of weathering (less so in Yolo than Brentwood and Capay); whereas Corning soils are derived from different parent material and are significantly older and more weathered. While soils are pedogenically similar in ways that drive soil C stabilization/storage, each has unique mechanisms that may be contributing to substantial carbon stocks at depth including the burial of A horizons in Yolo soils (Chaopricha & Marin-Spiotta, 2013), illuviation of clays in Brentwood soils (Torres-Sallan et al. 2017), and pedoturbation and the development of vertic cracks into which plant residues can fall, as well as occasional oxygen-limitations, which may contribute to greater preservation in Capay (Mathieu et al. 2015; Kogel Knabner and Amelung 2021). The absolute difference in soil C (0-100 cm) was highest in Capay, which has the greatest prevalence of 2:1 smectitic clays and vertic cracking, followed by Corning, which had the lowest soil carbon content overall. The differential in soil C stocks between hedgerows and cultivated fields did not differ significantly by soil type at any depth or across the whole profile. This suggests that hedgerows may have a universally positive impact on soil C storage across soil types.

2.4.4 GHG Reduction Scenario

When considering their limited extent across an agroecosystem, hedgerows may not account for the greatest potential carbon sink on-farm (Follain et al. 2007), although one California study found that hedgerows accounted for 18% of total on-farm C, despite only occupying 6% of the area (Smukler et al. 2010). To better understand the potential towards greenhouse gas reduction goals, we used measured results in this study to estimate total county and state-wide potential for reforestation of farm edges with hedgerows. Based on the

assumptions laid out in the methods above, it is estimated that there are 2,591 and 154,334 hectares of farm edges in Yolo County and California state, respectively, that could be revegetated in hedgerows. Assuming a 50% adoption rate and an average increase in C storage of 38.3 Mg/ha, this could amount to 49,616 Mg C or 2,955,498 Mg C stored in Yolo County and California soils, respectively. This translates to 181,925 Mg CO₂e and 10.8 MMT CO₂e, respectively. At an 80% adoption rate, soil C storage would increase to 79,386 Mg C and 4,728,797 Mg C, respectively, or 291,090 Mg CO₂e and 17.3 MMT CO₂e. These adoption scenarios could account for 33-53% of Yolo County's GHG reduction goals and between 7 and 12% of statewide GHG reduction goals for one year. This takes at least 10 years to accumulate. This also does not include contributions from aboveground biomass. At an estimated average household emissions of 4.83 MT CO₂e (US Census 2011; Goldstein et al. 2020) this could offset 2-3.5 million cars on the road for one year.

It was estimated that 100 m of hedgerows could be implemented per hectare across all of the European Union's agricultural lands (3 times our estimate per farm), amounting to 178 million ha of hedges (Aertsens et al. 2013). At 0.10 Mg C ha⁻¹ yr⁻¹ of hedgerow, they estimated that hedgerows could store 65 MMT CO₂e/yr, or 2% of total annual emissions in the EU (based on 2007 data). In the UK, it was estimated that the C sequestration potential for field margins was between 0.1 and 2.4 % of their 1990-level CO₂ emissions (Falloon et al. 2004). A 2008 report to the California Air Resources Board estimated 202,350 hectares of field edges could be revegetated on crop and rangelands across the state, providing an additional technical potential of 2.9 MMT CO₂e in aboveground biomass (Asmus 2008).

2.4.5 Soil Physicochemical Properties

Soil physicochemical properties followed an opposite trend to soil C, differing among soil types at all depths, but not across management types (Table 2.2). Soil texture ranged from loam to clay among soil types with the lowest amount of silt + clay in surface horizons of Corning and the highest in Capay.

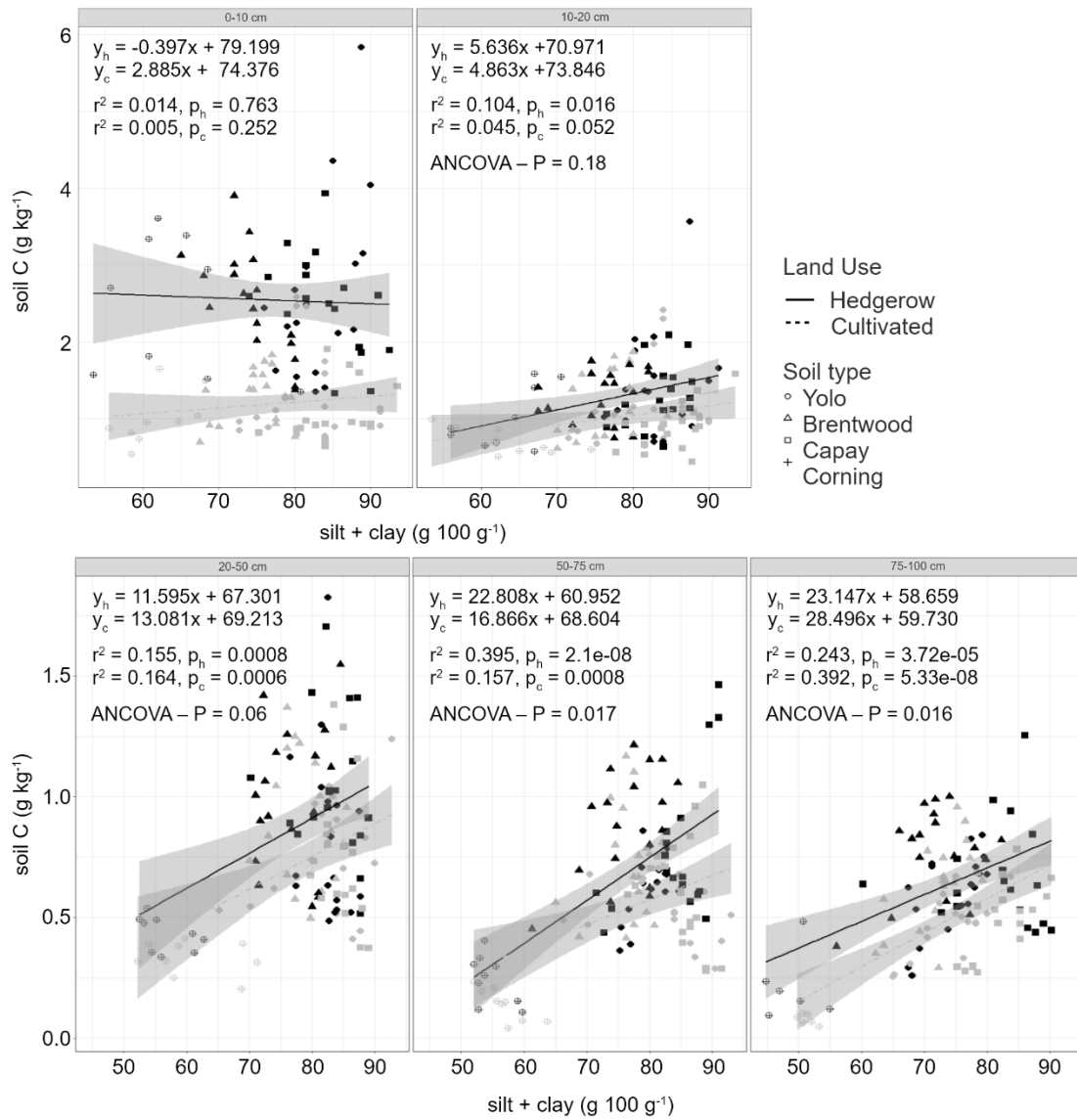


Figure 2.5. Correlations between carbon concentration (g kg⁻¹) and silt + clay content (g 100g⁻¹ soil) for each management type and sampling depth. P-values represent ANOVA's for each treatment individually. Analysis of covariance (ANCOVA) indicated significant differences in the relationship between carbon concentrations and silt + clay content by management type at 50-75 and 75-100 cm.

Silt + clay content (%) was not correlated with soil OC in the surface 0-20 cm (Figure 2.5), likely due to the similarity of management and climate across sites and the dominant effects of these factors in near surface horizons. However, there was a weak to moderate correlation between silt + clay content and soil C at 20-50, 50-75, and 75-100 cm (Figure 2.5). Corning, which had significantly lower silt+clay content (higher sand content) in surface 0-50 cm relative to all other soils, consistently had the lowest soil C concentrations and stocks. Silt + clay was a better predictor of soil C than clay alone, as other studies have similarly shown (Hassink et al. 1997; Rasmussen 2018; Matus 2021). This is consistent with findings that both particle size fractions play key roles in stabilization of soil C (Sollins et al. 2006; von Lutzow et al. 2006; Wiesmeier et al. 2019) and aggregate formation (Six et al. 2002; Totsche et al. 2018).

Bulk density was significantly different by soil type at all depths (Table 2.3). Capay and Corning were denser relative to Yolo and Brentwood (which did not differ from each other). Bulk density was not significantly different by management type on any soil types or at any depth, except at 0-10 cm in Corning, where cultivated fields are significantly denser than hedgerows. Although few studies have evaluated bulk density under hedgerows, lower bulk densities have been reported relative to cultivated fields at 0-50 cm (in 10 cm increments) (Holden et al. 2019) and 0-20 and 20-40 cm (Thiel et al. 2015). In our study, fields were recently tilled, which can reduce BD by 10% or more (Onstad et al. 1984), potentially minimizing differences in the surface 0-20 cm of some soils. Spearman's correlations (SI Table 2.1) showed a strong negative relationship between soil C and bulk density in the surface 0-20 cm on both management types ($p < 0.001$).

pH was unaffected by management on most soil types (Table 2.2), except Corning, which had significantly lower pH under cultivated fields than hedgerows at 20-50, 50-75, and 75-100

cm depths. pH varied significantly by soil type, following similar trends as clay content, with Yolo and Brentwood not significantly different at most depths, but with differences across all other soil types. There was a positive relationship between pH and soil C across depths and management types (SI Table 2.1).

2.4.6 Regressions of Surface vs. Subsurface Soil Carbon

To accurately estimate soil C stock changes, it is necessary to sample to a sufficient depth (Harrison et al. 2011; Poeplau & Don 2015; Blanco-Canqui et al. 2021) and measure both the mass of C and the density (or mass per unit volume) of each sampling depth (Post et al. 2001; VandenBygaart et al. 2006; Poeplau et al. 2017). According to two recent meta-analyses, the average sampling depth in peer-reviewed assessments is only 20-25cm and over 50% of studies fail to report bulk density data (Aguilera et al. 2013, Haddaway et al. 2016). To examine whether surface soil C concentrations are a sufficient proxy for determining quantitative SOC stock change, we used linear regression to compare surface SOC concentrations at a depth of 0-20 cm with whole profile stocks at a depth of 0-100 cm (Figure 2.6). Surface soil C concentrations (0-20 cm) (g kg^{-1}) were weakly correlated with whole profile C stocks (kg m^{-2}) in hedgerows ($F = 21.61$, $df = 61$, $P < 0.001$, $R^2 = 0.38$) and moderately correlated in cultivated fields ($F = 71.70$, $df = 61$, $P < 0.001$, $R^2 = 0.53$). The relationship between surface carbon concentrations (g kg^{-1}) and whole profile carbon (kg m^{-2}) significantly depended on management type (ANCOVA, $F = 5.65$, $df = 122$, $P < 0.02$). While surface carbon concentrations increased with whole profile carbon stocks at both locations, the slope of the relationship in the cultivated fields (0.25) was 175% greater than the slope of the relationship in the hedgerows (0.09) (Figure 2.6).

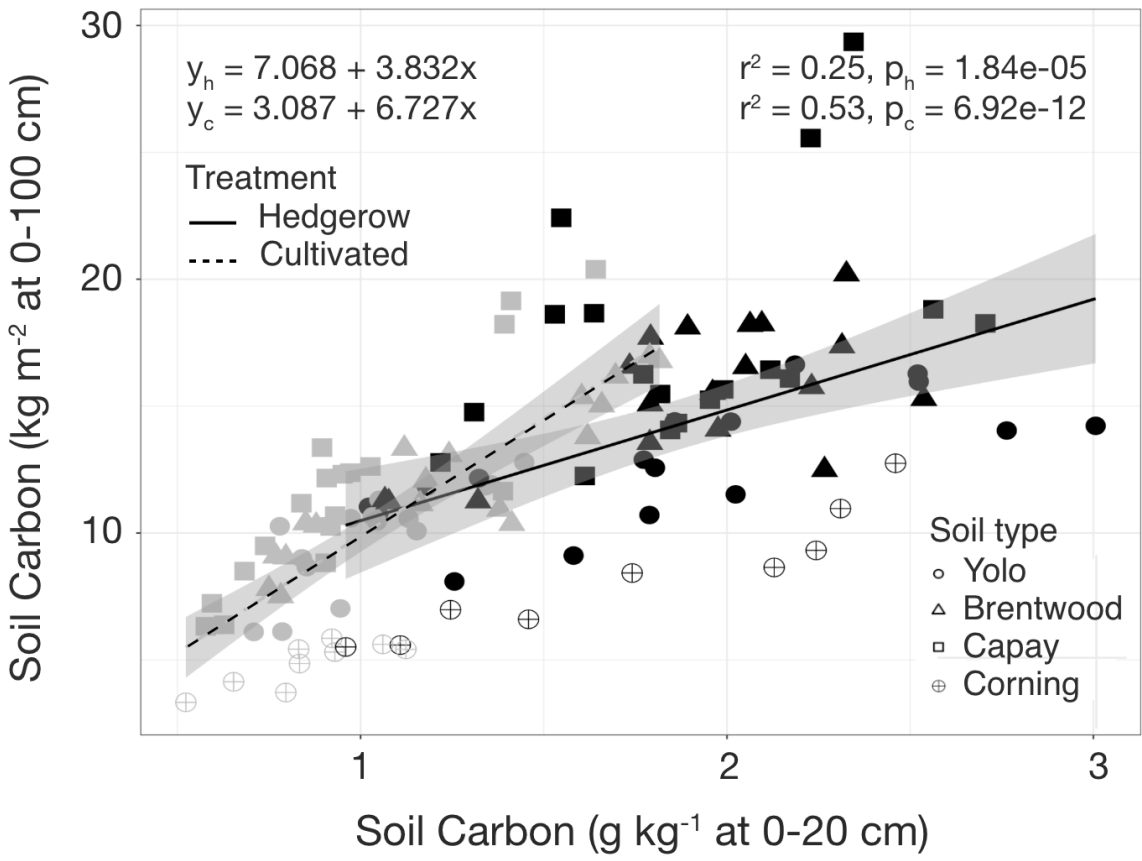


Figure 2.6. Correlation between surface carbon concentration (g kg⁻¹) at 0-20 cm depth and whole profile stocks (kg m⁻²) at 0-100 cm depth. Carbon concentrations at 0-20 cm represent an average of depths sampled at 0-10 cm and 0-20 cm. P-values represent ANOVA's for each treatment individually. Analysis of covariance (ANCOVA) indicated significant differences in the relationship between surface carbon concentrations and whole profile stocks by management type at P=0.02.

Since soil C at 0-20 cm accounted for 28-52% of total stocks and thus, has a strong influence on the values along the y-axis in our regressions, we also assessed the potential of surface C concentrations to predict C concentrations at 20-100 cm (g kg⁻¹) (SI Figure 2.2). Surface C concentrations were weakly correlated in both management types, but the slope of the relationship was once again greater (198%) in cultivated fields than hedgerows. Subsurface soil

C in cultivated fields may be driven largely by surface soil C inputs (residues, compost/manure, etc.), whereas in hedgerows, deposition of leaf litter may lead to faster accrual of soil C in surface than subsurface. Hedgerows may also be more heterogeneous at depth than in cultivated fields, due to perennial and deeper root systems and lack of mixing from tillage.

Several other studies have corroborated our findings that surface soil C concentrations are not good predictors of soil C at depth (Chabbi et al., 2009; Harrison et al., 2011; Jandl et al., 2014; Dal Ferro et al. 2020). Fresh organic inputs have been found to instigate a priming effect, or the mineralization of deep soil C (Fontaine et al. 2007; Wang et al. 2014; Bernal et al. 2016; Callesen et al. 2016; Shahzad et al. 2018). Conversely, surface inputs and deep roots have promoted increases of SOC at depth (Shi et al. 2013; Zhou et al. 2017; Cardinael et al. 2018; Tautges et al. 2019) and buried A horizons are common in aggregating landscapes, such as alluvial fans (Chaopricha & Marin-Spiotta, 2014). In this study, we found that, depending on soil type, 47 to 66% of C was below 20 cm in hedgerows and 49-75% under cultivated fields, which could lead to wide ranges in total stock change estimations using shallow sampling.

2.4.7 Principal Component Analyses

Ordination with PCA was performed to further examine the relationships between soil carbon, physiochemical properties, and variables from soil pit descriptions (Figure 2.7; SI Table 2.2). Three PCs accounted for a fairly high degree of variation in the data, for the surface (0-20 cm) and subsurface (20-100 cm) depths (72% and 85%, respectively). Cultivated and hedgerow soils formed distinct clusters along the x-axes of the biplots (PC1) and, for each management type, sites with the same soil type tended to group together along the y-axes (PC2) for both surface (0-20 cm) and subsurface (20-100 cm) depths. Vectors along the x-axes were positively

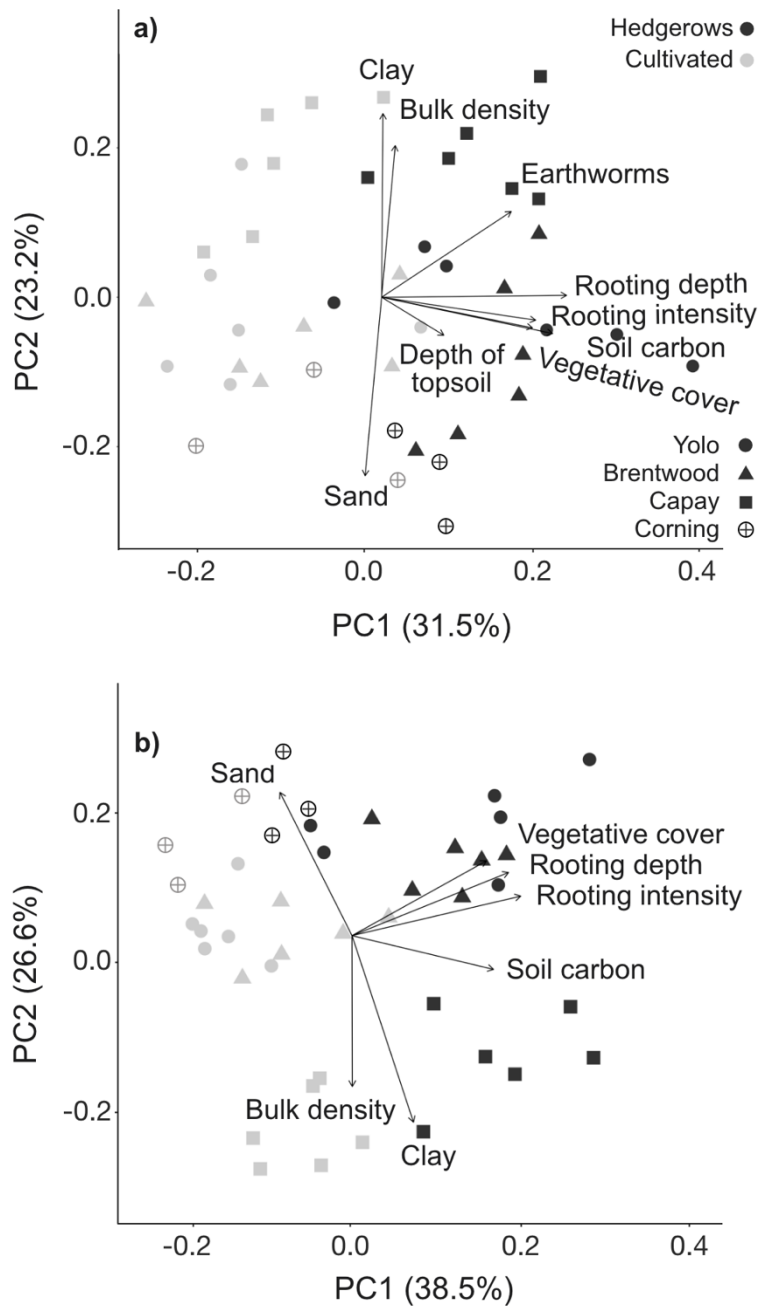


Figure 2.7. Principal Component Analysis biplots of soil physical properties and biological characteristics from soil pit descriptions in a) Surface (0-20 cm) and b) Subsurface (20-100 cm). Weighted averages of measured data collected at 0-10 cm and 10-20 cm depths were used for 0-20 cm values and from samples collected at 20-50 cm, 50-75 cm, and 75-100 cm for 20-100 cm values. Variable units described in the Materials and Methods and in Table 2 in the Supplementary Information.

associated with hedgerows, as compared with cultivated soils; represented by soil C and biological and morphological characteristics from soil pit descriptions. This supports our assumptions that roots and increased vegetative cover (and the associated increases in litter deposition) contribute to the differences in soil C under hedgerows. Topsoil depth and bulk density did not vary much between management types, potentially due to legacy effects of historic land use that have not yet been overcome through revegetation, and accordingly, explained the least amount of the variation at 0-20 cm.

At 0-20 cm, the variables most highly associated with PC1 (31.5% of the variation explained) were rooting depth > vegetative cover > root intensity > soil C > earthworm occurrence, all of which were positively associated with hedgerows (SI Table 2.3). PC2 had high positive loadings for bulk density and clay content and high negative loadings for sand content, representing 23.2% of the total variance. The clusters for Yolo soils are spread broadly across Axis 1 at surface 0-20 cm, reflecting their relatively larger variation in soil C as compared to the other soil types (CV = 44 for hedgerows and cultivated fields, 50 to 70% higher than other soil types). Physical properties, such as soil texture and bulk density, to a lesser extent, were highly associated with Axis 2 (23.2% of the variation).

Similar patterns occurred for the lower depths (20-100 cm) except for earthworm presence (Figure 2.7), which was not included in the analysis due to lack of presence below 20 cm at most sites. Differentiation across the y-axis by soil type is more distinct at 20-100 cm, corroborating the increased effect of soil type at lower depths in our study and others. PC1 accounted for 38.5% of the variance, with high positive loadings for root intensity > rooting depth > soil C > vegetative Cover (SI Table 2.3). PC2 represented 26.6% of the total variance and had high positive loadings for sand, vegetative cover and rooting depth and high negative

loadings for clay and bulk density. Capay soils were negatively associated with PC1 and PC2 indicating that the high clay contents were restrictive of root growth and C storage in cultivated soils. A two-dimensional visualization of the first two components shows strong differentiation by management types across the x-axis at both depths. Soil types tend to cluster across the y-axis, especially at 20-100 cm.

At 0-20 cm, the increased time in vegetative cover, intensive root systems, and increased soil C of hedgerow shrubs explained much of the variation in the data. Vegetative cover, rooting depth and intensity, and soil C also explained much of the variation at 20-100 cm, but inherent soil properties contributed more to variation and there was less distinction between management type than in the surface 0-20 cm.

2.5 Summary and Conclusions

Our results demonstrate that restoration of field edges with hedgerows has a pronounced impact on soil C storage with significant differences extending throughout the profile to a depth of 100 cm. The differential between hedgerows and cultivated fields was similar across soil types, indicating that hedgerows may have broad potential, although further investigation on soils with more diverse mineralogy, initial soil C content, and soil temperature/moisture regimes is necessary. Although farm edges do not make up a substantial proportion of total farm area, hedgerows provide a climate mitigation strategy with increased permanence, less leakage and additionality concerns. If implemented at scale, hedgerows could contribute to a small portion of GHG reduction goals, while promoting biodiversity, providing critical habitat in increasingly fragmented agricultural landscapes, and supporting a host of field and landscape scale co-benefits.

Increases in soil C concentrations in the surface 0-20 cm contribute greatly to climate change adaptation and increased resilience, but they are not an effective proxy for subsurface concentrations or whole profile C stocks. Policymakers and ecosystem markets should implement deeper sampling protocols, potentially through a national network of monitoring sites, representing major cropping systems and agriculturally relevant climates and soil types, to minimize costs and maximize the applicability of the data collected.

Management type had the strongest effect on soil C in the surface 0-10 cm and from 50-100 cm, with the perennial cover and root systems of the hedgerows explaining much of the variation in soil C. At 20-100 cm, inherent soil properties/soil type contributed more to variation in soil C than in the surface 0-20 cm. Soil type should always be considered in sampling design and model development, especially in regard to soils formed from different parent materials with different mineralogies, varying degrees of weathering, and contrasting climates.

Further research is needed 1) to identify appropriate hedgerow species for various contexts, 2) to better characterize contributions of above and below-ground woody C stocks (and relationships with tree/shrub dimensions), and 3) to investigate the impact on SOC at varying distances from the hedgerow to see if gains extend into field or are offset by losses, due to light interception or competition between tree/shrub roots and crops.

2.6 Acknowledgements

I would like to acknowledge Rachael Long for sharing her deep knowledge and understanding of hedgerows in Yolo County and for all the great research she has contributed as to the many co-benefits they provide – and for helping us track down a backhoe when we couldn't quite reach 100 cm. To all the incredible Yolo County growers who were kind enough

to let us come out and dig holes in the middle of their hedgerows and fields and for sharing information on your past management and intentions/experience with hedgerows. To the late John Anderson and Hedgerow Farms for all your work to increase hedgerow adoption across Yolo County and for making all of this work possible. To Garrett Long for your organizational and physical support in the field from the first to the last day of sampling – and for making sure we didn't all dehydrate. To Emily Lovell, Gavin Chaboya, and Jacob Grey for providing some critical muscle in the field and all your dedicated work in the lab to support sample processing and analysis. To Jake Spertus and Irfan Ainuddin for their support with statistics and R.

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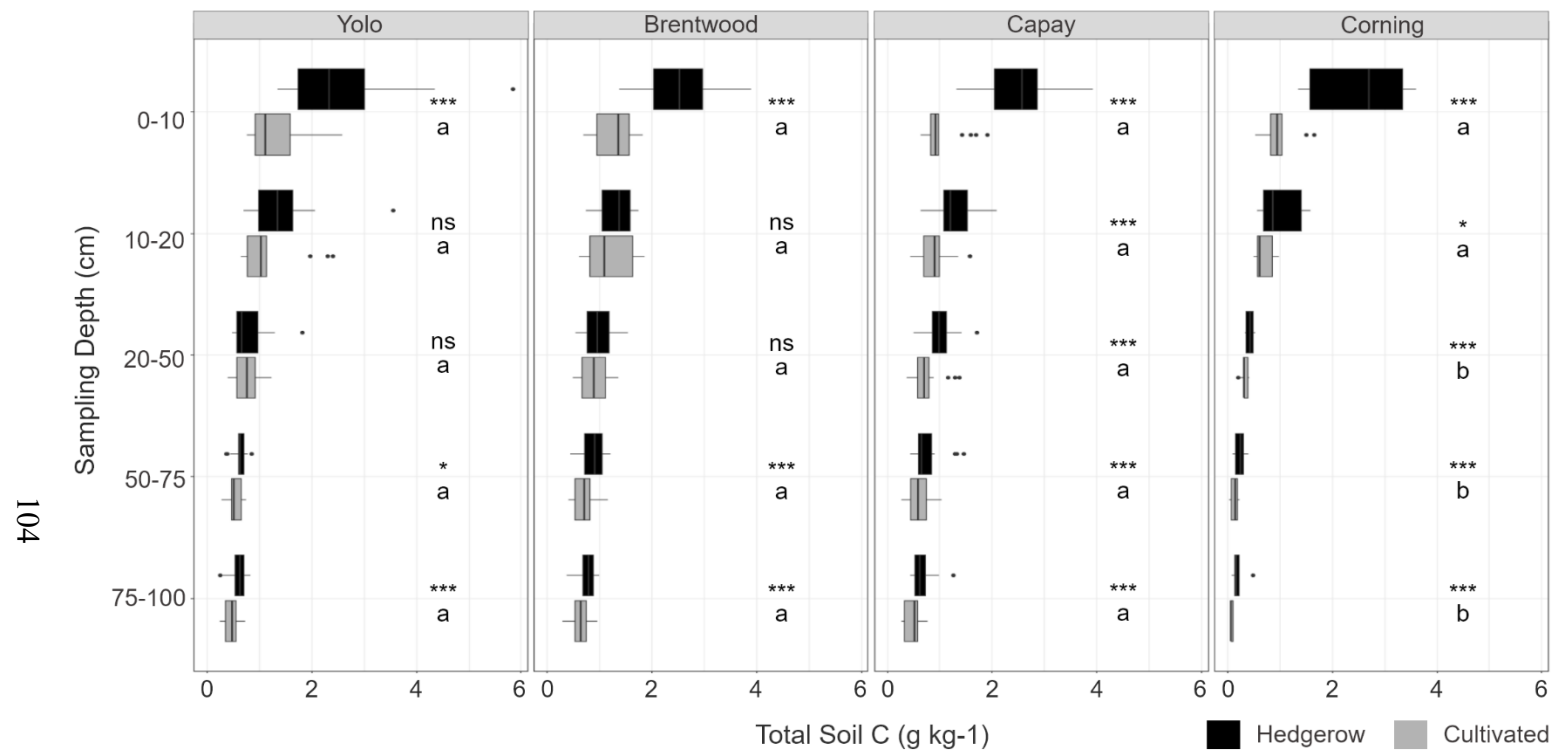
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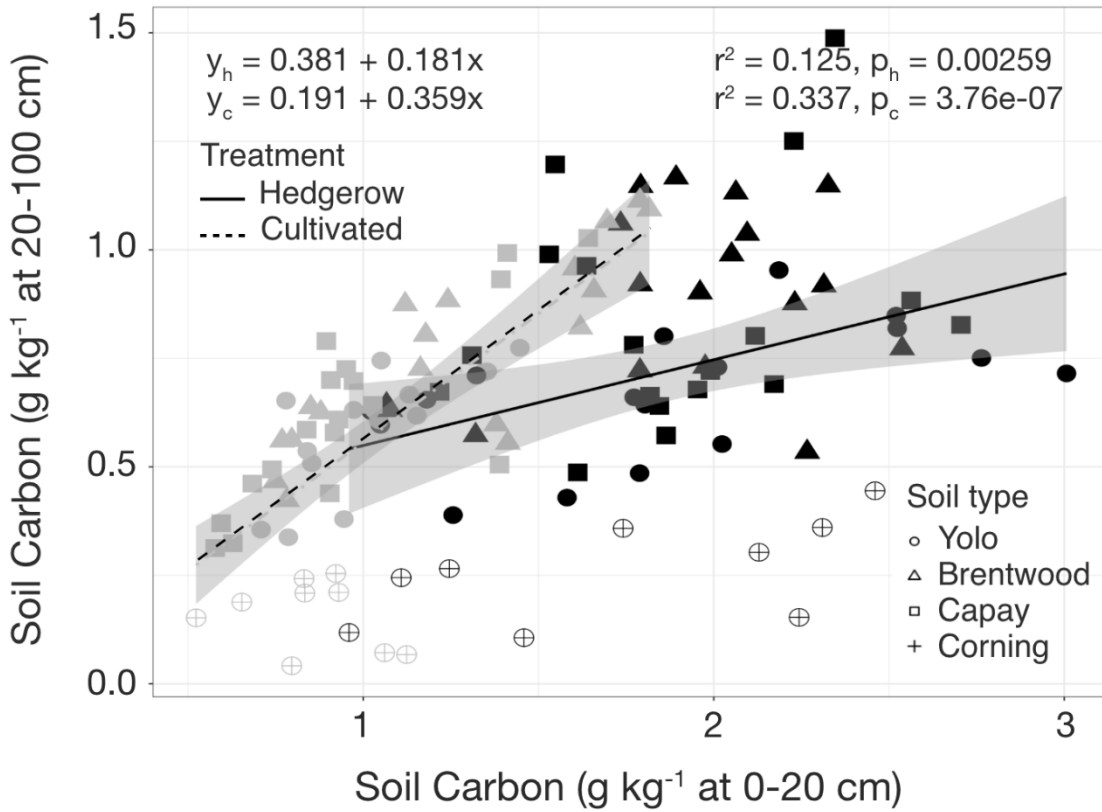
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2.8 Supplemental Information



SI Figure 2.1. Box plots of soil carbon concentration by management type and depth for each soil type. Letters indicate significant differences between soil types and asterisks indicate significant differences by management type in Tukey means comparisons at $P < 0.05$ ($n=21$ for hedgerow, $n=21$ for cultivated fields, $n=6$ for each soil type, except Corning $n=3$). Vertical line in each box represents the mean and the left and right sides of the boxes represent the first and third quartiles, respectively. The whiskers extend to the highest and lowest values within 1.5 times the inter-quartile range and points outside of the “box and whiskers” represent outliers.



SI Figure 2.2. Correlation between surface carbon concentration (g kg⁻¹) at 0-20 cm depth and subsurface carbon concentration (g kg⁻¹) at 20-100 cm depth. P-values represent ANOVA's for each treatment individually. ANCOVA indicated significant differences in the relationship between surface carbon concentrations and subsurface carbon by management type at P = 0.041.

SI Table 2.1. Total soil nitrogen concentrations and carbon-to-nitrogen ratios to a depth of 1m by soil type and management type for 21 sites in Yolo County, California.

Soil Type	Total Soil Nitrogen (g kg ⁻¹)						Soil C:N Ratio					
	Hedgerow		Cultivated				Hedgerow		Cultivated			
	0-10 cm depth											
Yolo	0.26	(0.03)	0.15	(0.01)	***	a	10.09	(0.28)	9.06	(0.46)	*	a
Brentwood	0.22	(0.01)	0.13	(0.01)	***	ab	11.42	(0.30)	9.67	(0.24)	***	b
Capay	0.22	(0.01)	0.11	(0.01)	***	b	11.46	(0.30)	9.52	(0.26)	***	b
Corning	0.21	(0.02)	0.09	(0.01)	***	b	11.83	(0.22)	10.58	(0.33)	**	b
	10-20 cm depth											
Yolo	0.16	(0.01)	0.14	(0.01)	ns	a	9.01	(0.40)	8.48	(0.38)	ns	a
Brentwood	0.12	(0.01)	0.12	(0.01)	ns	ab	10.94	(0.27)	9.78	(0.3)	***	b
Capay	0.13	(0.01)	0.10	(0.01)	***	bc	10.17	(0.23)	9.25	(0.24)	**	ab
Corning	0.09	(0.01)	0.08	(0.01)	ns	c	10.69	(0.49)	9.29	(0.49)	**	ab
	20-50 cm depth											
Yolo	0.09	(0.01)	0.09	(0.01)	ns	a	9.22	(0.22)	8.60	(0.33)	ns	a
Brentwood	0.09	(0.01)	0.09	(0.01)	ns	a	10.61	(0.26)	10.30	(0.34)	ns	b
Capay	0.10	(0.01)	0.09	(0.01)	*	a	10.03	(0.37)	8.59	(0.32)	**	c
Corning	0.05	(0.01)	0.04	(0.01)	*	b	9.42	(0.72)	8.42	(0.54)	ns	ac
	50-75 cm depth											
Yolo	0.09	(0.01)	0.08	(0.01)	**	a	7.50	(0.47)	7.52	(0.42)	ns	a
Brentwood	0.08	(0.00)	0.07	(0.00)	**	a	10.70	(0.30)	10.06	(0.30)	ns	b
Capay	0.08	(0.01)	0.07	(0.01)	**	a	9.36	(0.56)	8.89	(0.49)	ns	b
Corning	0.03	(0.00)	0.02	(0.00)	***	b	7.41	(0.80)	10.33	(1.04)	*	b
	75-100 cm depth											
Yolo	0.08	(0.01)	0.06	(0.00)	**	a	7.82	(0.64)	7.66	(0.58)	ns	a
Brentwood	0.08	(0.00)	0.06	(0.00)	**	a	10.05	(0.32)	9.74	(0.36)	ns	b
Capay	0.08	(0.01)	0.06	(0.01)	**	a	9.22	(0.60)	8.36	(0.62)	ns	ab
Corning	0.02	(0.01)	0.01	(0.00)	*	b	8.94	(0.36)	9.20	(0.45)	ns	ab

Within a column, for each depth, soil types followed by the same letter are not significantly different at P< 0.05. Asterisks indicate significant difference between management treatments (* = P< 0.05, ** = P< 0.01, *** = P< 0.001). Numbers in parentheses indicate standard error (n = 18); for Corning (n=9)

SI Table 2.2. Spearman's correlation coefficients between soil properties and management practices in surface (0-20cm) and subsurface (20-100 cm). Asterisks represent a significant correlation between variables at * = P < 0.05, ** = P < 0.01, and *** = P < 0.001.

Depth	SOC (g kg ⁻¹)		TN (g kg ⁻¹)		Silt (g kg ⁻¹)		Clay (g kg ⁻¹)		BD (g cm ⁻³)		pH		C:N Ratio	
	H	C	H	C	H	C	H	C	H	C	H	C	H	C
0-20 cm														
TN (g kg ⁻¹)	0.94 ***	0.89 ***	-	-	-	-	-	-	-	-	-	-	-	-
Silt (g 100 g ⁻¹)	-0.01	0.15	0.08	0.23 **	-	-	-	-	-	-	-	-	-	-
Clay (g 100 g ⁻¹)	0.05	0.18	0.01	0.2 *	-0.64 ***	-0.69 ***	-	-	-	-	-	-	-	-
BD (g cm ⁻³)	-0.54 ***	-0.29 ***	-0.48 ***	-0.27 ***	-0.29 ***	-0.17 *	0.38 ***	0.19 *	-	-	-	-	-	-
pH	0.19 **	0.39 ***	0.28 ***	0.45 ***	0.17	0.14	0.24 **	0.23 **	0.07	-0.11	-	-	-	-
C:N Ratio	0.45 ***	0.51 ***	0.19 *	0.13 *	-0.29 ***	-0.02	0.05	-0.02	-0.34	-0.17	-0.18 *	-0.02	-	-
20-100 cm														
TN (g kg ⁻¹)	0.82 ***	0.85 ***	-	-	-	-	-	-	-	-	-	-	-	-
Silt (g 100 g ⁻¹)	0.24 ***	0.32 ***	0.3 ***	0.37 ***	-	-	-	-	-	-	-	-	-	-
Clay (g 100 g ⁻¹)	0.27 ***	0.18 ***	0.16 *	0.13	-0.57 ***	-0.63 ***	-	-	-	-	-	-	-	-
BD (g cm ⁻³)	-0.17 **	-0.11	-0.24 ***	-0.11	0.62 ***	-0.53 ***	0.61 ***	0.6 ***	-	-	-	-	-	-
pH	0.36 ***	.21 ***	0.41 ***	0.24 ***	0.02	-0.04	0.52 ***	0.49 ***	0.25 ***	0.4 ***	-	-	-	-
C:N Ratio	0.47 ***	0.39 ***	-0.08	-0.08	0.04	-0.06	0.22 ***	0.06	0.04	-0.1	0.01	-0.18 *	-	-

SI Table 2.3. Input data for Principal Component Analysis

	Hedgerow		Cultivated	
	mean	se	mean	se
Soil carbon (g kg ⁻¹ at 0-20 cm)				
Yolo	2.15	0.56	1.27	0.25
Brentwood	1.85	0.18	1.27	0.17
Capay	1.87	0.19	0.96	0.15
Corning	1.87	0.32	0.76	0.05
Soil carbon (g kg ⁻¹ at 20-100 cm)				
Yolo	0.71	0.11	0.57	0.05
Brentwood	0.91	0.09	0.73	0.09
Capay	0.87	0.09	0.61	0.10
Corning	0.24	0.05	0.16	0.06
Root intensity (0-20 cm)				
Yolo	1.16	0.35	0.54	0.06
Brentwood	1.34	0.16	0.52	0.09
Capay	0.98	0.12	0.55	0.11
Corning	1.03	0.18	0.97	0.16
Root intensity (20-100 cm)				
Yolo	3.15	0.70	0.29	0.03
Brentwood	1.72	0.41	0.52	0.05
Capay	3.01	0.64	0.23	0.05
Corning	0.79	0.03	0.18	0.06
Root depth (cm)				
Yolo	59.67	7.54	28.17	3.10
Brentwood	63.50	2.55	32.67	8.58
Capay	57.50	2.08	36.33	3.03
Corning	55.00	5.57	34.67	5.46
Vegetative cover (months)				
Yolo	12.00	0.00	6.42	0.82
Brentwood	12.00	0.00	8.50	0.71
Capay	12.00	0.00	8.00	0.32
Corning	12.00	0.00	9.67	1.20
Topsoil depth (cm)				
Yolo	11.67	1.69	13.33	2.06
Brentwood	12.17	1.30	11.83	0.75
Capay	11.00	0.73	11.33	0.92
Corning	10.00	2.08	11.67	2.33
Earthworm index				
Yolo	3.33	0.33	2.17	0.48
Brentwood	2.50	0.43	2.50	0.43
Capay	3.17	0.31	2.33	0.33
Corning	2.00	0.58	2.00	0.58

SI Table 2.4. Summary results from the first three principal components of a principal component analysis (PCA) of soil biological indicators collected from soil pit descriptions (n=42). Weighted averages were calculated for 0-20 cm and 20-100 cm depths. Analysis was conducted on all sites without separation by soil type. Units provided in Table 2 in Supplementary Information.

	0-20 cm depth			20-100 cm depth			
	PC1	PC2	PC3	PC1	PC2	PC3	
Eigenvalues	1.68	1.45	1.23	Eigenvalues	1.64	1.36	1.19
% Variance	31.52	23.21	16.82	% Variance	38.52	26.61	20.14
Cumulative % Variance	31.52	54.73	71.55	Cumulative % Variance	38.52	65.13	85.27
Factor Loading				Factor Loading			
Rooting Depth	0.513	0.051	-0.177	Root Intensity	0.533	0.131	0.014
Vegetative Cover	0.478	-0.077	-0.216	Rooting Depth	0.495	0.210	-0.224
Root Intensity	0.433	-0.034	-0.351	Soil Carbon	0.447	-0.112	0.455
Soil Carbon	0.427	-0.062	0.401	Vegetative Cover	0.425	0.249	-0.388
Earthworms	0.325	0.248	0.223	Clay	0.194	-0.621	-0.224
Topsoil Depth	0.179	-0.114	0.558	Bulk Density	0.001	-0.502	-0.560
Bulk Density	0.003	0.485	-0.411	Sand	-0.228	0.477	-0.477
Clay	-0.033	0.588	0.043	Topsoil Depth	NA	NA	NA
Sand	-0.016	-0.575	-0.326	Earthworms	NA	NA	NA

Chapter 3: Hedging Our Bets – Assessing Long-Term Impacts of Afforestation on Soil Health

3.1 Abstract

Government and industry have begun incentivizing farmers for practices thought to improve soil health, as an approach to mitigating and adapting to climate change, while ensuring the long-term sustainability and viability of agriculture. No clear threshold values exist, however, to achieve soil health, especially in response to climate change, cropping system management, and soil type. Using a historical planting of hedgerows established in the mid-1990s, we assessed the impact of afforestation on commonly used soil health indicators (0-20 cm) across four soil types in Yolo County, CA. Hedgerows satisfy many of the key principles of soil health management, including continuous ground cover/roots, reduced disturbance (tillage), and increased diversity. By comparing soils under long-term hedgerows relative to the adjacent cultivated field, this study explores the extent to which agricultural soils respond to a common soil health promoting practice. Sampling included biological (microbial biomass carbon (MBC) and nitrogen (MBN), permanganate-oxidizable C, C- and N- cycling enzymes), chemical (pH, total C and N, KCl extractable C and N), and physical (bulk density, infiltration rate, surface/subsurface hardness, aggregate stability) variables. At 0-10 cm, soil C, available C, MBC and MBN, enzyme potential activity, and aggregate stability were two times higher under hedgerows, relative to adjacent cropland, while infiltration rates were at least two times faster. At 10-20 cm, only soil C, MBC, C-cycling enzymes, and surface hardness were higher under hedgerows. While some metrics were more sensitive than others (i.e. total C, MBC, MWD), a composite of biological, chemical, and physical indicators was necessary to explain the variation in the data.

3.2 Introduction

Agriculture is under increasing pressure to feed a growing population, on less land, with less environmental externalities, and amidst a changing climate (Foley et al. 2011; Pittelkow et al. 2014; Smith et al. 2016). The UN FAO International Year of Soils and the NRCS's Soil Health Division have spurred widespread interest in soil health management (FAO 2015; USDA–NRCS, 2018; Karlen et al. 2019), garnering bipartisan support and the attention of multiple stakeholders, including industry and the general public. Soil health management is a no-regrets solution that achieves the triple bottom line of sustainability – promoting people, planet, and profit (Elkington 1994; Carreon et al. 2011) – and importantly, resonates with growers (Andrews et al. 2004, Carlisle 2016; Roesch-McNally et al. 2017). Soil health is commonly defined as “the capacity of a soil to function within ecosystem and land-use boundaries to sustain biological productivity, maintain environmental quality, and promote plant and animal health” (Doran and Parkin, 1994; Kibblewhite et al. 2007). It is considered an emergent property that lies at the intersection of the physical, chemical, and biological characteristics of a given soil (Karlen et al. 1997; Kibblewhite et al. 2007; Bunemann et al. 2018). Thus, soil health is dynamic in nature, making it responsive to changes in land use/management, but also challenging to measure (Doran and Jones, 1996; Moebius-Clune et al. 2007; Lehmann et al. 2020).

Analogous to human health, no one metric can capture the health of the soil system (Kibblewhite et al. 2008; Baveye et al. 2016). Several attempts have been made to develop minimum viable data sets that encompass a broad suite of biological, chemical, and physical indicators (Andrews et al. 2003; Morrow et al. 2016; Stott et al. 2019; Norris et al. 2020; Nunes et al. 2020). Effective indicators must be accessible (logistically and economically), accurate, reliable, sensitive to management, and have relatively low spatial and temporal variability

(Karlen et al., 2006; Idowu et al., 2009; Morrow et al. 2016; Hargreaves et al. 2019). They must also be interpretable and useful in informing on-farm, adaptive management.

While there is a fairly strong consensus as to the physicochemical properties to include in soil health assessments (i.e. texture, aggregate stability, bulk density, pH, and organic carbon) (Doran and Parkin 1996; Cardoso et al. 2013; Bunemann et al. 2018; Stewart et al. 2018), biological and biochemical indicators are still poorly understood and frequently underrepresented (Stott et al. 2019; Lehmann et al. 2020; Fierer et al. 2021). The inclusion of biological and biochemical indicators is critical as soil organisms are central to many soil processes (i.e. decomposition, nutrient cycling, and aggregate formation) and the provision of ecosystem services (i.e. plant growth promotion, carbon (C) sequestration, pest/pathogen resistance, and drought resilience) (Bach et al., 2020; Fierer et al. 2021; Lehmann et al. 2020). Indicators for measuring the biological community and biological activity, however, are arguably some of the most spatiotemporally variable; influenced strongly by soil texture, pH, temperature, and moisture content (Hurriso et al. 2018; Wade et al. 2018; Amsili et al. 2021; Fierer et al. 2021; Lazicki et al. 2021). Thus, while many biological/biochemical indicators are sensitive to management (Barrios, 2007; Bastida et al. 2008; Kibblewhite et al. 2008; Lazicki et al. 2021), they may not be sufficient in isolation and should be measured alongside a suite of other indicators to aid in interpretation (Griffiths et al., 2016; Bunemann et al. 2018).

The most appropriate set of indicators, ultimately, will vary by context (i.e. region, cropping system, soil type) and management goals (Andrews et al. 2004; Moebius-Clune et al. 2007; Fierer et al. 2021). There is also a pressing need to contextualize thresholds of indicator responses by edaphoclimatic properties (Fine et al. 2017; Nunes et al. 2020; Devine and O'Geen 2021). While there has been much research on soil health management and the associated

outcomes in the Midwest, there is a paucity of data in California and other Mediterranean-type climates (Lagacherie et al. 2018; Devine and O’Geen 2021). California’s high level of agricultural productivity often associated with intensive tillage, prolonged bare fallow, and high input use have resulted in environmental challenges including nitrate leaching, groundwater overdraft, and loss of soil organic matter (Jackson et al. 2003; Mitchell et al., 2012; Harter, 2015; Tautges et al. 2019).

Long-term trials have highlighted the potential for soil health management including reduced tillage (Madden et al. 2008; van Donk, 2010; Klocke et al., 2010; Mitchell et al., 2012), cover cropping (Poudel et al. 2002, Seiter and Horwath, 2004, Mitchell et al., 2015, Jahanzad et al., 2016), and organic management (cover crop + compost) (Kong et al. 2011, Li et al. 2019; Tautges et al. 2019) to improve agronomic and environmental outcomes for the state. Rarely, however, has there been an opportunity to assess the stacking of all four soil health principles (keep soil covered, maintain roots in the ground, promote diversity, and reduce disturbance) on a regional/cropping system/soil type basis (Kibblewhite et al. 2008). While differences in land use inform variations in desired outcomes on a farm (i.e. production and non-production areas), undisturbed/unmanaged areas can clarify or put bounds on the *capability* of a given soil to achieve a healthy condition (Brown and Herrick 2016; McBratney et al. 2019; Maharjan et al. 2020; Williams et al. 2020).

Hedgerows satisfy many of the key goals of soil health management (Kibblewhite et al. 2008; Long et al. 2010; Heath et al. 2017; Holden et al. 2019). By providing additional ground cover, hedgerows have been found to buffer soil temperatures (Clinch et al., 2009; Sanchez et al. 2010; Dubbert et al., 2014) and protect against erosion and runoff (Walter et al. 2003; Long et al. 2010; van Vooren et al. 2017). By maintaining roots in the ground, hedgerows have been shown

to increase infiltration and soil C (Ghazavi et al. 2008; Thiel et al. 2015; Viaud & Kunnemann 2021), increase saturated hydraulic conductivity and soil water storage (Marshall and Moonen 2002; Holden et al. 2019) and reduce nitrate leaching and runoff (Caubel et al. 2003; Long et al. 2010; van Vooren et al. 2017; Thomas and Abbot 2018). Hedgerows increase botanical diversity, serve as dispersal corridors for wildlife, and support organisms that provide critical ecosystem services to agriculture (Marshall and Moonen 2002; Ouin and Burel 2002; Long 2010). In California agricultural production areas, hedgerows have been shown to increase the prevalence of birds, beneficial insects, and native bees, relative to bare or weedy field edges, which attracted more pests than hedgerows (Vickery et al. 2004; Morandin 2011, 2013; Heath et al. 2017).

Historic multi-stakeholder efforts established hedgerows on field edges across Yolo County, California. This provided an opportunity to evaluate the long-term effects of hedgerow plantings on common indicators of soil health across soil types, but within the context of a specific climate, region, and similar cropping systems, *i.e.*, intensive row crop agriculture in a Mediterranean-type climate. To characterize differences between hedgerows and adjacent cultivated fields, we analyzed a broad suite of lab-based and in-field indicators commonly proposed to underpin soil health and function (Moeibus-Clune et al. 2016, Fine et al. 2017, Bunemman et al. 2018; Norris et al. 2020) at 20 sites across the county. Specifically, our objectives were to 1) compare soil health metrics in the surface 0-20 cm under hedgerows and cultivated fields at paired sites across Yolo County, 2) assess the impact of soil type on differences in soil health metrics between hedgerows and cultivated fields, 3) determine metrics that are most sensitive to hedgerows vs. cultivated management, and 4) evaluate relationships between biological and physiochemical indicators of soil health.

3.3 Material and methods

3.3.1 Site Description

This regional survey was conducted in the flat, lowland alluvial plains, fans, and terraces of Yolo County (Figure 2.1), situated in the southern Sacramento Valley, California, USA. Elevation ranged from 16 to 140 m above sea level across survey sites. The region is characterized by cool, wet winters and hot, dry summers; a xeric soil moisture regime (annual precipitation from 40-56 cm) and a thermic soil temperature regime (average annual temperatures of 10–17°C) (Andrews 1972). Soils are developed largely from parent material deposited from the Coast Range to the west.

The study area is situated in the California Floristic Province, one of 36 biodiversity hotspots globally. It was historically dominated by oak woodlands, savannas, and wetlands, until cultivation began approximately 150 years ago. Today, the region is characterized by increasing intensification of irrigated agriculture with crop rotations dominated by wheat (*Triticum aestivum*), processing tomato (*Solanum lycopersicum*), alfalfa hay (*Medicago sativa*), and seed crops such as sunflower and safflower; as well as wine grapes (*Vitis vinifera*), almonds (*Prunus dulcis*), and rice (*Oryza sativa*).

Since the mid-1990's multi-stakeholder collaborations have helped farmers establish hedgerows, filter strips, and vegetated riparian corridors on agricultural lands, including a 2001 campaign to "Bring Farm Edges Back to Life" (Earnshaw et al. 2004; Brodt et al. 2009). Native California plant species were selected for plantings due to drought-tolerance, successive, overlapping bloom periods, and the provision of resources for beneficial and natural predatory insects (Pickett & Bugg, 1998; Long et al., 1998). These efforts have resulted in approximately

175 acres of hedgerow plantings in Yolo County as of 2020 and have supported numerous research projects into the agronomic and environmental benefits of hedgerows in the region (Morandin et al. 2011, 2013, 2016; Long et al. 2010, 2017; Heath et al. 2017; Heath & Long 2019).

3.3.2 Site Selection

A census of extant hedgerows in Yolo County was conducted in collaboration with UC Cooperative Extension. Soil types for each site were identified using Soil Web (Beaudette and O'Geen, 2009). Four soil types that represented a range of common soils in the region and presented a sufficient sample size were selected. Sites visits were then conducted to ground truth soil texture in surface 0-20 cm and identify hedgerows that satisfied the following criteria: 1) greater than or equal to 10 years in age; 2) greater than or equal to 5 feet in height; 3) contiguously planted with woody species; 4) immediately adjacent to a cultivated row or field crop; 5) not situated in or along a waterway or irrigation canal; and 6) where soil had not been reworked, moved, or made into a berm.

Twenty-one paired sites were selected (Figure 2.1); six on Yolo silt loam (*Fine-silty, mixed, superactive, nonacid, thermic Mollic Xerofluvents*), six on Brentwood clay loam (*Fine, smectitic, thermic Typic Haploxerepts*); six on Capay silt clay (*Fine, smectitic, thermic Typic Haploxererts*); and three on Corning loam (*Fine, mixed, semiactive, thermic Typic Palexeralfs*) (Table 3.1).

Table 3.1. Site information for 20 hedgerows and adjacent cultivated fields in Yolo County, California. Management practices represent the typical management for the past 5 years, while crop refers to the current crop at the time of sampling.

Site	Soil Type	Soil Textural Class	Hedgerow Age (yrs)	Compost (tons ha ⁻¹ yr ⁻¹)	Crop	Cover	Fallow Crop (months)
1	Yolo	Silt loam	20	0	wheat	N	6
2	Yolo	Silt loam	23	0	tomato	N	4.5
3	Yolo	Silt loam	11	0	tomato	N	7
4	Yolo	Silt loam	19	0	tomato	N	7
5	Yolo	Loam	10	4	tomato	N	7
6	Brentwood	Clay loam	13	12	diverse	Y	2
7	Brentwood	Clay loam	10	6	tomato	Y	2
8	Brentwood	Clay loam	10	6	tomato	Y	2
9	Brentwood	Clay loam	14	0	tomato	N	4.5
10	Brentwood	Clay loam	16	4	wheat	N	6
11	Brentwood	Clay loam	23	0	tomato	N	4.5
12	Capay	Silty clay	20	0	tomato	N	4.5
13	Capay	Silty clay	20	0	tomato	N	4.5
14	Capay	Silty clay	25	0	rye	N	3
15	Capay	Silty clay	25	0	rye	N	3
16	Capay	Silty clay	20	0	wheat	N	4.5
17	Capay	Silty clay	15	0	wheat	N	4.5
18	Corning	Loam	25	0	poppies	N	3
19	Corning	Loam	10	0	grapes	N	4
20	Corning	Loam	11	0	oat hay	N	2

Yolo = Mollic Xerofluvent, Brentwood = Typic Haploxerept, Capay = Typic Haploxerert, Corning = Typic Palexeralf
¹ diverse = cultivation of more than one species at the same time

Cultivated fields represented similar annual cropping systems but varied in management practices and crops in rotation at time of sampling. One perennial system (vineyard) was sampled to include the Corning series, a soil type with a markedly different texture, pH, mineralogy, and degree of weathering from other soils in the region. Corning soils are less commonly found in the county, especially under irrigated agriculture (Andrews 1972), necessitating the inclusion of a vineyard. Hedgerows consisted of predominantly shrubs with occasional tree species. Commonly occurring species included willow (*Salix spp.*), California

lilac (*Ceanothus spp.*), elderberry (*Sambucus mexicana*), California coffeeberry (*Rhamnus californica tomentella*), toyon (*Heteromeles arbutifolia*), saltbush (*Atriplex lentiformis*), coyote brush (*Baccharis pilularis*), western redbud (*Cercis occidentalis*), and milkweed (*Asclepias spp.*). Hedgerows ranged in age from 10-25 years (mean was 17 years) and were all established with irrigation and fertility in the first three years, although levels of maintenance (*i.e.*, pruning and weeding) may have varied over their lifetime.

3.3.3 Soil Sampling

In April 2019, prior to spring irrigation, soil samples were collected from all 21 sites for total C and N, bulk density (BD), aggregate stability, and particle size determination over the course of 12 days. In-field assessments of infiltration rate and surface hardness were also conducted at this time. On the 13th day, all 21 sites were revisited, collecting samples from undisturbed soils directly adjacent to initial sampling locations and immediately placing them on ice until they could be shipped for microbiological and enzymatic analysis within 24 h. Rapid sampling was conducted to control for variations in soil moisture/temperature and thus, minimize variability in responses to biological/biochemical indicators. Unfortunately, samples from the 21st site were not collected in time to include in overnight shipping and accordingly, were excluded from statistical analyses on all indicators discussed herein.

At each site, three locations were selected along a contiguous 100 m transect within the hedgerow using a random number generator (Figure 2.2). Within the cultivated field, three locations were selected 50-m directly parallel to hedgerow locations to avoid an edge effect and the impact of traffic/equipment, while minimizing variability in inherent soil properties. At the initial sampling, a shovel was used to dig small holes to 20 cm at each location and samples were

collected off the face to avoid disturbance of aggregates, at 0-10 and 10-20 cm, representing agriculturally relevant surface horizons (typical depth of plow layer). Soils from each sampling location were stored separately at 4°C until field sampling was complete. In the central location of each transect, bulk density (BD) samples were collected vertically at the midpoint of each sampling depth interval using a metal core (8.25 cm diameter and 7.5 cm length) (*i.e.*, 1.25 – 8.75 cm and 11.25 – 18.75 cm). On day 13, three samples were collected with a soil probe in the immediate vicinity of each initial sampling location, utilizing sterile technique between each location. Samples were lightly homogenized, placed into two bags, and immediately put on ice. One set of samples was shipped overnight on dry ice to the Soils Lab at University of Illinois, Urbana-Champaign. The second set was immediately air-dried at 25°C, prior to analysis for pH and active, or permanganate-oxidizable, carbon (POXc).

3.3.4 Physical Indicators

At each sampling location, surface residues/litter were gently cleared, taking care not to disturb the soil surface, and a double ring infiltrometer was hammered ~1 inch into the ground. After lining the central ring with plastic sheeting, the outer ring was filled with 2.5 cm of water to control for lateral flow, followed by 2.5 cm of water in the central ring. The plastic liner was removed and time for water to completely infiltrate was recorded in seconds (INF-1”) (Stott et al. 2019). After 15 minutes, the same procedure was repeated to provide a measure that accounts for saturated conditions and any slaking and filling over pores that may occur after an initial wetting event (INF-2”). After another 15 minutes had elapsed, surface (SH10) and subsurface hardness (SSH20) were measured at 10 and 20 cm, respectively, using a penetrometer inserted at three distinct points inside the central ring. This allowed for both co-location with infiltration and reduction in variability of moisture content, which could contribute additional variation to

penetrometer readings (Ayers & Perumpral, 1982; Herrick & Jones 2002). Soil samples from the initial sampling event were air-dried at 25°C for 72 h, sieved to <6mm, and a representative subsample was removed to assess aggregate stability (Kemper & Rosenau 1986; Stott et al. 2019). The remaining sample was sieved to <2mm for all further analyses.

For aggregate stability, a 25-g sample of the air-dried, <6mm sieved soil was placed on a <2mm sieve with a <0.25 sieve nested underneath and submerged in DI water in a sieving apparatus and run at approximately 30 cycles per minute (min) for 5 min. Remaining soil from each sieve was dried at 105°C. After weighing, dry soil from the <2mm sieve was returned to the sieve, remaining aggregates were crushed (avoiding crushing directly on the sieve), and the procedure was repeated to calculate sand and gravel content. Total initial sample weight was corrected for sand and gravel content prior to calculating percent macroaggregates (Agg_{ma}) (>2mm) and microaggregates (Agg_{mi}) (0.25mm-2mm). Mean weight diameter (mm) (MWD) was calculated, according to Van Bavel 1949, as:

$$MWD = \sum_i^n x_i w_i = 1$$

where x_i , is the mean diameter of the sieve size range, and w_i , is the fraction of soil remaining after sieving (after correction for sand and gravel).

Particle size was determined using a bouycous hydrometer, according to Gavlack et al., (2005). BD samples were oven-dried at 105°C until the mass did not change and density was determined using the total volume of the core(s) (Blake & Hartge, 1986). Rock fragments (>2mm) present in Corning soils were weighed, wrapped in paraffin wax and submerged in water to determine volume. Mass and volume of rock fragments, where present, were subtracted from the total mass and volume prior to calculating bulk density.

3.3.5 Chemical Indicators

All visible plant materials, including fine roots, were removed from <2mm sieved soil. Soil pH was measured in a 1:2 solution with 0.01M CaCl₂ using a pH electrode calibrated to pH 4, 7, and 10 standard buffers (Miller & Kissel, 2010). Subsamples were oven-dried at 60°C for 72 hours and ground for 12-24 hours, or until a fine powder, able to pass through a 100 mesh sieve, was achieved. Total C and N were determined on ground/ball milled samples by dry combustion using an ECS 4010 Costech Elemental Analyzer and a LECO soil standard (Blair et al., 1995). POXc was determined on air-dried, <2mm sieved soils in duplicate, using the method described by Weil et al. (2003a) as adapted by Culman et al. (2012). In 50 mL centrifuge tubes, 2.5 g of oven-dried equivalent soil was combined with 0.2 M KMnO₄ and 9ml MΩ cm⁻¹ water, yielding 20 mL of 0.02 M KMnO₄. The mixture was immediately shaken at 120 rpm for 2 min, then allowed to settle for 10 min. The supernatant was diluted (1:50) and absorbance at 550 nm was quantified by spectrophotometry. POXC was calculated assuming 9,000 mg C oxidized mol⁻¹ permanganate (Weil et al., 2003b).

3.3.6 Biological Indicators

Samples were stored at 4 C, until further analyses could be completed. Soil samples were analyzed using field-moist soil on an oven-dry mass basis. Soil enzymes that catalyze depolymerization of C-containing substrates were assayed to assess potential differences in maximum rates of hydrolysis, *i.e.*, cellobiohydrolase (CEL) (Enzyme Commission 3.2.1.91), β-glucosidase (BG) (EC 3.2.1.21) and N-acetyl-β-glucosaminidase (NAG) (EC 3.2.1.30). Assays were performed using field-moist soils based on Tabatabai (1994) as modified by Margenot et al. (2018). The equivalent of 1 g of oven-dried soil was incubated for 1 h (BG, NAG) or 2 h (CEL)

at 37°C in 5 mL of 18.2ΩM·cm water. No buffer was used because it has been shown that modified universal buffer does not maintain assay pH better than water (Li et al., 2021) and because buffer requires an assumption of a single pH optimum that is incorrect for many soil enzymes, including CEL and BG (Niemi and Vepsäläinen, 2005; Turner, 2010; Wade et al., 2021). A final substrate concentration of 10 mmol L⁻¹ for BG and NAG, and 5 mmol L⁻¹ for CEL was used to ensure substrate saturation (Malcolm, 1983; Margenot et al., 2018). Reactions were immediately alkalized after 1 h by the addition of 4 mL of 0.1 mol L⁻¹ Tris (pH 12.0) and 1 mL of 2 mol L⁻¹ CaCl₂.

Assays were centrifuged and an aliquot was used to quantify para-nitrophenol (pNP) colorimetrically using absorbance at 410 nm (Turner, 2010). Three corrections were performed to account for non-enzymatic absorbance (Margenot et al., 2018; Daughtridge et al., 2021). Mean absorbance of negative controls (*i.e.*, substrate but no soil) were subtracted from absorbance of soil assays to account for non-enzymatic hydrolysis of substrate during the incubation (Neal et al., 1981; Turner et al., 2002). Enzyme activities were corrected for potential dissolved organic matter contribution to absorbance with a soil-only control. Finally, potential sorption of pNP by soil components was corrected by measuring recovery of pNP in the same assay conditions using a relevant pNP concentration (1 mmol L⁻¹ per g soil) (Cervelli et al., 1973; Margenot et al., 2018).

Microbial biomass C (MBC) and N (MBN) and extractable organic C (EOC) and N (EON) were determined on field-moist soils. Microbial biomass was measured by sequential fumigation-extraction using chloroform gas (Vance et al., 1987). First, plant roots were removed prior to analysis. An oven-dry soil mass equivalent of 6 g was subjected to 16 h chloroform gas in fumigated samples; a second set of non-fumigated samples was also prepared. Both fumigated

and non-fumigated soils were then extracted by 2 mol L⁻¹ KCl (1:5 m/v, 30 min shaking at 120 rpm), which has been found to provide a more complete extraction than 0.5 M K₂SO₄ (Murage & Voroney 2007). Total organic C and N in extracts was quantified by UV-persulfate oxidation. The labile fractions of EOC and EON were obtained in extracts of non-fumigated samples. MBC and MBN were calculated as the difference between extractable C and N in fumigated and non-fumigated samples. No correction factors were applied.

3.3.7 Statistical Analyses

Each soil health variable was tested for multivariate normality and homogeneity of covariance and, where necessary, transformed to meet MANOVA assumptions. Total C and N were adjusted with log (x + 1) transformations, microbial biomass and enzyme potential activity were adjusted with square root transformations, while KCl extractable C and N were adjusted with reciprocal transformations (1/n). According to Hatcher and Stepanski (1996), Two-way multivariate analysis of variance (MANOVA) was conducted to determine if there were significant effects of treatment and/or soil type across the set of soil health variables, followed by Two-way ANOVAs on each individual variable, using a mixed-effects model named nlme, in the R statistical package (Pinheiro et al. 2018). Management (within-subject factor) and soil type (between-subject factor) were considered fixed effects, while site was considered a random effect (based on repeated measures). Data was analyzed separately for each sampling depth. Differences between means were calculated using Tukey's Honestly Significant Difference (HSD) tests. Statistical significance was evaluated at P < 0.05 unless otherwise stated.

Although transformations were able to adjust data to largely meet ANOVA assumptions, some variables, especially biological variables, still appeared slightly positively or negatively

skewed or only confirmed equality of variance using the Levene's test, which is known to be less sensitive to departures from normality than the Bartlett's test. It is also possible that a rejection of the null hypothesis of normality after transformation is simply due to insufficient sample size. Furthermore, recent work has discovered that soil biological activity occurs in hotspots, (Kuzyakov & Blagodatskaya 2015; Baveye et al. 2018; Tian et al. 2020; Fierer et al. 2021), challenging the assumption that samples are drawn from a symmetric distribution. Non-parametric methods control the rate of false rejections of the null hypothesis without any distributional assumptions (Webster and Lark 2019). In this sense, they are more robust than normal-theory MANOVA. Non-parametric methods are also often more powerful than parametric alternatives when the null is false (Lehmann and Romano 2005). We used a non-parametric MANOVA based on permutation tests to assess two global null hypotheses: a) no treatment effect on any of the soil health outcomes, and b) no interaction effect of treatment with soil type on any of the soil health outcomes.

For the test of treatment effect (a), we first ran paired two-sample permutation tests on each of the soil health outcomes. Specifically, to run these partial tests we simulated the "permutation distribution" of the difference-in-means under the null hypothesis by randomly flipping the treated/control labels within each pair 100,000 times, recomputing the difference in means each time. The original difference-in-means was then compared to these 100,000 draws from the permutation distribution. The fraction of draws with magnitude larger than the original difference-in-means was a non-parametric p-value for that partial test. The p-value for the global null was then computed from the partial permutation distributions using the non-parametric combination of tests (NPC) approach (Pesarin & Salmaso 2010).

For the test for interaction (b), we ran a permutation one-way ANOVA of the within-pair differences on soil type for each individual outcome. Specifically, the mean soil health outcome within each soil type was squared and weighted by the number of plots corresponding to that soil type. These weighted squared mean outcomes were then summed across soil types to compute the original ANOVA test statistic. The permutation distribution was simulated by shuffling the soil type labels 10,000 times, recomputing this ANOVA test statistic each time. The partial p-value was then the fraction of draws greater than the original ANOVA statistical test. The global p-value for interaction was again computed by combining these partial tests using NPC. In addition to testing the two global nulls, we examined p-values for the partial tests to understand which soil health outcomes were affected by management. Corrections were made for multiple testing, using the Benjamini-Yekutieli procedure (Benjamini & Yekutieli 2001).

Linear regression models were used to evaluate the relationship between C or N concentrations and MBC or MBN, respectively, as well to understand specific enzyme activity, the relationship between enzyme potential activity and MBC, and the relationship between aggregate stability, total C and silt/clay content. Where regressions were significant ($P < 0.05$) in both management types, residuals were tested for normality and homogeneity of variance, and analysis of covariance (ANCOVA) was used to determine if relationships were different between management types. Relationships between variables were further explored using Spearman's correlation coefficients. Significance of correlations at $P < 0.05$ were calculated for biological, chemical, and physical properties, excluding in-field performance indicators (infiltration rate, surface/subsurface hardness) using the *Hmisc* package in R (Harrell & Dupont, 2018).

Sources of variability in the dataset were characterized by Principal Components Analysis (PCA) (Figure 2.7) on a standardized correlation matrix using the *vegan* package in R

(Oksanen et al., 2012). Loadings and proportions of variance, as well as the raw data for included variables are presented in SI Table 2.3 and 2.4. The first 6 components were selected based on visual interpretation of the scree plots and criteria of having eigenvalues >1 and a cumulative variance of at least 70% (Jolliffe 2002).

3.4 Results

3.4.1 Soil Health Metrics

Hedgerow soils and adjacent cultivated fields differed dramatically in physical, chemical, and biological properties (Table 3.2). Soil type and depth had less pronounced effects. At 0-10 cm, Two-way MANOVA across all variables showed management ($P = 1.12e-07$) and soil type effects ($P=0.0002$), but not an interaction effect. At 10-20 cm, there was a significant soil type effect, ($P=9.418e-09$), a weak management effect ($P=0.058$), and no interaction effect. Two-way ANOVA for each individual property at 0-10 cm showed higher responses under hedgerows than cultivated fields, except for KCl extractable N, pH, gravimetric water content, surface hardness, and bulk density, which were not different between management types (Table 3.2). At 10-20 cm, similar trends occurred but to a lesser degree. Permutation tests, a non-parametric approach to comparing means showed the same results at 0-10cm. At 10-20 cm, however, only total C, total N, BG, and SSH20 were significant by management type, differing from MANOVA results in regards to MBC and CEL.

Soil texture was not different between paired sites (hedgerow vs. cultivated) at either depth (Table 3.2), indicating good correspondence between paired sampling locations. Dynamic physical properties differed between paired sites. At 0-10 cm, aggregate stability ($F=156.27$) was 2.6 times higher in hedgerows than in adjacent cultivated fields (Table 3.2). INF-1” was 2.4

times slower in cultivated fields than hedgerows, while INF-2” (F=30.89) took 6.9 times as long in cultivated fields. At 10-20 cm, management had an effect on SSH20 (F=54.91), which was lower under hedgerows than cultivated fields. Bulk density, however, was not different between the two management types.

Soil chemical and biochemical properties also differed under hedgerows, relative to cultivated fields. Total C (F=141.4) and total N (F=98.22) were 1.9 to 2.3 times higher in hedgerows than in adjacent cultivated fields at 0-10 cm (Table 3.2). At 10-20 cm, management also increased total C (F=16.67) and total N (F=7.35), but differences were only 1.2 or 1.3 times higher, respectively. In the surface 0-10 cm, microbial biomass C (F=45.22), β -glucosidase (F=149.56), β -glucosaminidase (F=36.56), and cellulase (F=62.78) were 1.8 to 3.0 times higher in hedgerows than in adjacent cultivated fields (Table 3.2). At 10-20 cm, management affected MBC (F=7.01), BG (F=13.62), and CEL (F=9.21), but differences were less pronounced compared to 0-10 cm. Although MBC, BG, and CEL were higher under hedgerows relative to cultivated fields at both depths, follow up Tukey’s HSD tests did not identify any single soil type as significantly different by management type (data not shown). Analysis of the ratio between enzyme activities and MBC, enzyme activities and SOC, as well as MBC:MBN were not different between management types at either depth (data not shown).

3.4.2 Variability in Soil Health Metrics

Physical indicators measured in-field, including infiltration rate and surface hardness, were the most variable, both overall and between management types. Coefficients of variation

Table 3.2. Mean and standard error values for measured soil health variables at 0-10 cm and 10-20 cm depths by management type (hedgerow and cultivated fields). Two-way ANOVA results are listed for each depth by management type, soil type, and their interactions. Asterisks indicate significant differences at * = P< 0.05, ** = P< 0.01, *** = P< 0.001.

Indicator	0-10 cm depth							10-20 cm depth						
	Hedgerow		Cultivated		M	ST	M x ST	Hedgerow		Cultivated		M	ST	M x ST
	Mean	SE	Mean	SE				Mean	SE	Mean	SE			
Soil carbon (g kg ⁻¹)	2.57	0.11	1.13	0.05	***	ns	ns	1.29	0.06	0.97	0.04	***	ns	*
Soil nitrogen (g kg ⁻¹)	0.23	0.01	0.12	0.01	***	ns	ns	0.13	0.01	0.11	0.01	***	ns	**
pH	6.44	0.07	6.40	0.07	ns	***	ns	6.43	0.07	6.42	0.07	ns	***	*
MBC (mg kg ⁻¹)	156.0	9.6	87.0	4.9	***	ns	*	85.5	6.9	66.7	4.4	**	ns	ns
MBN (mg kg ⁻¹)	60.8	7.4	28.4	2.4	***	ns	ns	25.6	2.6	21.6	2.4	ns	ns	ns
MBC:MBN	3.8	0.5	4.2	0.6	ns	ns	*	5.1	1.0	5.8	1.4	ns	ns	ns
POXc (mg kg ⁻¹)	747.2	32.1	546.6	23.3	***	ns	ns	526.6	27.6	472.5	26.3	ns	ns	ns
EOC (mg kg ⁻¹)	38.7	3.1	29.2	2.3	***	ns	*	27.5	4.1	25.3	3.0	ns	ns	ns
EON (mg kg ⁻¹)	32.7	2.7	30.6	2.4	ns	ns	ns	18.2	1.5	22.5	2.4	ns	ns	ns
β-glucosaminidase (μmol pNP g ⁻¹ soil h ⁻¹)	0.44	0.03	0.24	0.02	***	ns	ns	0.22	0.02	0.17	0.02	ns	ns	ns
β-glucosidase (μmol pNP g ⁻¹ soil h ⁻¹)	2.57	0.13	0.97	0.06	***	ns	ns	0.85	0.07	0.55	0.05	***	ns	ns
cellulase (μmol pNP g ⁻¹ soil h ⁻¹)	0.55	0.06	0.18	0.02	***	ns	ns	0.13	0.01	0.10	0.01	**	ns	ns
Macroaggregates (%)	46.4	2.0	34.4	1.4	***	*	*	-	-	-	-	-	-	-
Microaggregates (%)	21.7	1.1	30.3	1.2	***	*	*	-	-	-	-	-	-	-
Mean Weight Diameter (mm)	1.94	0.08	0.76	0.03	***	**	**	-	-	-	-	-	-	-
Infiltration dry (sec/in)	14.0	1.6	33.4	6.5	***	ns	ns	-	-	-	-	-	-	-
Infiltration wet (sec/in)	35.9	4.3	248.4	41.8	***	ns	ns	-	-	-	-	-	-	-
GWC (g g ⁻¹)	0.15	0.01	0.14	0.01	ns	*	ns	0.13	0.01	0.16	0.01	**	*	ns
Surface hardness (psi)	28.3	2.2	31.9	2.7	ns	ns	ns	89.9	3.1	132.1	5.4	***	ns	ns
Bulk density (g cm ⁻³)	1.34	0.02	1.33	0.02	ns	*	***	1.52	0.02	1.48	0.02	ns	***	ns

(CV) for infiltration rates were 87% under hedgerows and 131% in cultivated fields. Surface hardness at 0-10 cm had a CV of 54% under hedgerows and 94% in cultivated fields. Surface hardness at 10-20 cm, however, was less variable with a CV of 25% in hedgerows and 31% in cultivated fields. Lab-based physical indicators, including BD and MWD were also less variable. For MWD (0-10 cm only), CV was 30% under hedgerows and 27% in cultivated fields. BD had a CV of 9-11% in both systems and at both depths.

Biological indicators were also highly variable overall, but less so between management types, as compared to physical properties. At 0-10 cm, microbial biomass had a CV of 47-64% under hedgerows and 43-66% under cultivated fields. Enzyme potential activity had a CV of 48-80% for hedgerows and 51-63% for cultivated. The greatest variability was observed in MBN and the lowest in BG. Chemical/biochemical indicators were some of the least variable metrics, overall and between management types. pH had a CV of 8-9% under both management types, while total C and N and POXc had CV's of 33-38% across management types. EOC and EON, however, were more variable C and N pools with a CV of 60-64%.

Variability was higher at the 10-20 cm depth for most biological and chemical indicators, except pH which was the same at both depths. CV's for microbial biomass ranged from 52 to 112% and potential enzyme activities ranged from 55% to 87% across management types. The various C and N pools had CVs ranging from 35%-116% in hedgerows and 36-92% in cultivated fields.

3.4.3 Soil Type Effects

Based on Two-way ANOVA, only pH, Agg_{ma}, Agg_{mi}, MWD, and BD differed by soil type at 0-10 cm (Table 3.2). Such differences were only apparent for pH and BD at 10-20 cm.

At 0-10 and 10-20 cm, pH was lower in Corning than all other soil types ($P < 0.0001$), with a mean of 5.3 at both depths and a mean of 6.5 at both depths for Yolo, Brentwood, and Capay. At 0-10 cm, Capay soils tended to be denser than Brentwood ($P = 0.062$), but all other soils were similar. At 10-20 cm, Capay was denser than Yolo ($P = 0.013$), Brentwood ($P = 0.001$), and Corning ($P = 0.049$), which were all similar to one another. MWD were lower for Corning than all other soil types ($P < 0.05$). Agg_{ma} tended to be lower on Corning soils than Capay and Brentwood ($P = 0.073$ and $P = 0.056$, respectively), and Agg_{mi} lower on Corning soils than Brentwood ($P = 0.013$). Thus, lower pH and aggregate stability of Corning, and the higher density of Capay, were the main differences in soil type across the study sites.

3.4.4 Interaction Effects

Soil type played a role in the magnitude of the management effect for several physiochemical properties and microbial biomass indicators, according to MANOVA. No universal trends existed, but interaction effects often originated from Yolo and/or Corning, relative to other soils (Table 3.2). For example, at 0-10 cm, the Corning hedgerow soils increased MBC much more strongly relative to cultivated soils ($+132.8 \text{ mg kg}^{-1}$) as compared to the Yolo ($+48.2 \text{ mg kg}^{-1}$) and Brentwood ($+49.2 \text{ mg kg}^{-1}$) (SI Figure 3.1). The increase in EOC under hedgerows was greater for Yolo ($+26.56 \text{ mg kg}^{-1}$) compared to Capay ($+2.37 \text{ mg kg}^{-1}$) (SI Figure 3.1); while the increase in MWD was greater for Yolo ($+1.53 \text{ mm}$) compared to Capay ($+1.00 \text{ mm}$) and Corning ($+0.79 \text{ mm}$) (SI Figure 3.4). Overall, at 0-10 cm, the management effect was mediated by soil type for MBC, MBC:MBN ratio, EOC, Agg_{ma} , Agg_{mi} , MWD, and BD.

Soil type interactions were less prominent at the 10-20 cm depth. None of the microbial or physical indicators showed soil type interactions at this depth, and of the chemical indicators, only total C, total N, and pH. The increase in pH under hedgerows in Corning was significantly

greater (+0.19) than the negligible decrease on Brentwood (-0.09) and Capay (-0.06). The increase in total soil C and N (+0.52 g kg⁻¹ and +0.04 g kg⁻¹, respectively), for Yolo was significantly greater than Brentwood (+0.09 g kg⁻¹ and +0.00 g kg⁻¹), but not other soils, which increased on average +0.35 g kg⁻¹ and 0.023 g kg⁻¹, respectively.

Using Permutation Tests to compare mean differences between hedgerows and cultivated fields by soil type, similar interaction effects were observed (Table 3.3). At 0-10 cm, pH, BD, MBC:MBN ratio, Agg_{ma}, Agg_{mi}, and MWD all differed by interaction of management and soil type; aligning with parametric results. Total N followed a similar trend at 10-20 cm and POXc and EOC were also found to be significantly different using permutation tests, despite no significance in parametric tests. Differences in POXc and EOC were attributed to differences in Corning. pH was significant in both tests, but BD was only significant by soil type, not the interaction effect, according to parametric tests.

3.4.5 Relationships between soil carbon, biological, and physical properties

Total soil C was more strongly correlated with other soil variables under hedgerows than cultivated fields, and at 0-10 cm than 10-20 cm. Under hedgerows (0-10 cm), total soil C and all other variables, except pH and EON, were positively related based on results of the Spearman's correlation tests ($P < 0.001$) (Table 3.4). In cultivated fields (0-10 cm), total soil C was correlated with MBN and POXc ($P < 0.05$) and to MBC, NAG, and CEL ($P < 0.05$) (Table 3.5). Linear regressions of soil C and MBC further corroborated a correlation under hedgerows ($y_h = 37.0 + 45.7x$, $R^2 = 0.42$, $P = 4.92e^{-16}$) (Figure 3.1) and in cultivated fields ($y_c = 48.8 + 33.7x$, $R^2 = 0.09$, $P = 0.013$) (Figure 3.1). ANCOVA showed no differences in the relationship between soil C and MBC across management types.

Table 3.3. Results from permutation tests of soil health variables at 0-10 and 10-20 cm for management type and soil type. P-Values were adjusted to account for multiple tests, using the Benjamini-Yekutieli procedure.

Soil Health Indicator	0-10 cm depth		10-20 cm depth	
	P-value		P-value	
	Treatment	Soil Type	Treatment	Soil Type
Soil carbon (g kg ⁻¹)	<2e ⁻¹⁶	1.000	0.005	0.129
Soil nitrogen (g kg ⁻¹)	<2e ⁻¹⁶	0.106	0.041	0.003
pH	0.459	<2e ⁻¹⁶	0.969	<2e ⁻¹⁶
POXc (mg kg ⁻¹)	<2e ⁻¹⁶	0.269	0.526	0.030
EOC (mg kg ⁻¹)	0.051	0.194	1.000	0.030
EON (mg kg ⁻¹)	1.00	1.000	0.526	0.618
MBC (mg kg ⁻¹)	1.95e ⁻⁰⁴	1.000	0.334	1.000
MBN (mg kg ⁻¹)	<2e ⁻¹⁶	0.715	0.526	0.217
MBC:MBN Ratio	1.000	0.008	1.000	0.167
β-glucosaminidase (μmol pNP g ⁻¹ soil h ⁻¹)	<2e ⁻¹⁶	1.000	0.189	1.000
β-glucosidase (μmol pNP g ⁻¹ soil h ⁻¹)	<2e ⁻¹⁶	1.000	0.010	1.000
cellulase (μmol pNP g ⁻¹ soil h ⁻¹)	<2e ⁻¹⁶	0.737	0.098	0.288
Macroaggregates (%)	0.002	<2e ⁻¹⁶	NA	NA
Microaggregates (%)	0.002	0.003	NA	NA
MWD	<2e ⁻¹⁶	0.015	NA	NA
Infiltration dry (sec/in)	0.029	0.106	NA	NA
Infiltration wet (sec/in)	<2e ⁻¹⁶	1.000	NA	NA
GWC (g g ⁻¹)	0.445	<2e ⁻¹⁶	0.002	<2e ⁻¹⁶
Surface hardness (psi)	0.453	0.106	<2e ⁻¹⁶	1.000
Bulk density (g cm ⁻³)	0.970	<2e ⁻¹⁶	0.293	<2e ⁻¹⁶

At 0-10 cm in hedgerow soils, MBC was strongly correlated with the potential activities of all three enzymes (Table 3.4, Table 3.5), but ANCOVA results showed they were more strongly related in hedgerows than cultivated fields for both BG (P=0.001) and CEL (P=0.02) (Figure 3.2). Of the three enzyme potential activities measured, β-glucosidase had the strongest relationship with total soil C under both management types, according to Spearman correlation coefficients. Linear regressions across management and soil types indicate positive relationships between BG and soil C at 0-10 cm ($y=0.36 + 0.76x$, $R^2=0.44$, $P<2.2e^{-16}$) and at 10-20 cm

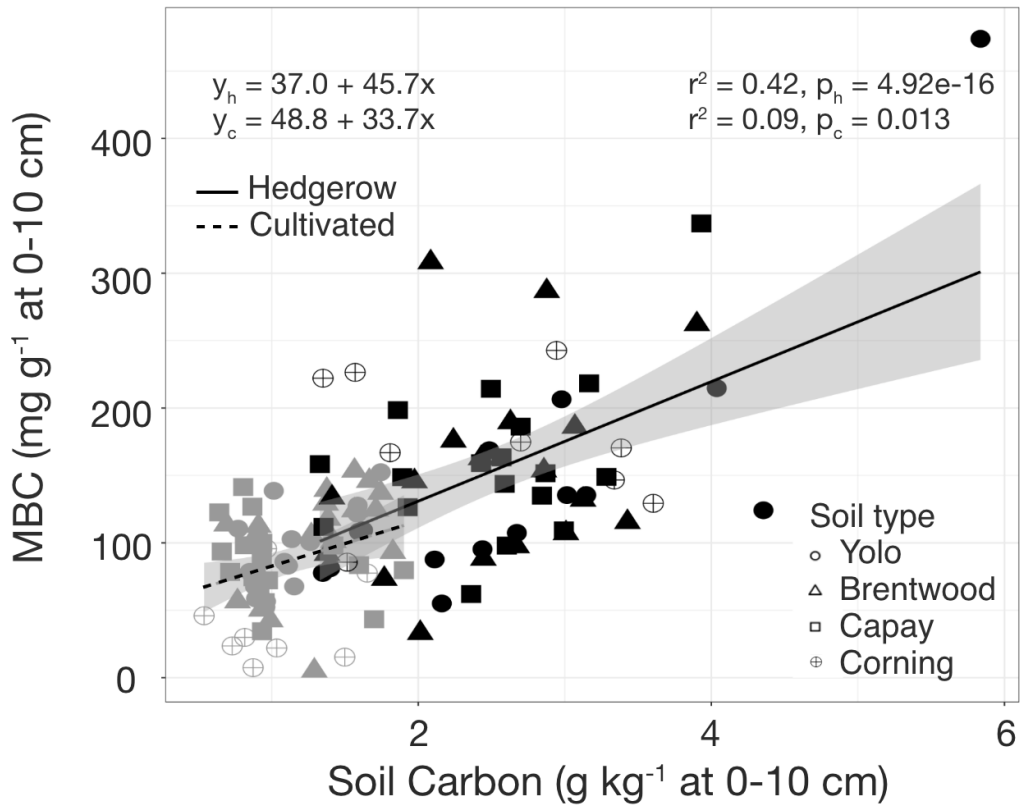


Figure 3.1. Correlation between total soil carbon concentrations (g kg^{-1}) and microbial biomass carbon (mg kg^{-1}) at 0-10 cm depth. P-values represent ANOVA results for each treatment individually. Analysis of covariance (ANCOVA) indicated no significant difference in the relationship by management type.

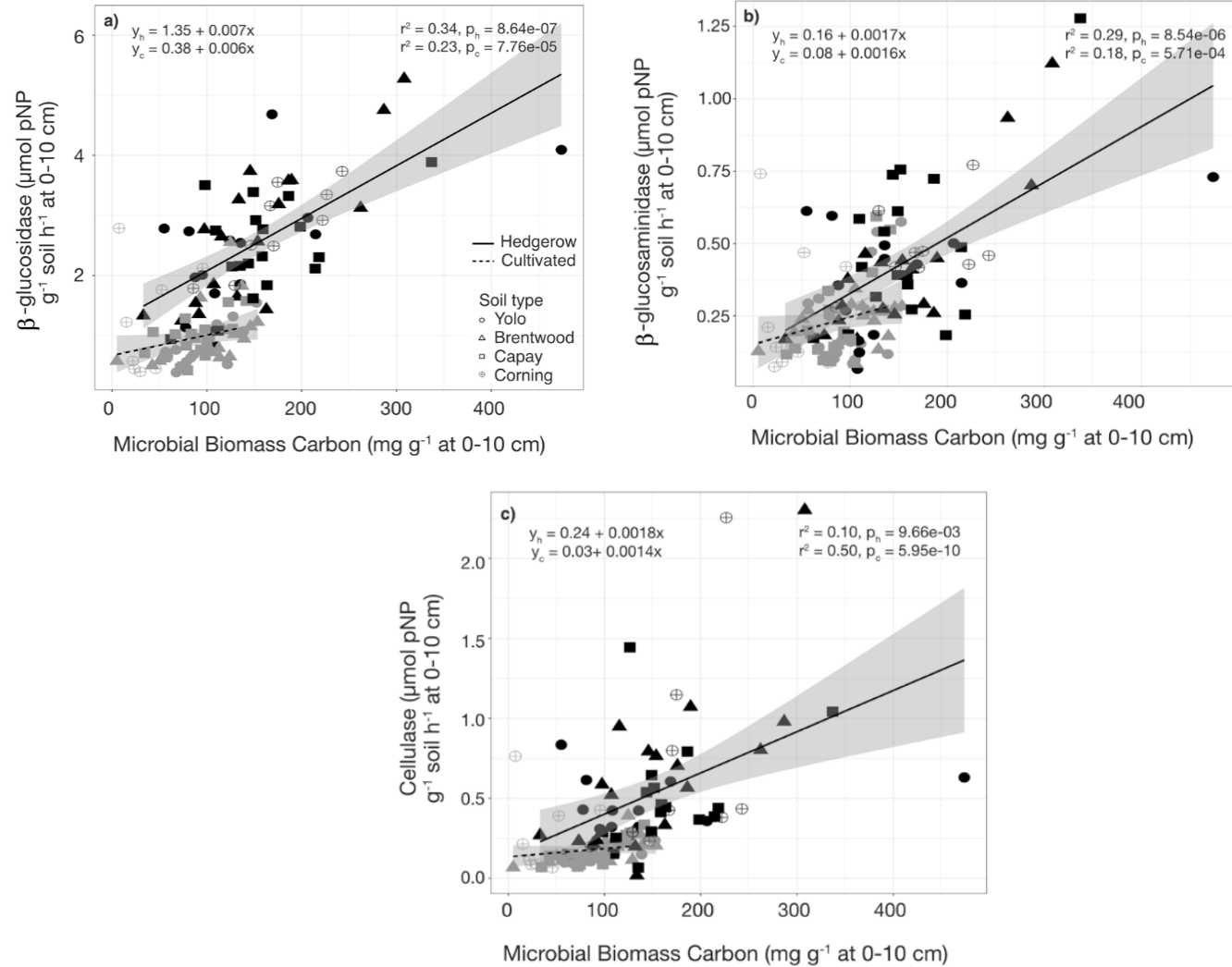


Figure 3.2. Correlation between microbial biomass carbon (mg kg^{-1}) and enzyme potential activity at 0-10 cm depth. P-values represent ANOVA results for each treatment individually. Analysis of covariance (ANCOVA) indicated significant differences by management type between microbial biomass carbon and a) β -glucosidase at $P=0.001$ and c) cellulase at $P=0.02$.

Table 3.4. Spearman's correlation coefficients (adjusted for multiple testing) between soil physiochemical and biological properties in the surface (0-10cm) of hedgerows in Yolo County, California. Asterisks represent a significant correlation between variables at * = P < 0.05, ** = P < 0.01, and *** = P < 0.001.

	Hedgerows												
	Soil C	Soil N	pH	GWC	MBC	MBN	EOC	EON	POXc	BG	NAG	CEL	MWD
Soil nitrogen (g kg ⁻¹)	0.94 ***	-	-	-	-	-	-	-	-	-	-	-	-
pH	0.12 ns	0.22 *	-	-	-	-	-	-	-	-	-	-	-
GWC (g g ⁻¹)	0.28 ***	0.31 ***	0.32 ***	-	-	-	-	-	-	-	-	-	-
MBC (mg kg ⁻¹)	0.60 ***	0.56 ***	0.11 ns	0.39 ***	-	-	-	-	-	-	-	-	-
MBN (mg kg ⁻¹)	0.50 **	0.51 ***	0.29 ***	0.40 ***	0.65 ***	-	-	-	-	-	-	-	-
EOC (mg kg ⁻¹)	0.31 **	0.25 **	0.01 ns	-0.10 ns	0.26 ***	0.16 ns	-	-	-	-	-	-	-
EON (mg kg ⁻¹)	0.16 ns	0.21 *	0.13 ns	0.33 ***	0.22 *	0.26 ***	0.48 ***	-	-	-	-	-	-
POXc (mg kg ⁻¹)	0.60 ***	0.61 ***	0.27 ***	0.20 *	0.36 ***	0.25 **	0.23 **	0.20 *	-	-	-	-	-
β-glucosaminidase (μmol pNP g ⁻¹ soil h ⁻¹)	0.71 **	0.62 ***	-0.02 ns	0.15 ns	0.70 ***	0.50 ***	0.43 ***	0.24 **	0.41 ***	-	-	-	-
β-glucosidase (μmol pNP g ⁻¹ soil h ⁻¹)	0.44 **	0.34 ***	-0.08 ns	0.12 ns	0.57 ***	0.22 *	0.24 **	0.14 ns	0.28 ***	0.69 ***	-	-	-
cellulase (μmol pNP g ⁻¹ soil h ⁻¹)	0.63 **	0.54 ***	0.01 ns	0.11 ns	0.65 ***	0.43 ***	0.36 ***	0.13 ns	0.34 ***	0.82 ***	0.69 ***	-	-
Macroaggregates (%)	0.34 ***	0.30 ***	0.24 *	0.26 ***	0.27 ***	0.09 ns	0.23 **	0.14 ns	0.28 ***	0.34 ***	0.11 ns	0.33 ***	-
Microaggregates (%)	-0.30 ***	-0.25 **	0.07 ns	0.10 ns	-0.13 ns	-0.17 ns	-0.25 **	-0.15 ns	-0.14 ns	-0.35 ***	-0.15 ns	-0.03 ***	-0.50 ***

Table 3.5. Spearman's correlation coefficients (adjusted for multiple testing) between soil physiochemical and biological properties in the surface (0-10cm) of cultivated fields in Yolo County, California. Asterisks represent a significant correlation between variables at * = P < 0.05, ** = P < 0.01, and *** = P < 0.001.

	Cultivated												
	Soil C	Soil N	pH	GWC	MBC	MBN	EOC	EON	POXc	BG	NAG	CEL	MWD
Soil nitrogen (g kg ⁻¹)	0.84 ***	-	-	-	-	-	-	-	-	-	-	-	-
pH	0.31 *	0.42 ***	-	-	-	-	-	-	-	-	-	-	-
GWC (g g ⁻¹)	0.40 ***	0.49 ***	0.40 ***	-	-	-	-	-	-	-	-	-	-
MBC (mg kg ⁻¹)	0.27 *	0.39 ***	0.25 *	0.51 ***	-	-	-	-	-	-	-	-	-
MBN (mg kg ⁻¹)	0.50 ***	0.58 ***	0.34 **	0.52 ***	0.50 ***	-	-	-	-	-	-	-	-
EOC (mg kg ⁻¹)	0.12 ns	-0.04 ns	-0.11 ns	-0.16 ns	-0.03 ns	-0.16 ns	-	-	-	-	-	-	-
EON (mg kg ⁻¹)	0.12 ns	0.29 *	0.15 ns	0.34 **	0.22 ns	0.16 ns	0.40 ***	-	-	-	-	-	-
POXc (mg kg ⁻¹)	0.37 ***	0.37 ***	0.25 ns	0.08 ns	0.34 **	0.14 ns	0.28 *	0.29 *	-	-	-	-	-
β-glucosaminidase (μmol pNP g ⁻¹ soil h ⁻¹)	0.26 *	0.18 ns	-0.06 ns	0.06 ns	0.46 ***	0.25 *	0.27 *	0.27 *	0.19 ns	-	-	-	-
β-glucosidase (μmol pNP g ⁻¹ soil h ⁻¹)	-0.09 ns	-0.17 ns	0.05 ns	-0.12 ns	0.32 ***	-0.06 ns	0.01 ns	-0.13 ns	0.09 ns	0.50 ***	-	-	-
cellulase (μmol pNP g ⁻¹ soil h ⁻¹)	0.26 *	0.18 ns	0.00 ns	0.05 ns	0.55 ***	0.28 *	0.12 ns	0.05 ns	0.23 ns	0.75 ***	0.70 ***	-	-
Macroaggregates (%)	0.10 ns	0.00 ns	0.08 ns	0.35 **	0.16 ns	-0.18 ns	0.22 ns	0.27 *	0.12 ns	0.05 ns	-0.15 ns	-0.16 ns	-
Microaggregates (%)	0.12 ns	0.23 ns	0.21 ns	0.17 ns	0.35 **	0.13 ns	-0.04 ns	-0.26 *	0.10 ns	0.14 ns	0.30 *	0.19 ns	-0.16 ns

($y=0.02 + 0.64x$, $R^2=0.38$, $P=1.89e^{-07}$). Under hedgerows, soil C was not correlated with NAG or CEL at 10-20 cm and was only weakly correlated with CEL in cultivated fields.

Linear regressions of soil C and MWD (Figure 3.3) showed a positive relationship across most soil types ($y=0.42 - 0.50x$, $R^2 = 0.45$, $P=2.56e^{-14}$). The relationship was stronger in hedgerows than cultivated fields for Yolo and in cultivated fields for Capay and Corning. Soils with poor correlations between MWD and soil C (except Brentwood) tended to have a better correlation between MWD and texture. Specifically, for Capay hedgerow, there was a poor negative relationship between MWD and soil C ($y_h = 2.52 - 0.24x$, $r^2 = 0.11$, ns), but a strong positive relationship between MWD and silt + clay content ($y_h = -4.39 + 0.08x$, $r^2 = 0.63$, $P<2.2e^{-16}$) (Figure 3.3). Corning hedgerow soils had a weak positive relationship between MWD and soil C ($y_h = 1.12 + 0.11x$, $r^2 = 0.05$, ns), but a stronger negative relationship ($y_c = 3.25 - 0.03x$, $r^2 = 0.29$, $P=0.031$).

Several biological soil health variables were related with physical soil health variables in both hedgerows and cultivated fields. In hedgerows, Spearman's correlation coefficients indicated MBC was strongly correlated with Agg_{ma} ($P<0.001$), but not Agg_{mi} (Table 3.4). C-cycling enzymes (BG and CEL) were strongly correlated with both Agg_{ma} and Agg_{mi} ($P<0.001$). In cultivated fields, MBC and BG were related with Agg_{mi} . Linear regressions also showed significant relationships between MWD and MBC ($y = 0.72 + 0.005x$, $R^2 = 0.23$, $P = 3.2e^{-08}$). C-cycling enzymes, BG ($y = 0.59 + 0.43x$, $R^2 = 0.43$, $P=3.8e^{-16}$) and CEL ($y=0.99 + 0.99x$, $R^2=0.24$, $P=9.9e^{-09}$), were also positively correlated with MWD.

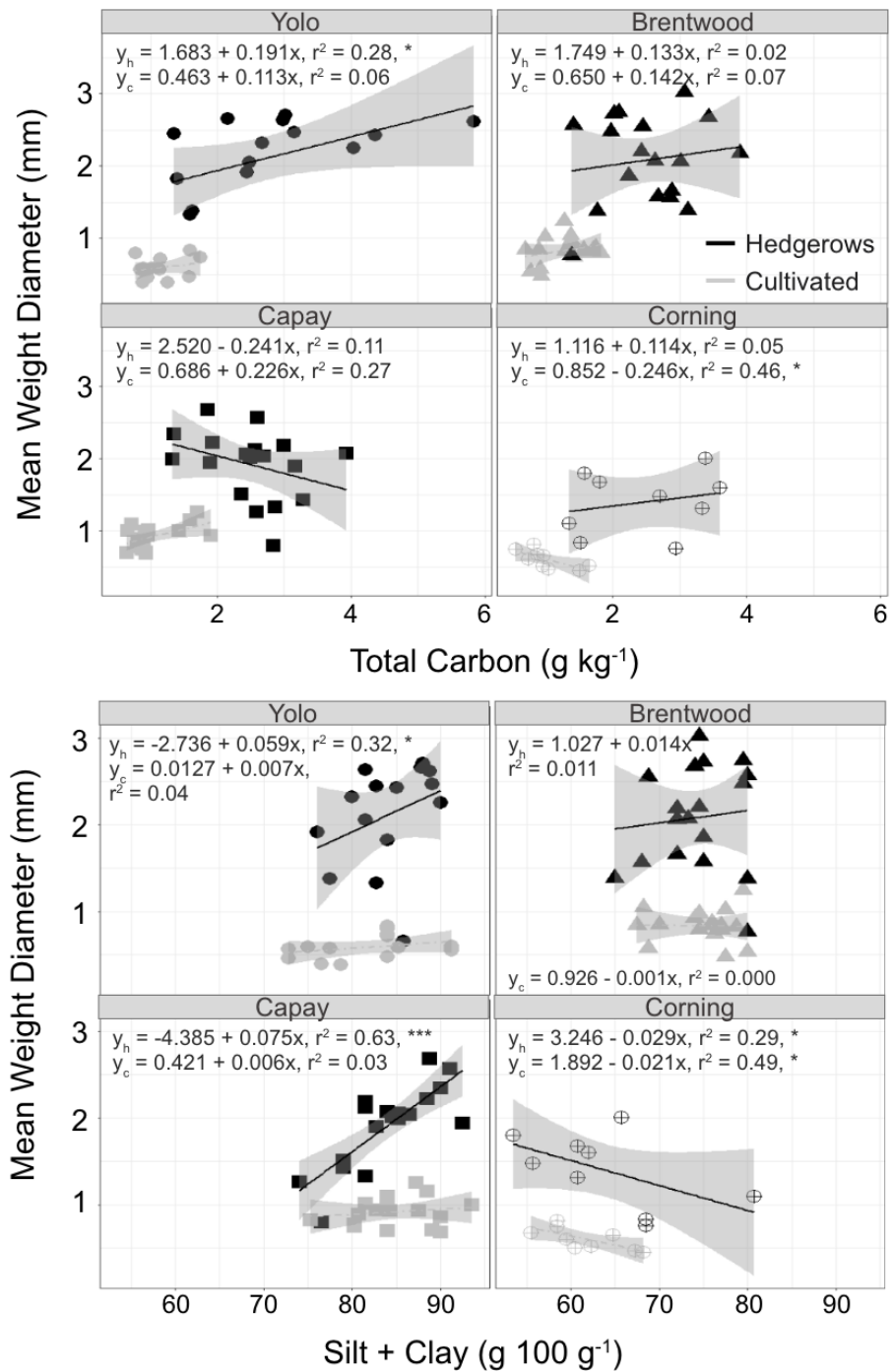


Figure 3.3. Correlations between aggregate mean weight diameter (mm) and a) total soil carbon (g kg⁻¹) and b) silt and clay content (g 100 g⁻¹ soil). P-values represent ANOVA results for each treatment individually at * = P < 0.05, ** = P < 0.01, and *** = P < 0.001.

3.4.6 Principal Components Analysis (PCA)

Ordination with PCA, to assess relationships between all measured soil variables, demonstrated distinct differences between management and soil types (Figure 3.4a and 3.4b). PC1 and PC2 explained a moderate degree of variation at both 0-10 cm (45.4%) and 10-20 cm depths (44.7%). Hedgerow and cultivated soils formed distinct clusters by management type with pronounced differentiation across Axis 1 at 0-10 cm (Figure 3.4a), but not 10-20 cm. (Figure 3.4b). Sites with the same management and soil types, however, tended to group together at both depths. Soil types tended to cluster across Axis 2, which was dominated by physical soil properties associated with soil type differences, including sand, clay, pH, and to a lesser degree, BD.

For the surface soil at 0-10 cm, variables with high negative loadings on PC1 at 0-10 cm included total soil C > MBC > BG > total N > POXc > NAG > MBN > CEL (Figure 3.4a; SI Table 3.1); all are biological and chemical variables. These variables were negatively associated with Axis 1, with hedgerows having higher absolute values, indicating that together they form a composite of soil characteristics that are typically associated with soil health. The physical variables that were measured in the field (SH10, INF-1”, INF-2”) as well as Agg_{mi} were moderately positively associated with Axis 1 with hedgerows having more negative values on Axis 1 as compared with cultivated soils. The clustering of these physical variables with chemical and biological variables on the left side of Axis 1 points out the complexity of relationships that generate soil health. Variables with high loadings on PC2 included Sand > Clay > pH > GWC > Agg_{ma} > BD, contributing to groupings by soil types.

At 10-20 cm, PC1 was dominated by high positive loadings for total soil C > total soil N > BG > MBC > MBN > CEL > POXc ((Figure 3.4a; SI Table 3.2)), while PC2 had high positive

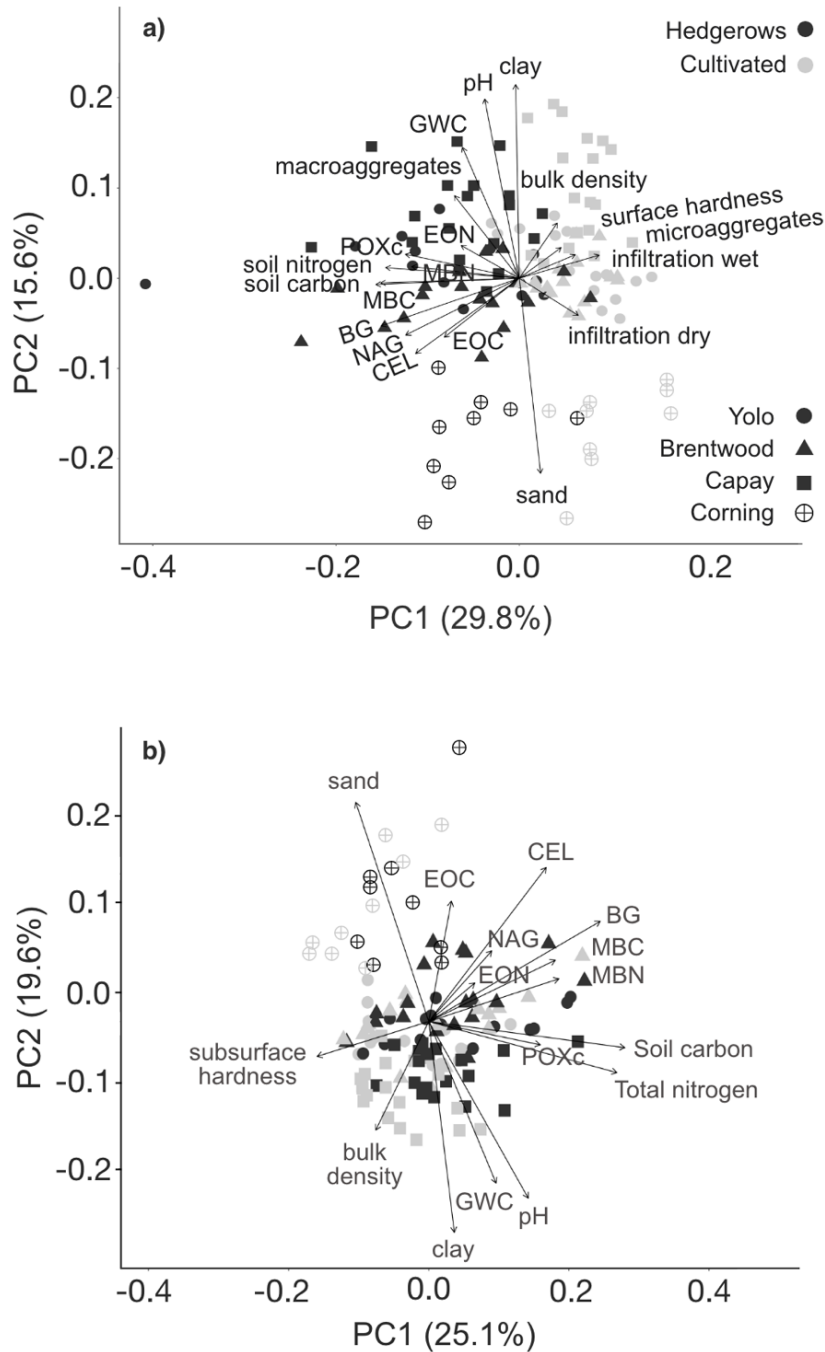


Figure 3.4. PCA ordination biplot of the 20 sites, classified by 4 soil types. Vectors represent soil health variables and soil physical properties at a) 0-10 cm and b) 10-20 cm depths. Values in parentheses on x- and y- axes labels represent the variation in data explained by each principal component. EOC, extractable organic carbon; EON, extractable organic nitrogen; MBC, microbial biomass carbon; MBN, microbial biomass nitrogen; POXc, permanganate oxidizable carbon; BG, β -glucosidase; NAG, β -glucosaminidase; CEL, cellulase; GWC, gravimetric water content

loadings for Sand > CEL > BG and high negative loadings for Clay > pH > GWC > BD.

Variables clustered similarly to those at 0-10 cm, but with little distinction between hedgerow and cultivated soils.

3.5 Discussion

Using a regional survey, this study provided an in-depth analysis of the range of soil responses to hedgerows on field edges on working farms, as compared with adjacent cultivated fields. We observed that hedgerows produced a pronounced improvement in soil properties that are typically considered to be indicators of soil health, such as accumulation of MBC, POX_c, total C, improved aggregate stability, and faster infiltration rate across all soil types. This is likely due to the concomitant implementation of a set of principles known for improving soil health (do not disturb, continuous ground cover, maintain living roots, and diversify). Improved biological indicators like higher MBC and enzyme potential activity in hedgerows were associated with improved physical indicators such as increased macroaggregates/total aggregates, higher MWD, and decreased surface hardness and infiltration rates. The consistent positive effects of hedgerows on soil C, MBC, infiltration, and other soil health variables may provide a ‘reference point’ as to the potential for soil health management in cultivated fields and marginal areas; providing site specific context for effective interpretation (McBratney et al. 2019; Maharjan et al. 2020; Fierer et al. 2021).

Hedgerows and cultivated fields are very different systems with different management goals; resulting in contrasting processes that contribute to differences in response to soil health metrics. Cultivated fields in row crop systems in Yolo County, often receive a high input of C

from crop roots and residues across the growing season, but also tend to experience long periods of bare fallow (average 4.3, range of 2-7 months in our study) with low C inputs, which has been found to contribute to lower SOM and MBC in Mediterranean agroecosystems in the Pacific Northwest (Collins et al. 1992; Machado et al. 2011; Ghimire et al. 2019). They are also frequently tilled; breaking open macroaggregates, increasing microbial access to soil C, injecting oxygen, and driving increased metabolic activity (Six et al. 2002). While this can be a major driver of critical nutrient cycling in cropping systems, it may also contribute to the lower soil C and reduced prevalence of macroaggregates found in our study. Further, residue C inputs are predominately protected from degradation through occlusion inside the very aggregates that tillage disrupts (Six et al. 2002; Jastrow et al. 2006; Schmidt et al. 2011). Several other studies have similarly shown 50% or greater reductions in MWD in cultivated fields relative to adjacent hedgerows or native forest (Gupta et al. 2009; Emadi et al. 2009; Thiel et al. 2015).

Low soil C and low crop diversity may have contributed to the lower MBC and MBN and in turn, the lower potential enzyme activity in cultivated fields. Conversely, in hedgerows, there is high litter deposition, abundant fine roots in the ground year-round (potentially sloughing off and releasing a constant supply of exudates), and little physical disturbance, contributing to higher soil C, MBC, Agg_{ma} , and MWD (Lynch and Bragg, 1985, Maaß et al., 2015, Vezzani et al., 2018). Recent studies have shown that root C has a higher carbon use efficiency, or amount of C stored per unit carbon consumed, contributing to greater gains in microbial biomass, which may be more readily retained as SOM and/or stabilized on mineral surfaces (Rasse et al. 2005; Kong et al. 2011; Schmidt et al. 2011; Cotrufo et al. 2013, 2015). Furthermore, higher plant diversity has been shown to result in higher soil C, MBC, MBN, and potential enzyme activity (Lange et al. 2015; Brockerhoff et al. 2017; Isbell et al. 2017). While the lower microaggregate

percent in hedgerows relative to cultivated fields may seem counterintuitive, it is likely affected in part by the method, as microaggregates inside macroaggregates would be included in the total weight of stable macroaggregates. In cultivated fields, tillage frequently disrupts macroaggregates, but microaggregates can persist, whereas in hedgerows the lack of disturbance allows for macroaggregate formation to precede unimpeded with microaggregates forming first and providing a nucleus for macroaggregate formation (Six et al. 2002). Saha et al. 2012 similarly found greater microaggregate concentration under agricultural fields relative to adjacent agroforestry systems.

Our results did not indicate differences in the ratio between MBC and soil C or potential enzyme activity and soil C by management type, suggesting that observed differences in microbial biomass and potential enzyme activity by management types are largely due to increased total soil C, rather than major shifts in C-cycling processes. The relationship between BG:MBC and CEL:MBC, however, did differ by management type, suggesting differentiation in microbial/metabolic activity in these two systems. Hedgerow litter likely has greater diversity in quality and higher lignin and cellulose content, which could lead to increased enzyme production per unit biomass (Mungai et al. 2005, Yadav et al. 2008). Aboveground litter accumulation and reduction of tillage may be larger drivers of improved soil health than root activity, since there was less accumulation of MBC and soil C in the 10-20 cm layer.

In other studies, soil health practices have been shown to positively impact soil properties (Idowu et al. 2008; Hargraeves et al. 2019; Norris et al. 2020) with afforestation typically leading to even greater improvements (Mungai et al. 2005; Udawatta et al. 2009; Smukler et al. 2010; Guillot et al. 2019). Here, hedgerows showed higher MBC, MBN, and MWD than other soil health interventions in Yolo Co. cropping systems (Fennimore & Jackson 2003; Burger 2003;

Bowles et al. 2015), but lower than native stands of oak and shrub communities in the region (Young-Mathews et al., 2010; Smukler et al. 2010; Hodson et al., 2014), indicating that soil health variables may continue to improve. Other agroforestry studies in Mediterranean-type climates have shown a similar magnitude of difference between hedgerows and a no-tree control, with a 156-197% increase in MBC and a 161-211% increase in MBN, relative to 180% and 214% increases, respectively, in this study (Yadav et al. 2010; Guillot et al. 2019).

Despite higher levels of total C and N, MBC in our study, values were approximately 20-60% lower in comparison to organically and conventionally managed plots at a nearby long-term research station (Lazicki et al. 2021). Sampling occurred in April of the same year in both studies and on similar soils, but moisture content was not reported, limiting ability for comparison. POXc was similar in organic and conventional fields to cultivated fields in our study but was nearly 1.5-2x greater under hedgerows (Lacizki et al. 2021). In an organic farmscape in Salinas, CA, Smukler et al. 2010, found similar levels of aggregate stability with MWD higher in hedgerows (2.1 mm) than all other habitats on farm (~0.9mm).

Significant improvements in Agg_{ma} , MWD, and INF-2'' under hedgerows, indicate that hedgerows are beginning to have a positive impact on overall soil structure. Increases in soil C, Agg_{ma} and MWD may be contributing to the faster infiltration rates observed under hedgerows, due to increases in macropores, greater connectivity of pores, and/or limited slaking and clogging of pores after initial wetting often found with cessation of tillage (Benjamin 1993; Van Eerd et al. 2014). At the time of sampling, recent tillage to prepare beds, could have increased slaking and clogging of pores after initial application of water. Improvements in infiltration rate and aggregate stability are often preceded by increases in soil C and microbial biomass/activity, which we also observed here.

At 0-10 cm, there was only a significant interaction effect (of management and soil type) on a few variables, most of which are commonly considered or associated with inherent soil properties (i.e. pH, MWD, and BD). For instance, MWD was highly correlated with silt and clay content, which is known to play a major role in aggregate formation (Six et al. 2002; Totsche et al. 2018). The minimal differences in effect sizes by soil type (for either management type) could speak to the broad capacity of hedgerows to enhance surface soil health and the impact of microclimate (Sanchez et al. 2010; Kanzler et al. 2019; Veste et al. 2020) in these otherwise harsh climatic conditions. It may also be a result, however, of limited differences in soil types included in the study. Yolo, Brentwood, and Capay are all formed from the same parent material, have similar dominant mineralogy, pH, and degree of weathering. Further, while these soil types exhibit a large range in clay percentage, silt + clay content does not vary significantly between Yolo and Capay soils. At 10-20 cm, there was an interaction effect on GWC, POX_c, and EOC, all of which were attributed to differences in Corning. Corning was the only soil type in the study to differ in terms of parent material, dominant mineralogy, and pH; all of which are major determinants of response to soil health indicators (Rousk et al. 2009; Fine et al. 2017; Waldrop et al. 2017; Rassmussen et al. 2018; Adeyolanu et al. 2015) and specifically, microbial community structure and activity (Girvan et al. 2003; Bossio et al. 2005; Kuramae 2012).

Based on the magnitude of differences, a composite of variables may provide value in distinguishing differences in soil health. MBC, BG, and CEL appeared to be the most sensitive biological indicators to land use change. Total C, total N, and POX_c were the most sensitive chemical indicators; and MWD, INF-2", and SSH20 were the most sensitive physical indicators. Similarly, a multi-year study at a nearby long-term research station found that total C, total N, MBC, POX_c, and MWD (when corrected for soil texture) were some of the most sensitive soil

health indicators (Lazicki et al. 2021). Based on the high variability in INF-2” and SSH20 both in this study and others (Leonard & Andrieux 1998; Kılıç et al. 2004; Haws et al. 2005), they may not be effective indicators, except insofar as they provide value to the grower as simple, cost-effective in-field indicators (Andrews et al. 2004, Carlisle et al. 2016). It is well documented that metrics of soil health tend to correlate with soil C (Blanco-Canqui & Benjamin 2013; Karlen et al. 2019; Nunes et al. 2020; Wu & Congreves 2021) and that SOM is central to soil health and ecosystem function (Weil & Magdoff 2004; Lal et al. 2016). Indeed, many of the indicators measured correlated strongly with soil C (i.e. MBC, POXc, EOC, MWD, etc.), especially under hedgerows. Nonetheless, PCA indicated that a combination of multiple indicators was necessary to explain a substantial portion of the variation in the dataset and thus, are important in understanding soil health.

Conclusions

Our regional survey found that implementing the four principles of soil health management, through revegetation of field margins with hedgerows, had a marked impact on soil health on common soil types in the Sacramento Valley, California. In the surface 0-10 cm, most of the commonly used soil health indicators were sensitive to the different management. Only pH, SH10, and BD (which may be more associated with inherent soil properties) and EON (which may be more spatiotemporally variable and associated with management) were not different between systems. At 10-20 cm, variability of biological indicators increased and overall differences between systems were less apparent.

While many of the soil health variables correlated strongly with soil C, it is evident that no single indicator is sufficient to distinguish between cultivated fields and hedgerows. Although careful consideration should be given as to the desired goals/outcomes of a given system or landscape feature on-farm, the consistent positive effects of hedgerows on soil health variables may help elucidate the “Soil Health Gap” and provide an effective reference as to the biogeochemical potential of a given landscape to achieve soil health. The ability of soil health management practices in cultivated row crop systems to narrow the gap between managed and unmanaged/restored areas should be further explored across a range of edaphoclimatic contexts.

Vegetated marginal areas on farms (road edges, creek and slough berms, ditches) in the area have been found to contribute to a broad set of ecosystem services, (e.g., pollination and pest control, wildlife habitat, nitrate removal, woody C sequestration). The observed improvements in hydrological function under hedgerows in this study, may further support their use on marginal lands. Potential tradeoffs for agricultural production such as edge effects from competition for light and nutrients should be further investigated, as well as the potential for cultivable hedgerows species.

3.7 Acknowledgements

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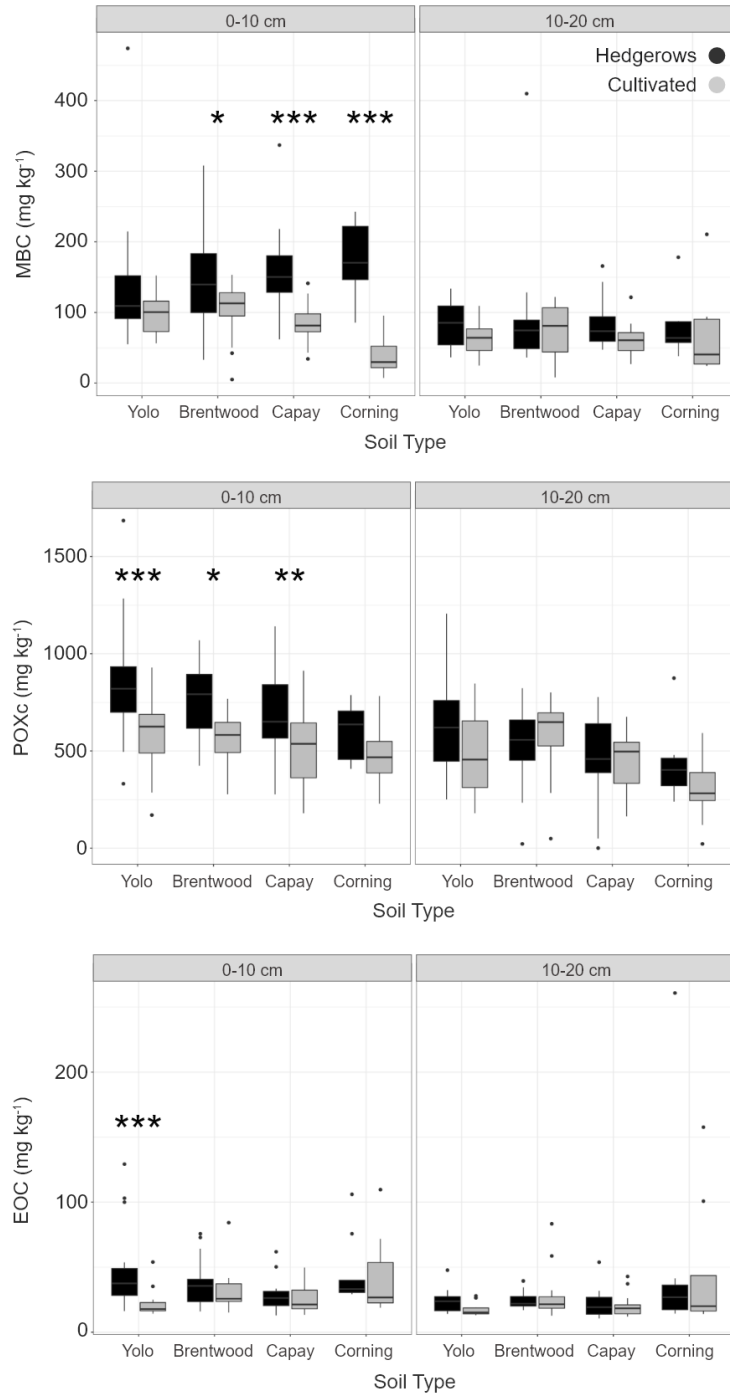
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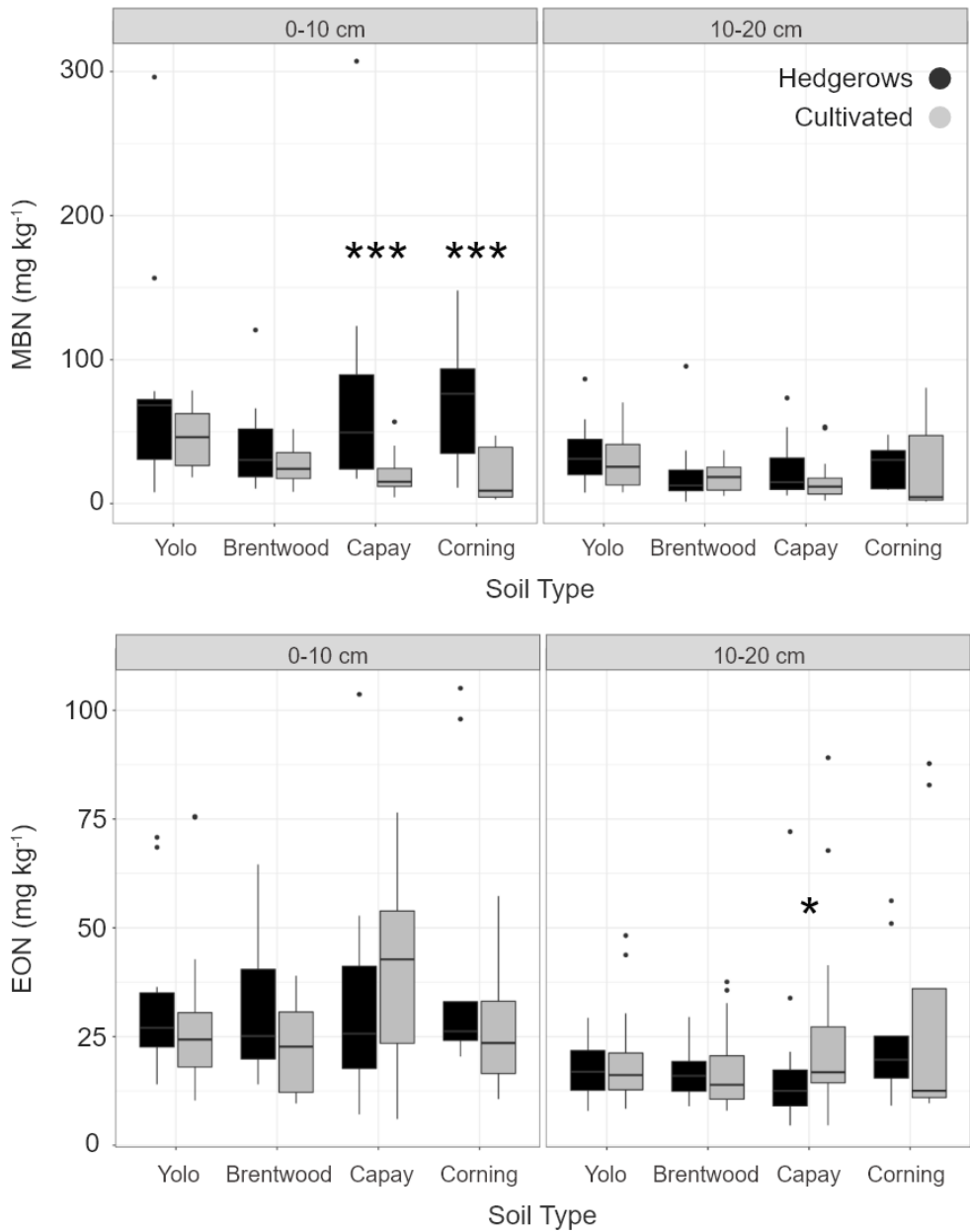
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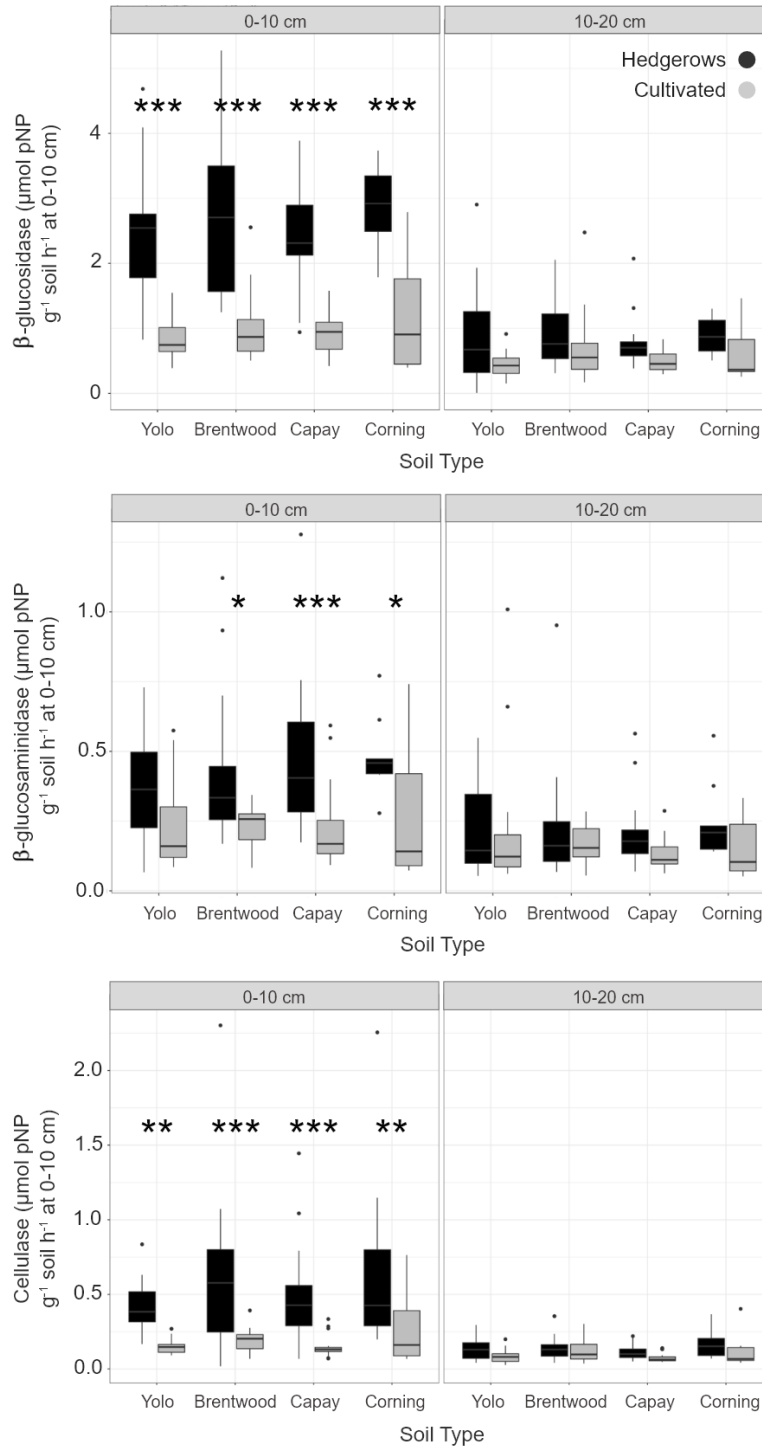
3.9 Supplemental Information



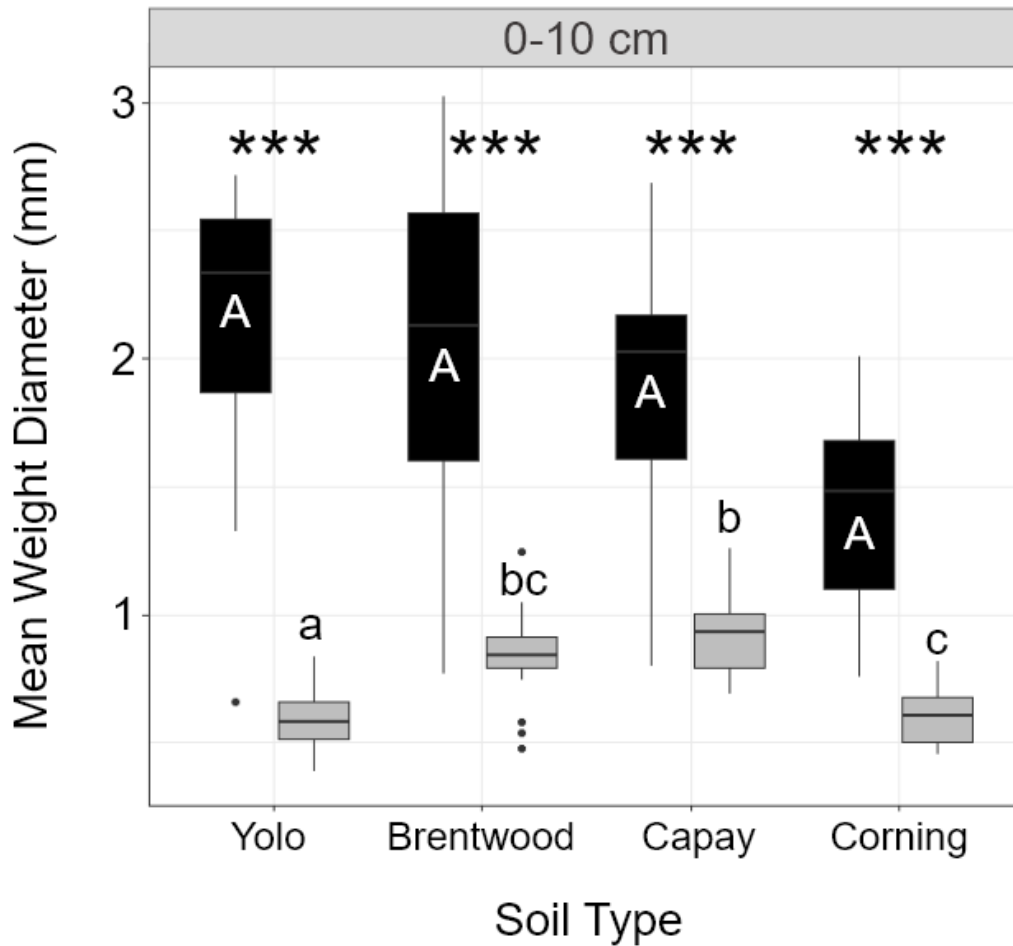
SI Figure 3.1. Box plots of different soil carbon fractions by soil type at 0-10 and 10-20 cm depths, including a) microbial biomass carbon (mg kg⁻¹), b) permanganate oxidizable carbon (mg kg⁻¹), and c) KCl extractable organic carbon (mg kg⁻¹). Horizontal lines represent mean values, bars represent standard error. Asterisks represent significant differences between management types at * = P < 0.05, ** = P < 0.01, *** = P < 0.001.



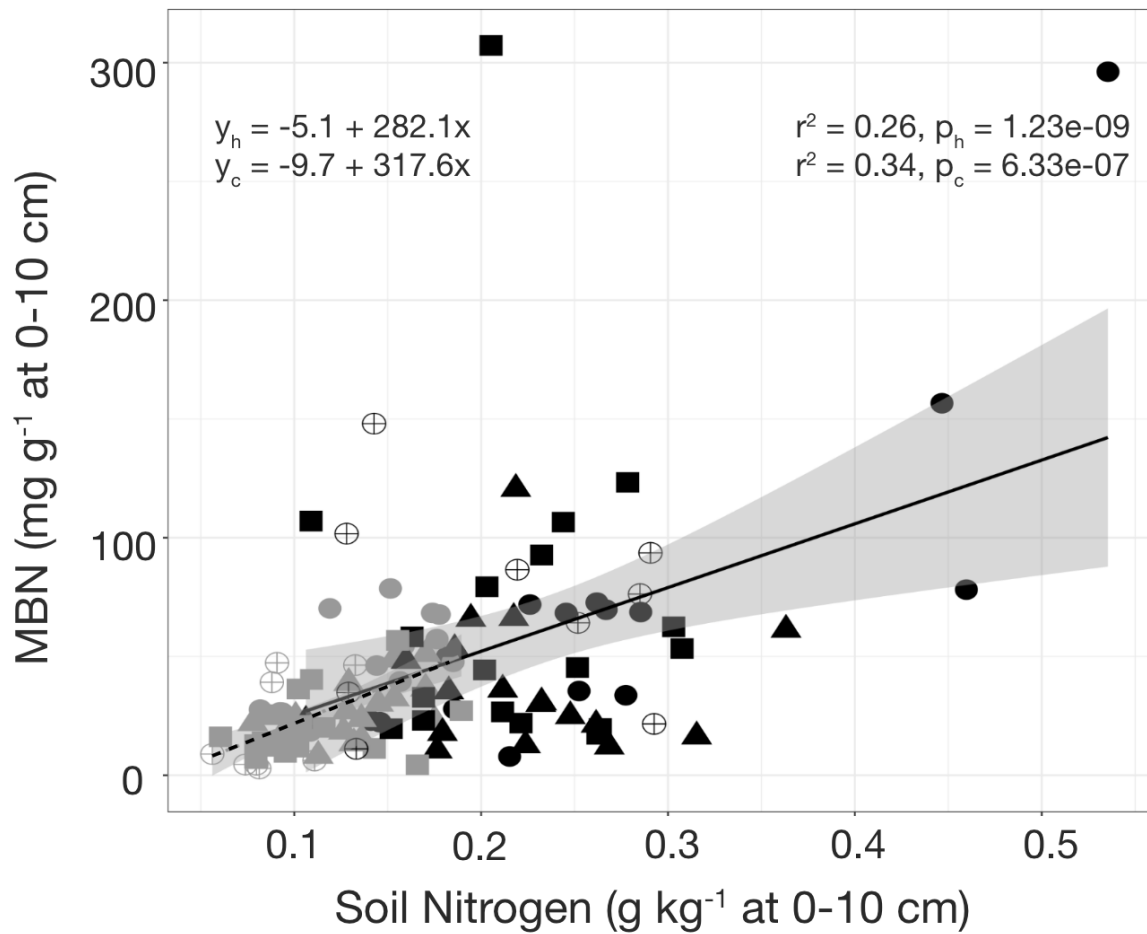
SI Figure 3.2. Box plots of different soil nitrogen fractions by soil type at 0-10 and 10-20 cm depths, including a) microbial biomass nitrogen (mg kg⁻¹) and b) KCl extractable organic nitrogen (mg kg⁻¹). Horizontal lines represent mean values, bars represent standard error. Asterisks represent significant differences between management types at * = P < 0.05, *** = P < 0.001.



SI Figure 3.3. Box plots of soil enzymes by soil type at 0-10 and 10-20 cm depths, including a) β -glucosaminidase ($\mu\text{mol pNP g}^{-1} \text{ soil h}^{-1}$) and b) β -glucosidase ($\mu\text{mol pNP g}^{-1} \text{ soil h}^{-1}$), and c) cellulase ($\mu\text{mol pNP g}^{-1} \text{ soil h}^{-1}$). Horizontal lines represent mean values, bars represent standard error. Asterisks represent significant differences between management types at * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$.



SI Figure 3.4. Box plots of Mean Weight Diameter (mm) by soil type at 0-10 cm. Black boxes represent hedgerows, while gray boxes represent cultivated fields. Hedgerow soils with the same uppercase letters and cultivated soils with the same lowercase letters are not significantly different at $P < 0.05$. Asterisks represent significant differences between management types at *** = $P < 0.001$. Horizontal lines represent mean values, bars represent standard error.



SI Figure 3.5. Correlation between total soil nitrogen concentration (g kg⁻¹) and microbial biomass nitrogen (mg kg⁻¹) at 0-10 cm depth. P-values represent ANOVA results for each treatment individually. Analysis of covariance (ANCOVA) indicated no significant difference in the relationship by management type.

SI Table 3.1. Results from the first six principal components of a principal component analysis (PCA) of all measured variables from 0-10 cm depth (n=20). Analysis was conducted on all sites without separation by soil type.

0-10 cm depth						
	PC1	PC2	PC3	PC4	PC5	PC6
Eigenvalues	2.41	1.68	1.47	1.32	1.19	0.90
% Variance	28.90	14.20	10.77	8.70	7.04	5.86
Cumulative % Variance	28.90	43.10	53.87	62.57	69.61	75.47
Factor Loading						
Carbon	-0.359	-0.016	0.034	-0.092	0.028	-0.317
MBC	-0.347	-0.012	0.058	-0.101	-0.120	0.197
β -glucosidase	-0.346	-0.121	0.030	0.142	-0.149	0.101
Nitrogen	-0.335	0.027	0.045	-0.175	0.094	-0.373
POXc	-0.286	0.061	-0.065	-0.075	0.232	-0.340
β -glucosaminidase	-0.285	-0.144	-0.064	0.071	-0.265	0.317
MBN	-0.270	0.029	0.229	-0.218	0.189	0.228
cellulase	-0.260	-0.191	0.037	0.212	-0.352	0.201
EOC	-0.188	-0.150	0.165	-0.006	0.450	0.085
Macroaggregates	-0.163	0.210	0.091	0.440	-0.187	-0.231
EON	-0.146	0.084	0.245	-0.073	0.341	0.422
GWC	-0.142	0.333	0.114	-0.274	-0.312	0.073
pH	-0.087	0.456	-0.040	-0.105	0.023	-0.069
Clay	-0.009	0.492	-0.009	0.154	-0.121	0.195
Infiltration wet	0.200	0.059	0.523	-0.157	-0.091	-0.019
Infiltration dry	0.148	-0.095	0.487	-0.240	-0.136	-0.033
Microaggregates	0.140	0.060	-0.336	-0.453	-0.100	0.197
Surface hardness	0.105	0.078	0.428	0.164	-0.172	-0.175
Bulk Density	0.095	0.140	0.083	0.451	0.360	0.207
Sand	0.053	-0.496	0.083	0.011	-0.092	-0.052

SI Table 3.2. Results from the first six principal components of a principal component analysis (PCA) of all measured variables from 10-20 cm depth (n=20). Analysis was conducted on all sites without separation by soil type.

10-20 cm depth						
	PC1	PC2	PC3	PC4	PC5	PC6
Eigenvalues	2.00	1.77	1.32	1.09	1.08	1.02
% Variance	25.05	19.58	10.96	7.49	7.34	6.51
Cumulative % Variance	25.05	44.63	55.59	63.08	70.42	76.93
Factor Loading						
Carbon	0.433	-0.055	0.130	-0.111	0.145	-0.228
Nitrogen	0.416	-0.107	0.069	-0.220	0.159	-0.146
β -glucosidase	0.379	0.210	0.030	0.008	-0.085	-0.088
MBN	0.288	0.090	-0.193	-0.482	-0.179	0.184
MBC	0.280	0.129	-0.198	-0.119	-0.382	0.336
cellulase	0.260	0.323	-0.047	0.152	-0.194	0.141
POXc	0.246	-0.049	0.291	0.353	0.267	-0.065
pH	0.220	-0.371	0.095	0.163	0.112	0.156
GWC	0.149	-0.339	-0.087	0.040	0.212	0.407
β -glucosaminidase	0.139	0.148	-0.029	0.661	-0.219	0.290
EON	0.102	0.081	-0.544	0.049	0.490	-0.003
Clay	0.057	-0.443	-0.230	0.108	-0.217	-0.050
EOC	0.050	0.253	-0.495	0.160	0.310	-0.123
Subsurface hardness	-0.247	-0.073	-0.053	-0.161	0.213	0.571
Sand	-0.161	0.460	0.121	-0.021	0.119	0.008
Bulk Density	-0.117	-0.228	-0.430	0.119	-0.334	-0.360