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UNIVERSITY OF CALIFORNIA, MERCED

Rooted In Health: Significant Benefits of Compost
and Cover Crops to Soil Health in Nut Orchards in
the Central Valley of California

by

Alexia Ila Cooper

Professor Asmeret Asefaw Berhe

Professor Sat Darshan S Khalsa

Professor Jennifer Pett-Ridge

Professor Rebecca Ryals

2024

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The Dissertation of Alexia I. Cooper is approved, and it is acceptable in quality and form for publication on microfilm and electronically:

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University of California, Merced

2024

DEDICATION

Soil

For her. For you. For us.

This work is dedicated to the soil—the giver of life, the foundation that sustains our world. To those who have been called to action for the sake of our shared future: the soil and its vast potential for climate mitigation have given me hope. I pursue this work so that my future children can grow up in a world with healthy soil, clean water, and pure air—a legacy rooted in resilience and care for our planet.

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LIST OF SYMBOLS

BD: Bulk Density

C: Carbon

CEC: Cation Exchange Capacity

CH₄: Methane

CO₂: Carbon Dioxide

EC: Electrical Conductivity

ESM: Equivalent Soil Mass

FD: Fixed Depth

MBC: Microbial Biomass Carbon

MBN: Microbial Biomass Nitrogen

N: Nitrogen

N₂O: Nitrous Oxide

POXC: Permanganate Oxidizable Carbon

SOC: Soil Organic Carbon

SOM: Soil Organic Matter

VWC: Volumetric Water Content

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To my family, whose love has profoundly shaped my journey into soil science: my mother, Tammie, whose early teachings and exuberant love for the natural world fostered my curiosity and critical thinking which has helped me tremendously throughout this program. To my father, Lewis, whose steady support and belief in me have been a critical foundation, encouraging me to persevere and reminding me of my ability to overcome any challenge. And to my brother, Cole, whose encouragement has been a source of strength. His insightful mind and grounded perspective have been guiding lights, helping me find resilience in dark times and fueling my determination to keep moving forward.

To my community in Mariposa, thank you for grounding me and giving me the balance I needed amidst the demands of my program. The days spent by the river, climbing in Yosemite, and late nights gazing at the stars were vital for keeping my heart and mind refreshed.

To Jeff, my project PI and now dear friend—thank you for turning even the most grueling days in the field into moments I looked forward to. Your laughter, kindness, and lighthearted energy made the long hours bearable. I am grateful for all those days spent out in the orchards and am so glad to have had you by my side through it all. Here's to more projects and adventures together in the future!

To my advisor, Becca, thank you for seeing my potential from the beginning. Your wisdom, kindness, and belief in my abilities allowed me to grow into the soil scientist I am today. And to my committee members, JP, Sat, and Asmeret, thank you for your guidance, insights, and dedication, all of which have been invaluable on this journey. This is only the beginning of my path, and I am deeply grateful to have had each of you by my side.

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CURRICULUM VITA

Alexia Ila Cooper

EDUCATION

University of California Merced — PhD Candidate, Merced, CA **2020 - 2024**

Ph.D. in Environmental Systems, Dissertation Title: “ Rooted Health: Benefits of Compost and Cover Crops to Soil Health in Nut Orchards,”
School of Engineering

Western Colorado University — M.E.M., Gunnison, CO **2018 - 2020**

Sustainable and Resilient Communities
School of ENVIS Scholarship | Mountaineer Fellowship | GPA 4.0
Advisor: Dr. Jennie DeMarco

Westminster University — B.S., Salt Lake City, UT **2013 - 2017**

Environmental Science
Presidential Scholarship | Honor Roll | GPA 3.65
Focus on environmental writing, political science, and agroecology.

PROFESSIONAL EXPERIENCE

Graduate Researcher — UC Merced | Conservation Innovation Grant **Jun’21
Dec ‘24**

As a graduate student researcher, I designed and implemented the sampling plan for a five-year project monitoring the effects of integrative soil health practices across eight orchards in California's Central Valley.

Teaching Assistant — UC Merced | Agroecology ESS 172 **Aug ‘21
Dec ‘21**

In this role, I helped to prepare discussion section lessons and activities, and graded exams and homework. I organized and held workshops lead by the undergraduate student in the on-campus garden. I was available to field questions by undergraduates about course material that covered soil science, ecology, and agricultural practices.

Graduate Researcher — UC Merced | Agroecology Lab **Aug ‘20
Aug ‘21**

As a field technician on a composting research project for the Healthy Soils program, in Central California I monitored the flux of GHGs i.e., CO₂, NH₃, N₂O and water vapor with a Picarro analyzer from grassland soils. In the soil lab, I worked with soil samples processing them for a

suite of soil health parameters. I mentored undergraduate students, and interns in the lab on the basics of soil science and field sampling.

Graduate Mentor — Western Colorado University | Organics Guild **Oct '18 - May '20**

In this leadership role, I mentored undergraduate club members to achieve their desired garden project outcomes. I ensured the club moved towards efficiency and sustainability goals. During my leadership, we increased the quantity and quality of local food produced by the community. I provided expertise regarding permaculture practices, soil health, and companionship planting methods to increase garden resiliency in a cold climate. Responsibilities included: garden planting design, coordinating over 1,000+ internship hours, workshop planning, and garden classes.

Graduate Researcher — Western Colorado University | Soils Lab **Aug '18 – May '20**

As a graduate student researcher, I designed and initiated a long-term experiment on the effects of a single biosolid application in irrigated pastures in Gunnison, CO. This study involved weekly monitoring of soil moisture and a one-year sampling of soils to assess changes in soil carbon storage. In addition to soil sampling, biomass quadrats were used to identify species types present, which were then sorted into forbs and graminoids after harvest to measure total biomass.

FELLOWSHIPS & GRANTS

Traina Fellowship	\$1,000	2024
Provides support to researchers that study sustainable agriculture		
CONDESA Fellow	\$34,000	2022
NRT Trainee for nano-engineered soil C development and application.		
USDA Sustainable Agriculture Research and Education (SARE)	\$24,746	2019
Collaborative project with ranchers on biosolid compost amendments.		
Coldharbour Institute Graduate Research Grant	\$2,500	2019
Provided research funding for soil health amendments.		
Hayley Fund Western Colorado University	\$3,380	2019
Provided funding for Soil Science course at CSU along with soil moisture probe		

Mountain Club Foundation Graduate Grant

\$780.00 2019

Provided funding for continued research on compost.

RESEARCH & PROJECTS

University of California Merced | Jan 2021-23

Integrative effects of compost and cover crops on soil health indices in California orchards

As part of a NRSC Conservation Innovation Grant (CIG) grant, 8 orchards were selected for the implementation of integrated soil health practices of compost and cover crops. The influence of these practices on soil health indices, nitrogen dynamics, and microbial diversity was studied for 4 years.

Western Colorado University | Dec 2018

Biosolid compost amendments in arid temperate irrigated grasslands

Researching the effect from 5 cm of biosolid compost on irrigated pasturelands in Gunnison Colorado, on soil health, specifically the water holding capacity, plant productivity, species diversity, fungal relationships, and carbon sequestration. This project addressed an expressed community need to be more resilient to drought periods by improving the health of the soil.

Western Colorado University | Oct 2018

Empower Workshop

Project manager working on a Habitat for Humanity's grant from the Community Foundation to organize and hold a workshop on energy efficiency and conservation for the community of Gunnison. We taught energy efficiency winterization techniques and brought awareness to energy saving tactics attendees could implement at home, and provided 30 energy efficiency kits which included LEDs, programmable thermostats, and other energy saving products.

Western Colorado University | Sept 2018

Biomimicry Project

This was a research project where we developed a biomimicry product from the spruce beetle, *dendroctonus rufipennis*, outbreak in Colorado. This product would theoretically use the bark beetles actinomycete egestion as an anti-fungal paint to apply within homes to prevent the growth of black mold and support healthy building and people.

Westminster College | Apr 2017

Monitoring the Effects from Air Pollution on Plant Productivity and Health

Lead undergraduate research technician on Dr. Christy Clay's agricultural research on the effect of increased CO₂ and Ozone on plant health and biomass production along the Wasatch Valley at 6 sites. Trials were set up where genetically modified and heirloom varieties of beans, beets, onions, and tomatoes were compared to see their resilience to pests, and experienced effects from the air pollution.

Westminster College | Nov 2016

Unprotected & Protected Watershed Comparisons: Dog Presence Effect to Water Quality

Designed and implemented a water quality research project to determine how great of an effect dogs had on watersheds water quality. In this research we took water samples from non-watershed protected canyons that allowed dogs, and canyons that were protected watersheds and compared the growth of bacteria.

Westminster College | Nov 2016

Medicinal Permaculture Garden

A two-year project where I designed, organized, and implemented a demonstration garden on permaculture techniques. This spiral garden was built by students with my guidance, it was planted with perennial medicinal herbs that I grew in the on-campus greenhouse. This garden demonstrated how to build healthy soil, how to companion plant, and how to design a garden for an arid climate.

PUBLICATIONS

Hicks Pries, C. E., Ryals, R., Zhu, B., Min, K., **Cooper, A.**, Goldsmith, S., Pett-Ridge, J., Torn, M., & Berhe, A. A. (2023). The Deep Soil Organic Carbon Response to Global Change. *Annual Review of Ecology, Evolution, and Systematics*, 54(1), 375–401. <https://doi.org/10.1146/annurev-ecolsys-102320-085332>

Cooper, A., & DeMarco, J. (2023). Composted biosolids amendments for enhanced soil organic carbon and water storage in perennial pastures in Colorado. *Agriculture, Ecosystems & Environment*, 347, 108401. <https://doi.org/10.1016/j.agee.2023.108401>

Kutos, S., Stricker, E., **Cooper, A.**, Ryals, R., Creque, J., Machmuller, M., Kroegar, M., & Silver, W. L. (2023). Compost amendment to enhance carbon sequestration in rangelands. *Journal of Soil and Water Conservation*, 78(2), 163–177. <https://doi.org/10.2489/jswc.2023.00072>

PRESENTATIONS

Triple Societies, San Antonio TX

Nov
2024

Compost and Cover Crops Influence on Soil Carbon Dynamics in California Orchard Systems

COMPOST2024, Daytona FL

Feb
2024

Compost and Cover Crops Influence on Soil N Dynamics in California Orchard Systems

Soil Health Workshop, Gunnison CO

Nov
2021

Soil health 101 invited presentation to landowners, ranchers, and students

Annual Meeting Society for Range Management, Denver CO

Feb
2020

Carbon Sequestration on Irrigated Pasturelands: Compost for Drought Resiliency

Land Manager Conference, Paonia CO

Dec
2019

Land Management as Climate Mitigation: Carbon Sequestration Potentials

Four Corners Symposium, Gunnison CO

Sept
2019

Innovative Land Management: Carbon Sequestration on rangelands in Colorado

TedX Westminster College, Salt Lake City UT

Apr
2016

Backyard Solutions to Global Problems

DISSERTATION ABSTRACT

Rooted In Health: Significant Benefits of Compost and Cover Crops to Soil Health in Nut Orchards in the Central Valley of California

Alexia Ila Cooper

Environmental Systems

This dissertation explores the impact of sustainable management practices, specifically the use of compost and cover crops, on soil health, soil organic carbon storage, and microbial communities in Californian nut orchard systems. This research addresses key questions surrounding the role of these practices in enhancing climate resilience and promoting long-term sustainability. Chapter 2 focuses on assessing soil organic carbon (SOC) stocks across conventionally managed almond, pistachio, peach, and walnut orchards, revealing significant variation in cumulative SOC ranging from $55 \pm 5.2 \text{ Mg C ha}^{-1}$ to $109 \pm 11.6 \text{ Mg C ha}^{-1}$. This variation is driven by factors such as orchard type, soil type, and irrigation methods. The chapter underscores the importance of deep soil sampling, as limiting sampling to surface soils would have underestimated SOC stocks by 60%. Method comparisons between the equivalent soil mass (ESM) and fixed depth (FD) approaches confirmed that ESM is preferable in systems where bulk density shifts are expected.

Chapter 3 examines the effects of compost and cover crops on soil health indicators in almond and walnut orchards over three years. Experimental plots received annual applications of 19 Mg/ha of yard waste and manure compost, while cover crops were seeded at a rate of 13.8 kg/ha. Compost and cover crops significantly improved microbial biomass carbon (MBC) and nitrogen (MBN), with MBC increasing by $573 \pm 98\%$ in alleys and $307 \pm 100\%$ in berms, while MBN rose by $487 \pm 95\%$ in alleys and $55 \pm 14\%$ in berms. Additionally, permanganate oxidizable carbon (POXC) increased by $133 \pm 23\%$, and short-term carbon mineralization rates were $70 \pm 5\%$ higher, indicating enhanced biological activity. Soil carbon content doubled, leading to an average SOC of $60.7 \text{ Mg C ha}^{-1}$ across sites by the end of the study, alongside improvements in wet aggregate stability and soil structure.

Chapter 4 delves into the effects of these management practices on soil microbial diversity and structure, focusing on 16S and ITS sequencing of microbial communities in almond orchards. Soil samples were collected at four depth intervals (0-10 cm, 10-30 cm, 30-50 cm, and 50-100 cm) to assess how depth and management influence microbial communities. The results showed that compost and cover crops increased the abundance of beneficial bacterial and fungal phyla involved in organic matter decomposition and nutrient cycling. The study also emphasized that both soil depth and management practices must be considered to fully optimize microbial-plant interactions. Together, these findings highlight the critical role of compost amendments and the planting of cover crops in improving soil health, increasing carbon storage, and fostering resilient microbial communities in orchard systems. In conclusion, Chapter 5 discusses the feasibility of scaling these practices across California, limitations, and key takeaways to support management of these perennial systems for improved soil health, carbon storage, and agroecological resilience.

CHAPTER 1: INTRODUCTION

California's Central Valley is a critical hub for U.S. agriculture, producing over 250 crops and generating over \$17 billion in annual agricultural revenue (CDFA, 2020). The region is a leading producer of various nut crops, including almonds, walnuts, and pistachios, contributing significantly to the nation's food supply and generating substantial agricultural revenue (NASS, 2023). Almonds alone account for over \$3.5 billion in annual revenue, underscoring the Central Valley's critical role in domestic and global markets (California Almond Board, 2020; CDFA, 2023). The combination of fertile alluvial soils, a favorable Mediterranean climate, and extensive irrigation infrastructure has positioned it as the heart of the nation's food production. Yet, the success of California's agricultural sector is intertwined with practices that have exerted substantial pressures on the soil, leading to degradation that threatens the long-term sustainability of these systems (Fenster et al., 2021). The intensive nature of farming in this region, characterized by high input use and monoculture cropping systems, has contributed to a decline in soil health, undermining the resilience of these agroecosystems in the face of mounting climate challenges (Flint et al., 2018).

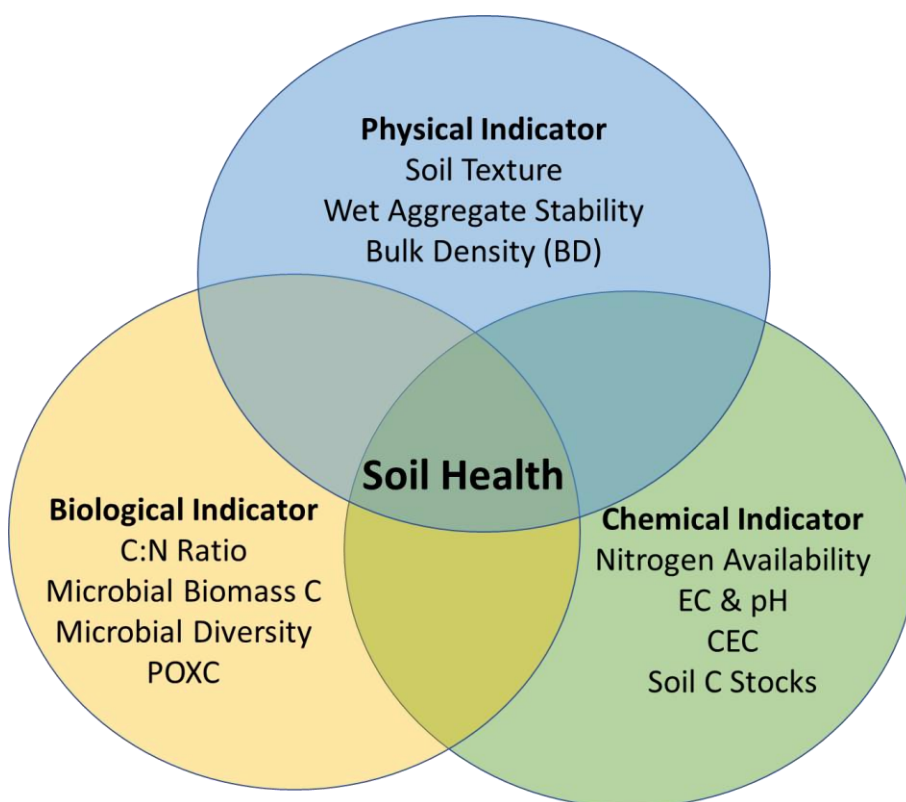


Illustration: 1-1 Soil's physical, biological, and chemical components, through their respective indices, shape the functioning of soil systems, with soil health at its nexus. Based on indicators from NRCS Soil Health.

As the challenges to sustain agricultural productivity grow, it is essential to consider the underlying factors that contribute to these issues, particularly the concept of

soil health. Soil health is a multidimensional concept that integrates physical, chemical, and biological properties to assess the soil's ability to function as a living ecosystem (Tahat et al., 2020), see Illustration: 1-1. Healthy soils support plant productivity, biological diversity, and water regulation while enhancing resilience against climate stresses (Lehmann et al., 2020). In the Central Valley, conventional orchard management often involves practices such as maintaining bare ground with herbicide use and heavy machinery, which degrade these critical properties by causing soil compaction, topsoil erosion, chemical pollution, and nutrient depletion (Acir et al., 2022). Soil compaction affects water infiltration and root penetration, while a decline in organic matter and soil structure exacerbates erosion and reduces soil resilience. Decreased soil health drastically increases the vulnerability of these systems to drought and intense rain events, which are predicted to increase in severity with climate change.

One element of soil health is soil organic matter (SOM), the key energy source for microbes in the soil. SOM plays a pivotal role in maintaining soil structure, enhancing water retention, and promoting nutrient cycling (Schjønning et al., 2018). Microbial communities process SOM into various forms of soil organic carbon (SOC) with differing resistance to decomposition (Cotrufo et al., 2019). Yet, in many Central Valley orchards, the cycling of SOM has been disrupted due to management practices that leave bare alley floors and limit soil inputs, leading to significant carbon depletion. A majority of SOC might have been lost before the planting of orchards, when most of the Valley was utilized as annual croplands such as corn or soy— systems that relies heavily only tillage (Spohn et al., 2023).

The depletion of SOM over time is particularly concerning within agricultural systems with minimal organic inputs (Oades, 1988). In perennial orchard systems, the lack of regular organic matter replenishment via plant residues has can result in a significant decrease in SOM retention (Xiang et al., 2022). This decline is directly linked to SOC levels, as approximately half of SOM is composed of SOC (Oldfield et al., 2019). Therefore, maintaining or increasing the retention and stabilization of SOM is critical for preserving SOC stocks and overall soil health. In Mediterranean orchards, previous studies in olive and peach found that SOC losses reached 0.20 to 0.31 Mg C/ha/yr with intensive conventional practices (Gómez et al., 2011; Montanaro et al., 2017). This depletion has far-reaching implications, not only for soil productivity but also for the role these soils can play in climate mitigation (Gelaye & Getahun, 2024). Rebuilding SOM and SOC is essential for improving soil health and restoring these systems' ability to support plant growth, mitigate climate change, and enhance resilience against environmental stresses (Javadinejad et al., 2021).

This brings us to the broader significance of SOC in the context of climate change. As the effects of climate change intensify, SOC plays an increasingly vital role in buffering orchard systems from environmental extremes (Hessen & Vandvik, 2022). SOC enhances water retention and reduces the risk of drought stress, while improving overall soil structure, nutrient availability, and biological activity (Flint et al., 2018). Thus, addressing SOC depletion is crucial, and the implementation of sustainable soil management practices, such as compost and cover cropping, offers a pathway to restore depleted SOC and enhance the soil's ability to sequester carbon (White et al., 2020).

These practices provide an opportunity to simultaneously mitigate climate change and build resilient orchard ecosystems (Azim et al., 2018)

This dissertation aims to explore how compost and cover crops can serve as effective soil health management practices in Central Valley orchards. Through the annual application of compost and the integration of cover crops, this research assesses changes in SOC, soil structure, microbial biomass, and nutrient availability. The results offer insights into the potential for these practices to improve soil health, sequester carbon, and enhance the long-term resilience of orchard systems.

Chapter Two explores the dynamics of SOC content in Central Valley orchard systems, highlighting the significant carbon depletion observed and its implications for soil health. The chapter discusses how conventional management practices have contributed to SOC loss and examines the potential for soil health management practices, such as compost application and cover cropping, to reverse this trend and restore soil carbon content. It calculates the potential of orchard soils to play a role in climate mitigation efforts in California with the support of soil health practices.

Chapter Three presents a detailed analysis of the effects of compost and cover crops on a suite of soil health indicators across physical, chemical, and biological components. The chapter reports significant improvements in soil structure, nutrient cycling, and microbial activity, demonstrating the effectiveness of these practices in enhancing soil health. It also considers the interactions between these indicators, demonstrating the synergistic effects of these practices in enhancing overall soil function.

Chapter Four focuses on the biological component of soil health, particularly the impact of compost and cover crops on the microbial communities in almond orchard soils. This chapter delves into the composition and diversity of bacterial and fungal communities, with a specific emphasis on how these communities vary with soil depth. The findings reveal significant shifts in microbial populations due to the implemented practices, with distinct differences between surface and subsoil microbial communities. These results underscore the importance of considering soil depth in soil health assessments and the potential for targeted management practices to enhance microbial-mediated soil functions.

Chapter Five concludes the dissertation by summarizing the critical role of compost and cover crops in improving soil health in orchard systems. The chapter emphasizes the substantial benefits observed in soil carbon content, structural stability, nutrient cycling, and microbial community dynamics, reinforcing the potential of these practices for long-term sustainability. It also highlights areas for further research, such as investigating the specific soil organic carbon fractions influenced by compost and cover crops, understanding the stabilization mechanisms of exogenous carbon, and exploring how to harness microbial communities for management goals. These research directions will be essential for refining management practices to maximize long-term carbon storage and enhance the resilience of orchard soils to future environmental challenges.

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CHAPTER 2: ASSESSING DEEP SOIL CARBON STOCKS IN CALIFORNIA ORCHARDS: INSIGHTS AND OPPORTUNITIES FOR CLIMATE MITIGATION

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HIGHLIGHTS

- Orchard soils exhibit a relatively low soil carbon (C) stocks, on average storing $36 \pm 0.9 \text{ Mg C} \cdot \text{ha}^{-1}$ in 0-30 cm depth.
- Deep soil sampling is imperative for comprehensively assessing the entire soil C stocks system: cumulative soil C to 1 m contained an average of $85 \pm \text{Mg C} \cdot \text{ha}^{-1}$.
- Sampling to only 30 cm would have overlooked 60% of orchard soil C to 1 m.
- Orchard type, soil type, and irrigation methods were significant factors impacting soil C stocks in orchards.
- The observed low soil C stocks in these orchards presents an opportunity for enhancing soil C stocks through soil health management practices.

ABSTRACT

This study provides a comprehensive assessment of soil carbon (C) stocks within conventionally managed orchard systems, investigating factors influencing soil C stocks across sites and depths. Across orchards of almond, pistachio, peach, and walnut in the Central Valley of California, we observed a significant range in cumulative soil C stocks to 1 m depth, with values spanning from $55 \pm 5.2 \text{ Mg C} \cdot \text{ha}^{-1}$ to $109 \pm 11.6 \text{ Mg C} \cdot \text{ha}^{-1}$, underscoring the influence of specific site conditions and management practices. Method comparison revealed the reliability of both the equivalent soil mass (ESM) and the fixed depth (FD) method in estimating cumulative soil C stocks but acknowledged the preference of ESM for long term monitoring where bulk density shifts are expected, especially in surface soils where there was a significant difference between methods. We highlight the importance of deep soil sampling, as limiting sampling to just surface soils (0-30 cm) would have underestimated orchard soil C stocks by 60%. We found that orchard type, soil type, and irrigation methods were significant factors impacting soil C stocks in orchards. Our study revealed the depleted nature of soil currently in orchards and discusses the substantial role they could play in soil C stocks for climate mitigation in California if soil health practices were implemented.

INTRODUCTION

Carbon (C) stocks within soils, and the potential for enhanced C sequestration attributed to different soil health management practices, can play a pivotal role in climate mitigation and agricultural adaptation. Deep soil C, often defined as soil below 30 cm, may be impacted by land management practices but is poorly characterized in agricultural systems (Hicks Pries et al., 2023). Orchards, as permanent cropping systems, are relatively understudied in terms of soil C dynamics and their potential for soil C sequestration (Gómez et al., 2022; Sharma et al., 2021). In the state of California, approximately 810,000 ha of bearing orchards exist, with almond orchards alone accounting for 607,000 ha (NASS, 2023). This extensive land coverage offers many opportunities for implementing innovative soil health practices to enhance soil C stocks. It is essential to establish the baseline levels of soil C within orchards, both in the surface (0-30 cm) and subsoil (30 - 100 cm), to better understand soil responses to future management strategies (Skadell et al., 2023). While there is existing research on soil C dynamics in orchard systems, the focus on deep soil C stocks in orchards, especially those in semi-arid climates, is limited. Investigating the factors that influence soil C stocks, such as soil texture, irrigation practices, and orchard type could provide valuable insights into enhancing soil C sequestration and mitigating climate change impacts in these regions.

Agricultural land management practices are an important factor affecting soil C sequestration and stocks (Lal, 2016; Yu & Song, 2023). Conventional orchard management practices such as keeping alleys bare, the use of large machinery, and high inputs of pesticides, fungicides, and chemical fertilizers can have negative effects on soil health (Acir et al., 2022; van Diepeningen et al., 2006). These practices contribute to key soil quality issues of compaction, low soil organic matter content, and poor water infiltration (Devine et al., 2021). Land management practices such as compost additions, cover crops, and reduced tillage have been shown to positively influence various soil health parameters in addition to promoting an increase in soil C (Chahal et al., 2020; Hodson et al., 2021; Lal, 2004). The effect of these practices on soil physical, chemical, and biological parameters in surface soils is well documented in annual cropping systems but lacking in permanent cropping systems (Norris & Congreves, 2018). Variations in those parameters at greater depths are also uncertain.

The average soil study does not sample to a depth greater than 24 cm (Yost & Hartemink, 2020, Baker et al., 2007; Davidson & Ackerman, 1993; Murty et al., 2002). This shallow sampling can promote assumptions about the effect of management practices along with incorrect projections about the influences of these practices on the whole soil profile (Tautges et al., 2019). To determine changes in soil C stocks in response to management, sampling should be conducted to the full depth of expected soil C changes which can extend beyond rooting depth or the direct zone of management practice (Olson & Al-Kaisi, 2015). Barriers to collecting soil at greater depths include the physical difficulty and high costs associated with deep soil sampling. Shallow sampling is often justified by assuming that the deeper soil horizons C content contains a larger and more persistent form of C that is considered stable, and less responsive to management (Z. Shi et al., 2020). Globally, the mineral associated organic matter (MAOM) portion

of soils contains ~60% of all C, and the more persistent larger MAOM pool is often found below 30 cm (Heckman et al., 2021). Shallow sampling could either under or overestimate the soil C present, and influence conclusions drawn about the effects of management on soil C. It is important to understand the current state of soil C stocks in the 1 m portion of the soil profile to better inform orchard managers about effective soil health management practices and climate mitigation market and policy mechanisms (Dynarski et al., 2020). Sampling to 1 m can also begin to capture soil C dynamics that occur beyond the active rooting zone, which is often deeper than 50 cm for orchards, as it has been found that management can influence subsoils (Pries et al., 2023; Y. Villa & Ryals, 2021).

The quantification of soil C stocks is often calculated via a fixed depth (FD) method which uses an equation that incorporates soil bulk density, depth, and total organic C concentration derived from elemental analysis from samples collected at fixed depth intervals. This method has been recognized as a "good practice" by the Intergovernmental Panel on Climate Change (IPCC), which supports protocols employing it for soil C calculations (Lehman et al., 2019). However, it has been observed that this method tends to systematically overestimate soil C stocks in areas with higher bulk density, leading to a bias in error for certain practices, such as no-till, where greater bulk density is often associated with management practices like minimal tillage (Poeplau et al., 2017). Variability in soil mass and compaction leads to an often-unequal basis between treatments to truly assess changes to soil C.

Some researchers argue for discontinuing the use of FD as a standard method in soil sciences and propose adopting the equivalent soil mass method (ESM) instead (Wendt & Hauser, 2013). Unlike FD, ESM method calculates soil C stocks by visualizing soil profiles in terms of soil mass layers rather than soil depth layers. This approach utilizes a fitted cubic spline to estimate cumulative soil C contents for specific reference soil masses within the profile and corrects extreme bulk density measurements (Slessarev et al., 2020; von Haden et al., 2020). By shifting the focus to soil mass, ESM offers a promising alternative that addresses the limitations associated with using bulk density as a sole determinant for soil C calculations. Very few studies have compared the difference between these two methods.

Our study quantifies soil C stocks in different orchard types and tests the differences between the two dominant methods to calculate soil C stocks. We hypothesize that current management practices of conventional orchards result in low soil C stocks. And that the methods of ESM and FD will have different outcomes to soil C stocks quantification. We anticipate that soil C content will be highest at topsoil depths (0-30 cm) and subsequently decline down to a depth of 1 meter. There will likely be an increase in C content around the rooting zone, which in these systems is at 30-50 cm depth. An increase in this area could be due to root exudation of photosynthate, which then accumulates as soil C (Brunn et al., 2022). We also expect that a majority of the soil C will be stored below 30 cm depth. We also anticipate that the berms have the potential to have greater soil C stocks than alleys, due to the location of the trees and decreased compaction. We anticipate that the most influential factors determining soil C pool size

will be texture, tree type, and tree age (Fenster et al., 2021, Dutta et al., 2018; McLauchlan, 2006).

MATERIALS AND METHODS

STUDY SITES

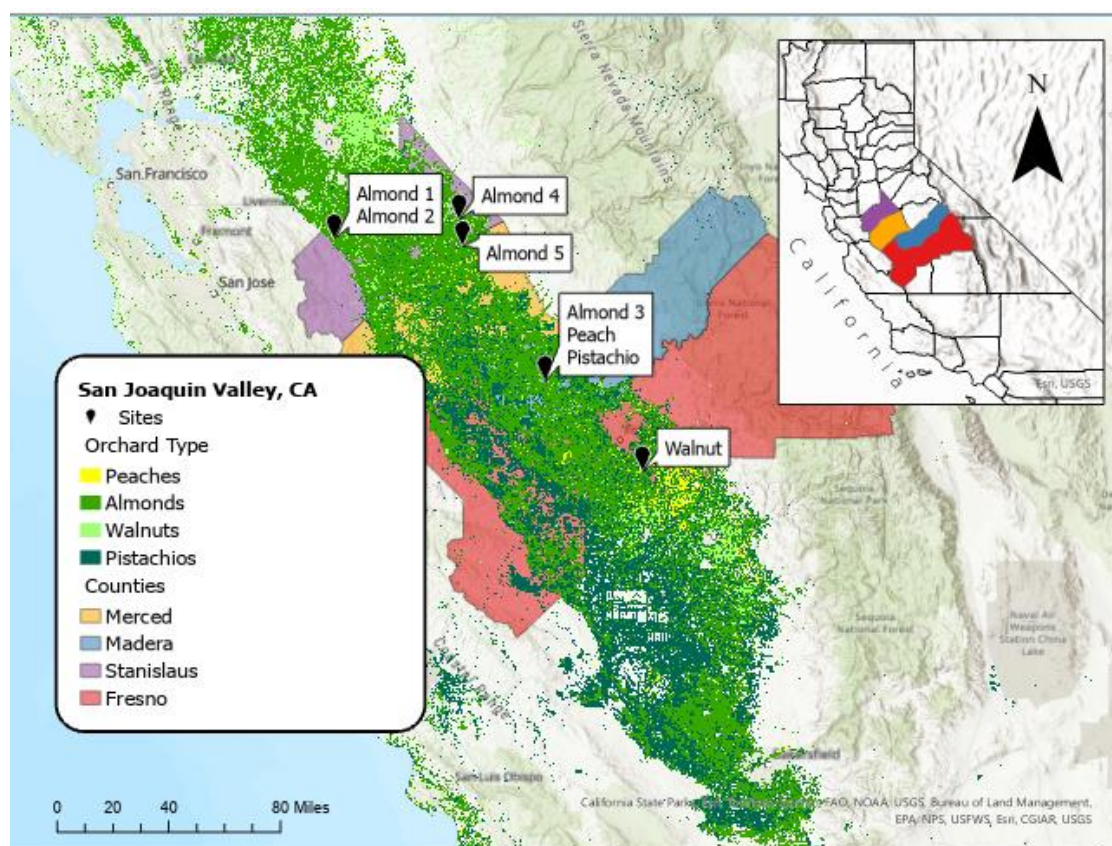


Figure 2-1. Map of study sites within the counties included in our study with overlay of orchard types across the Central Valley of California.

Table 2-1 Orchard site information for the eight sites included in this study, including age, irrigation type, and management prior to the study.

<i>Site Name</i>	<i>County</i>	<i>Orchard Age (yrs.)</i>	<i>Irrigation Type</i>	<i>Management</i>
Almond 1	Stanislaus	4	Drip	Conventional
Almond 2	Stanislaus	4	Drip	Conventional
Almond 3	Madera	15	Sprinkler	Conventional
Almond 4	Stanislaus	20	Sprinkler	Organic

Almond 5	Stanislaus	25	Drip	Conventional
Peach	Madera	15	Sprinkler	Organic
Pistachios	Madera	10	Drip	Conventional
Walnut	Fresno	25	Flood	Conventional

We sampled soil across eight sites in the San Joaquin Valley of California (Table 2-1) in June 2021. The sites included five almond orchards, along with one each of peach, walnut, and pistachios orchards. The San Joaquin Valley, the ancestral homeland of the Tejon, Kitanemuk, Yokuts, and Chamash Indigenous peoples of California, is situated south of the Sacramento-San Joaquin River Delta and is drained by the San Joaquin River. The San Joaquin Valley has a Mediterranean climate characterized by cool, wet winters and hot, dry summers. Mean annual temperatures in this region are approximately 14°C. The mean high temperature reaches approximately 25°C, while the mean low temperature is around 12°C (NOAA, 2021). Mean annual precipitation is 41 cm. Most of the precipitation occurs between November and March, with minimal rainfall during the summer months. The area is renowned for its agricultural activities, as more than half of the state's fruits, vegetables, nuts, and grains are cultivated here (Escriva-Bou et al., 2023).

SAMPLING DESIGN

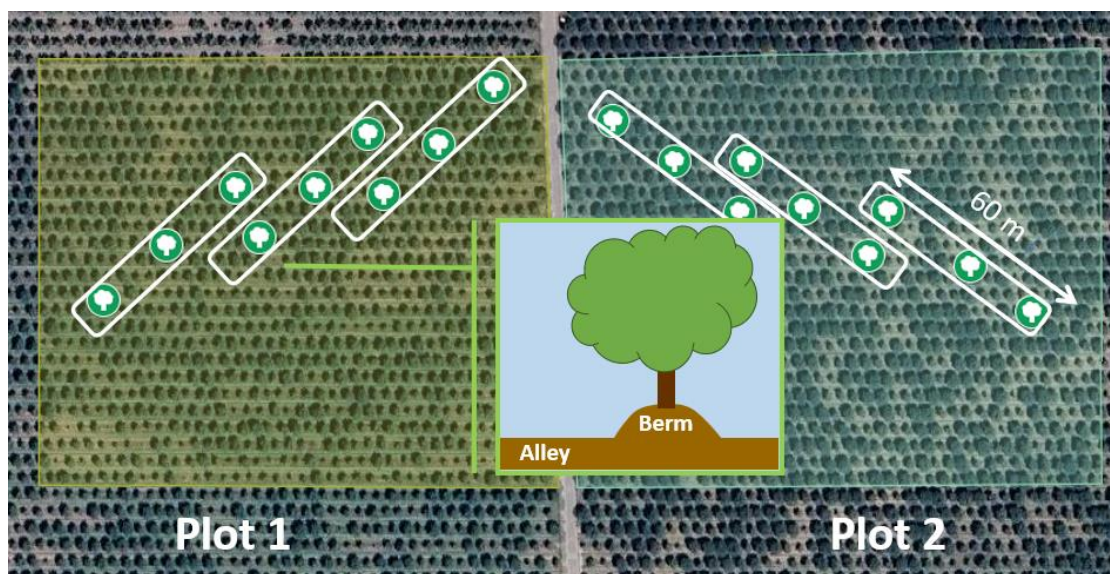


Illustration: 2-1. Site design for each of the 8 sites, satellite imagery and overlay of replicate transects within the plots for the pistachio site.

At each of the eight sites, two plots were designated within the orchards. Each plot was approximately 10 ha each. Within each subplot, three replicate transects of 60 m were established. The transects were arranged diagonally within the plots and contained three GPS marked trees (Illustration: 2-1). Transects started 10 m in from the edge of the

block to prevent potential edge effects. Soil samples were collected from both the berm and alley, defined as the location where tree trunks are planted and the spaces between the rows of trees, respectively. Soils were randomly sampled ~1 m away from each tree trunk along the berm and ~1 m away from the tree trunk into the alley. These locations were distinguished due to their different management practices, where alleys are subjected to heavy machinery traffic and herbicide use, while the berms are where water and fertilizer are applied near the base of trees.

Sampling was conducted to a depth of 1 m for one tree location in each transect replicate, while the other two tree locations were sampled down to a depth of 50 cm. The depth increments of 0-10 cm and 10-30 cm were collected using quantitative bulk density corers (inner diameter 5.74 cm), while deeper depths of 30-50 cm, 50-75 cm, and 75-100 cm were collected using a soil auger (diameter 6 cm) in the same sample hole. For each plot, we also excavated one soil pit, where we sampled down to 1 m, in 10 cm increments. These pits were used to correct the FD, whereas the ESM method used the auger data for its calculations. This sampling scheme resulted in a total of 1,128 soil samples, with 141 samples obtained per site, providing a broad regional assessment of orchard C stocks while capturing within site variability.

SOIL PHYSICAL PROPERTIES

For each site and its two corresponding plots, soil texture was determined via the hydrometer method (Gee & Bauder, 1986). Briefly, for each depth increment, 40 g of air-dried 2 mm sieved soil was mixed with 100 mL of sodium hexametaphosphate solution, let sit for 10 minutes, then mixed with an electric mixer for 5 minutes, then added to a glass sedimentation cylinder and brought to up a total volume of 1,000 mL. At the 30 second, 1 minute, 90 minutes, and 24-hour mark, a hydrometer was used to measure the density of the liquid and the corresponding liquid temperature was recorded. Afterwards, the measurements were then used to calculate the fractions of sand, silt, and clay via equations that were corrected for temperature. Soil pH was also measured on each sample. A 10 ± 0.1 g sieved soil g of soil was mixed with 20 ml of deionized water and then placed on an orbital shaker for 30 min. The soil slurry dilutions (1:2) were let to equilibrate uncapped for 10 minutes, then a calibrated pH meter (Mettler Toledo SS20, Columbus, OH) and EC meter (Mettler Toledo SS30, Columbus, OH) were used to measure pH and EC ($\mu\text{S}\cdot\text{ms}^{-1}$).

SOIL ORGANIC C AND N

Soil samples were air-dried, sieved at 2 mm, and pulverized by hand with a mortar and pestle. Carbonate presence was based on effervesce after addition of 4 N HCl to soil. No sites exhibited a response to the test for carbonates. Total soil C and N concentrations were measured on a Costech ECS 4010 CHNS-O Elemental Analyzer coupled to a Thermo Scientific Delta-V Plus continuous flow isotope ratio mass spectrometer at the Stable Isotope Ecosystem Laboratory of University of California, Merced.

Soil C stocks were calculated using the FD and ESM methods. For the FD, the bulk density in $\text{g}\cdot\text{cm}^{-3}$ was multiplied by percent soil C, then multiplied by the soil depth

increment, then converted to Mg ha^{-1} . Whereas, with the ESM method uses soil mass, volume, and the percent soil C and then uses a cubic spline of reference mass layers that was site specific (Wendt & Hauser, 2013). To calculate soil C stocks for each method, we used an R script developed by Von Haden et al. (2020). Data reported in the results is derived from the ESM method, unless otherwise stated. Comparisons of the two calculations can be found in Table 3. When reporting cumulative and depth soil C content, data were spatially weighted by berm and alley coverage. As berms account for approximately one-quarter of the area with an orchard, and alleys the remaining three quarters, the soil C stocks measured at each location was multiplied by each fraction of coverage and then summed to get a more accurate account of soil C stocks in the orchards.

MANAGEMENT VARIABLES

Information about management decisions, including tree and irrigation type, was collected from personal conversations with farmers and categorized into variables to test their effect on soil C stocks. Conventional management for our study is defined as orchard management that relies on chemical fertilizer inputs, along with pesticide and fungicide, in addition continually maintains bare alleys via tillage, scraping or herbicides. Whereas organic orchards included in our study are certified by USDA organic, meaning they do not use certain pesticides or herbicides. Four orchard types of almonds, peaches, pistachios, and walnuts were included in our study. Along with tree type, we gathered information on tree age, which ranged from 4 to 25 years old. There were various irrigation methods employed throughout the sites comprising of deep drip, regular drip, sprinklers, and flood irrigation.

STATISTICAL ANALYSIS

The study design incorporated several independent variables, including site, location (alley and berm), tree type, tree age, management, irrigation type, and soil type. The response variables of interest were soil C stocks and cumulative soil C stocks to 1 m. Prior to analysis, the normality of the data was assessed using a Shapiro-Wilk test, and if it was non-normal, tukey power of ladders transformation was applied. To investigate the differences in soil C stocks across sites, a linear mixed model was employed. Pairwise comparisons were conducted using a Tukey post hoc analysis, with significant interactions determined at a significant level of $p \leq 0.05$. Statistical analyses were carried out using R 4.1.0 (R Core Team, 2021), the *tidyverse* (v1.3.0; Wickham et al., 2019.) *ggplot2* (v3.3.3; Wickham, 2016) packages.

RESULTS

Table 2-2. Soil pH and electrical conductivity (EC) for the 8 sites that were included in this survey. The italicized *p* column indicates sites where pH or EC was significantly different between alley and berm ($p < 0.05$)

Site	pH			EC ($\text{uS}\cdot\text{cm}^{-1}$)		
	<i>Alley</i>	<i>Berm</i>	<i>p</i>	<i>Alley</i>	<i>Berm</i>	<i>p</i>

Almond 1	7.5 ± 0.1	7 ± 0.1	*	355.1 ± 64.5	569.3 ± 36.6	*
Almond 2	7.1 ± 0.2	7.3 ± 0.1		497.2 ± 57.9	505.1 ± 59.6	
Almond 3	7 ± 0.2	6.7 ± 0.2		475.6 ± 101.8	900.7 ± 318.7	
Almond 4	7 ± 0.1	6.7 ± 0.1	*	85.6 ± 5.1	96.2 ± 10	
Almond 5	7.6 ± 0.1	7.6 ± 0.1		163.6 ± 26.5	149 ± 43.1	
Peach	7.1 ± 0.5	6.9 ± 0.3		480.9 ± 143	698.5 ± 143	
Pistachio	6.6 ± 0.4	7.3 ± 0.1	*	446.9 ± 287.1	195.7 ± 138.4	
Walnut	7 ± 0.1	7.3 ± 0.1	*	421.7 ± 56.5	237.1 ± 22.9	*

SOIL PHYSICAL AND CHEMICAL PROPERTIES

Soil conditions across sites were similar, even across various soil types, and tree types (Table 2-2). The pH of the soils across our sites were neutral, with an average high of 7.2 ± 0.1 and low of 6.8 ± 0.1 in the 0-10 cm depth. Across sites, the pH of alley and the berm were not significantly different, 7.1 ± 0.07 , and 7.0 ± 0.07 , respectively. There was not a significant difference between sample locations ($p = 0.17$). There was a significant difference ($p < 0.05$) between sites of Almond 4 & 5 compared to Almond 1, which had the highest pH of all sites 7.22 ± 0.1 . The average soil EC for the sites ranged from a low of 90.9 ± 5.6 ($\mu\text{S}\cdot\text{cm}^{-1}$) to a high of 501.1 ± 39.6 ($\mu\text{S}\cdot\text{cm}^{-1}$). For EC, there was a significant difference between sites ($p < 0.001$), and an interaction effect between location and site ($p < 0.001$; Table 3). The conventionally managed almond sites were significantly different than that of the organically managed site ($p < 0.05$), with organically managed sites having a lower EC than conventional. There was a significant difference between Almond 1's alley and berm EC ($p=0.03$) and Walnut ($p<0.5$).

Soil bulk density across sites ranged from the lowest at the surface soils (0-10) with a mean of 1.14 ± 0.02 $\text{g}\cdot\text{cm}^{-3}$, with the highest bulk density being in the 50 cm depth with a mean of 1.85 ± 0.02 $\text{g}\cdot\text{cm}^{-3}$, thereafter it decreased to 1.59 ± 0.03 $\text{g}\cdot\text{cm}^{-3}$ at the max depth of 1 m. Across sites, there was a significant effect on bulk density of sampling location of alley and berm ($p < 0.001$), and depth ($p < 0.001$) and there was a significant interaction effect of the two factors ($p < 0.001$). A post hoc test revealed that the 10-30 cm depth had a significant difference between alley and berm bulk density ($p < 0.001$), with the alley having an 18% higher bulk density than the berm. Soil texture differed across the sites in our study (Table 2-2). The average clay content in the whole soil profile (0-100cm) across sites was 6 ± 0.2 %, with a low of 0.7 ± 0.2 % at the Walnut site, to a high of 20.7 ± 0.2 % at the Almond 4 site.

Table 2-3. Soil properties include soil type information, and the total soil C percentage (C%), total soil nitrogen percentage (%N), bulk density (BD), and soil texture proportions of clay, sand and silt.

Site	Soil Information	Depth cm	Carbon (C)	Nitrogen (N)	C:N	BD	Clay	Sand	Silt
			%	%		g/cm ³	%		
Almond 1	Soil Order	0-10	1.09 ± 0.05	0.14 ± 0.003	8:1	1.16 ± 0.05	14.7 ± 0.25	49.5 ± 0.1	36.4 ± 0.4
	Inceptisol	10-30	0.89 ± 0.04	0.11 ± 0.003	8:1	1.32 ± 0.06	17.9 ± 0.15	50.2 ± 0.1	33.1 ± 0.1
	Soil Series	30-50	0.65 ± 0.03	0.10 ± 0.003	7:1	1.88 ± 0.07	18.5 ± 0.28	53.4 ± 0.5	27.1 ± 0.2
	<i>Zacharias</i>	50-75	0.45 ± 0.04	0.06 ± 0.005	7:1	1.62 ± 0.11	17.3 ± 0.23	57.3 ± 0.3	25.4 ± 0
	Fine-loam	75-100	0.42 ± 0.04	0.05 ± 0.004	8:1	1.49 ± 0.11	17.5 ± 0.3	54 ± 0.9	28.5 ± 0.6
Almond 2	Soil Order	0-10	1.09 ± 0.03	0.14 ± 0.004	8:1	1.23 ± 0.05	14.3 ± 0.29	48.9 ± 0.1	36.3 ± 0.4
	Inceptisol	10-30	0.96 ± 0.04	0.11 ± 0.004	9:1	1.51 ± 0.07	16.9 ± 0.15	50.2 ± 0.1	33.6 ± 0.1
	Soil Series	30-50	0.74 ± 0.04	0.10 ± 0.003	7:1	1.75 ± 0.07	19.5 ± 0.28	53.4 ± 0.5	27.1 ± 0.2
	<i>Zacharias</i>	50-75	0.46 ± 0.06	0.06 ± 0.003	8:1	1.62 ± 0.14	17.3 ± 0.23	57.3 ± 0.3	25.4 ± 0
	Fine-loam	75-100	0.52 ± 0.14	0.06 ± 0.004	9:1	1.47 ± 0.12	17.4 ± 0.3	54 ± 0.9	28.5 ± 0.6
Almond 3	Soil Order	0-10	0.75 ± 0.07	0.08 ± 0.010	9:1	1.06 ± 0.03	6.4 ± 0.3	66.1 ± 0	27.5 ± 0.3
	Mollisols	10-30	0.71 ± 0.06	0.07 ± 0.010	10:1	1.30 ± 0.03	5.2 ± 0.53	66.5 ± 0.3	28.3 ± 0.3
	Soil Series	30-50	0.70 ± 0.08	0.08 ± 0.010	9:1	1.85 ± 0.04	4.5 ± 0.06	64.4 ± 0.1	31.1 ± 0
	<i>Visalia</i>	50-75	0.91 ± 0.11	0.11 ± 0.020	8:1	1.66 ± 0.09	4.2 ± 0.73	75.6 ± 1.2	20.2 ± 0.5
	Fine Sandy Loam	75-100	0.56 ± 0.14	0.05 ± 0.010	11:1	1.74 ± 0.08	3.5 ± 0.65	72.1 ± 0.2	24.4 ± 0.8
Almond 4	Soil Order	0-10	0.83 ± 0.05	0.10 ± 0.005	8:1	1.17 ± 0.05	19 ± 0.52	40.6 ± 0.1	40.4 ± 0.4
	Entisol	10-30	0.49 ± 0.02	0.06 ± 0.002	8:1	1.68 ± 0.07	22 ± 0.07	46.3 ± 0.6	31.7 ± 0.6
	Soil Series	30-50	0.34 ± 0.05	0.04 ± 0.003	8:1	1.99 ± 0.06	21.1 ± 0.14	46.8 ± 0.7	32.1 ± 0.5
	<i>Hanford</i>	50-75	0.33 ± 0.07	0.03 ± 0.004	11:1	2.01 ± 0.34	21.5 ± 0.12	44.3 ± 0	34.2 ± 0
	Coarse-loam	75-100	0.20 ± 0.03	0.02 ± 0.002	10:1	1.57 ± 0.30	20.2 ± 0.14	46.2 ± 0	33.6 ± 0
Almond 5	Soil Order	0-10	1.49 ± 0.13	0.15 ± 0.010	10:1	1.20 ± 0.04	1.3 ± 0.06	86.2 ± 0.7	12.5 ± 0.7
	Alfisol	10-30	0.56 ± 0.04	0.06 ± 0.004	9:1	1.54 ± 0.04	0.7 ± 0	83.4 ± 0.3	15.9 ± 0.3
	Soil Series	30-50	0.46 ± 0.04	0.05 ± 0.003	9:1	1.85 ± 0.08	1.7 ± 0.14	87 ± 0.4	11.3 ± 0.2
	<i>Montpellier</i>	50-75	0.28 ± 0.03	0.04 ± 0.002	7:1	1.65 ± 0.11	1.7 ± 0	82.6 ± 0.3	17.4 ± 0.3
	Fine-loam	75-100	0.24 ± 0.02	0.03 ± 0.002	8:1	1.51 ± 0.16	9.4 ± 2.15	64.4 ± 3.3	26.2 ± 1.1
Peach	Soil Order	0-10	1.27 ± 0.04	0.18 ± 0.010	7:1	1.06 ± 0.04	1.4 ± 0.01	74.8 ± 0.4	23.8 ± 0.4
	Mollisols	10-30	0.85 ± 0.06	0.10 ± 0.010	9:1	1.33 ± 0.05	1.4 ± 0.09	74.2 ± 0.5	24.4 ± 0.4
	Soil Series	30-50	0.50 ± 0.03	0.10 ± 0.003	5:1	1.82 ± 0.06	0.7 ± 0	76.2 ± 0.1	23.2 ± 0.1
	<i>Grangeville</i>	50-75	0.30 ± 0.03	0.05 ± 0.002	6:1	1.76 ± 0.10	0.3 ± 0.08	78.8 ± 0.7	20.9 ± 0.6
	Coarse-loam	75-100	0.23 ± 0.03	0.03 ± 0.002	8:1	1.51 ± 0.09	0.4 ± 0.09	82.5 ± 0.2	17.1 ± 0.1
Pistachio	Soil Order	0-10	1.37 ± 0.13	0.14 ± 0.010	10:1	1.15 ± 0.04	3.5 ± 0	75.5 ± 0.1	20.9 ± 0.1
	Mollisols	10-30	0.77 ± 0.05	0.08 ± 0.004	10:1	1.35 ± 0.06	3.5 ± 0.01	81.2 ± 0.3	15.2 ± 0.3
	Soil Series	30-50	0.61 ± 0.06	0.06 ± 0.003	10:1	1.72 ± 0.07	0.7 ± 0	83.8 ± 1.3	15.5 ± 1.3
	<i>Grangeville</i>	50-75	0.36 ± 0.04	0.04 ± 0.003	9:1	1.45 ± 0.08	0.3 ± 0.08	87.9 ± 1.9	11.8 ± 1.8
	Coarse-loam	75-100	0.21 ± 0.03	0.02 ± 0.003	11:1	1.62 ± 0.12	0.3 ± 0.07	92.2 ± 0.1	7.8 ± 0.1
Walnut	Soil Order	0-10	0.43 ± 0.03	0.06 ± 0.004	7:1	1.12 ± 0.04	0.7 ± 0	76.4 ± 0.3	22.9 ± 0.3
	Entisol	10-30	0.65 ± 0.13	0.07 ± 0.010	9:1	1.37 ± 0.04	0.7 ± 0	75.1 ± 0.1	24.2 ± 0.1
	Soil Series	30-50	0.36 ± 0.11	0.04 ± 0.005	9:1	2.04 ± 0.07	0.5 ± 0.06	78.7 ± 0	20.8 ± 0
	<i>Hanford</i>	50-75	0.17 ± 0.02	0.02 ± 0.001	9:1	1.71 ± 0.15	0.7 ± 0	81.4 ± 0.8	17.9 ± 0.8
	Coarse-loam	75-100	0.13 ± 0.01	0.01 ± 0.001	13:1	1.66 ± 0.06	0.7 ± 0.01	78.8 ± 1.4	20.5 ± 1.4

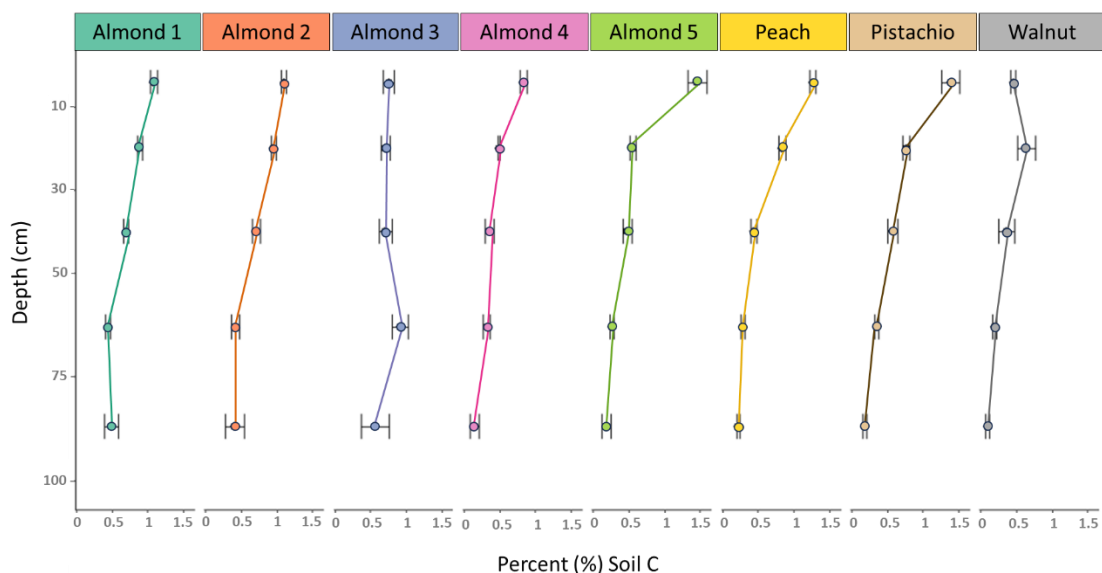


Figure 2-2. Soil C percent (%) changes along the depth (cm) gradient from surface to 1 m across the 8 sites.

SOIL C & N

Across sites the average SOC percent was highest in the surface soils (0-10 cm) with a mean of $1.1 \pm 0.03\%$, and gradually decreased to $0.34 \pm 0.03\%$ at 1 m (Figure 2-2). The average soil total N concentration across sites was also highest in the surface soils (0-10) with a mean of $0.13 \pm 0.00\%$ and decreased to $0.04 \pm 0.002\%$ at 1 m depth (Table 2-3. Soil properties include soil type information, and the total soil C percentage (C%), total soil nitrogen percentage (%N), bulk density (BD), and soil texture proportions of clay, sand and silt.). There was no significant difference between sampling location of alley and berm ($p = 0.83$), but there was a significant interaction effect by depth and location ($p < 0.001$). To test the impact of soil properties, such as soil texture, we conducted a regression analysis. The coefficient estimates revealed significant associations between each soil texture factor and SOC content ($p < 0.001$). Overall, the regression model was statistically significant (F-statistic = 22.13, $p < 0.001$), indicating that the combined effects of soil particle size collectively explain variability in SOC content. However, the model's explanatory power was limited ($R^2 = 0.04$), suggesting that other factors not included in the analysis may contribute to SOC variability, likely the site effects of management or soil mineralogy.

Table 2-4. Method comparison between fixed depth (FD) and equivalent soil mass (ESM) calculations for soil C stocks are reported. Cumulative soil mass is also report (Cum. Soil).

Site	Depth (cm)	Cum. Soil (Mg/ha)	FD	ESM	Site	Depth (cm)	Cum. Soil (Mg/ha)	FD	ESM
			Soil C Storage (Mg/ha)					Soil C Storage (Mg/ha)	
Almond 1	0-10	1187	13.1 ± 0.6	16.3 ± 0.8	Almond 5	0-10	1234	19.2 ± 1.8	21.2 ± 1.9
	10-30	3872	24.7 ± 1	25.1 ± 1.1		10-30	4347	19.1 ± 1.4	17.9 ± 1.3
	30-50	7676	26 ± 1.1	22 ± 1		30-50	8083	19.3 ± 1.9	13.1 ± 1
	50-75	11761	18.2 ± 1.5	17.7 ± 1.6		50-75	12197	9.8 ± 1.1	10.2 ± 0.9
	75-100	15527	17.4 ± 1.7	16.2 ± 1.9		75-100	15984	9.5 ± 1.4	9.7 ± 0.9
Almond 2	0-10	1252	13.7 ± 0.4	16.7 ± 0.6	Peach	0-10	1089	14.1 ± 0.5	16.8 ± 0.6
	10-30	4321	30 ± 1.2	29.4 ± 1.3		10-30	3795	25.1 ± 1.5	27.5 ± 1.8
	30-50	7870	26.6 ± 1.3	27.6 ± 1.8		30-50	7466	19.7 ± 1.2	16 ± 0.9
	50-75	11948	18.8 ± 2.6	19 ± 2.9		50-75	11895	13.5 ± 1.4	11.9 ± 1.2
	75-100	15647	17.7 ± 4	22.9 ± 6.4		75-100	15681	8.8 ± 1	9 ± 0.9
Almond 3	0-10	1073	8.2 ± 0.8	10.4 ± 1	Pistachio	0-10	1184	16.5 ± 1.6	14.7 ± 1.3
	10-30	3719	19.2 ± 1.6	20.7 ± 1.9		10-30	3916	20.7 ± 1.3	22.1 ± 1.4
	30-50	7464	27 ± 3.1	20 ± 2.4		30-50	7398	21.7 ± 2.5	20.9 ± 2.2
	50-75	11681	38.8 ± 4.8	34.1 ± 4.3		50-75	11056	13.4 ± 1.4	13.9 ± 1.5
	75-100	16071	19.9 ± 8.4	21.3 ± 5.1		75-100	15099	8.4 ± 1.3	8.4 ± 1.4
Almond 4	0-10	1186	10.6 ± 0.5	12.6 ± 0.6	Walnut	0-10	1135	5.1 ± 0.5	6.8 ± 0.6
	10-30	4585	19.2 ± 1.4	16.9 ± 1.3		10-30	3902	19 ± 4.1	21.6 ± 4.2
	30-50	8599	15.5 ± 1.4	12.4 ± 1.2		30-50	8014	16.1 ± 3.9	12.8 ± 4
	50-75	13660	14.2 ± 1.8	12.1 ± 1.5		50-75	12296	9.5 ± 1	7.2 ± 0.7
	75-100	17606	10.1 ± 0.9	9.6 ± 1.1		75-100	16464	4.3 ± 0.6	5.3 ± 0.5

SOIL C STOCKS

The distribution of soil C along the soil profile across the sites were similar across sites (Figure 2-3). Generally, the surface soils (0-10 cm) and deep soils (75-100) exhibited the lowest soil C stocks levels within the soil profile, with averages of $12.6 \pm 0.5 \text{ Mg C} \cdot \text{ha}^{-1}$ and $13.6 \pm 1.4 \text{ Mg C} \cdot \text{ha}^{-1}$, respectively. The 10-30 and the 30-50 cm had significantly ($p < 0.05$) higher soil C stocks compared surface and deep soils, averaging $21 \pm 0.7 \text{ Mg} \cdot \text{ha}^{-1}$ and $21 \pm 0.84 \text{ Mg C} \cdot \text{ha}^{-1}$, respectively. The subsequent depth increment of 50-75 cm experienced a decline to an average of $18 \pm 1.5 \text{ Mg C} \cdot \text{ha}^{-1}$. There was little variability in soil C stocks along the soil profile, but there was higher variation in the deeper depths. Cumulative soil C to 1m across sites ranged from $55 \pm 5.2 \text{ Mg C} \cdot \text{ha}^{-1}$ to $109 \pm 11.6 \text{ Mg C} \cdot \text{ha}^{-1}$. There was a significant effect of site on cumulative soil C stocks ($p < 0.001$). There was a grouping of higher cumulative soil C stocks in Almonds 1-3, however there was generally not much statistically significant variation between all orchard sites (Figure 3). ESM and FD were not significantly different in their estimation of cumulative soil C stocks ($p = 0.75$), but there was a significant difference of their estimation of soil C stocks within sites and depth ($p < 0.0001$). With a synergistic effect of site*depth*type ($p < 0.0001$), but that was primarily driven by the comparison between depths, like 10 to 100 cm. Even though there was not a significant difference, there was a relatively large percent difference between the methods for the 0-10 soils ($15 \pm 12\%$. and the 50-75 cm ($18.8 \pm 13.5\%$). Which indicates the various bulk densities and soil mass can have a large influence on the estimation of soil C stocks.

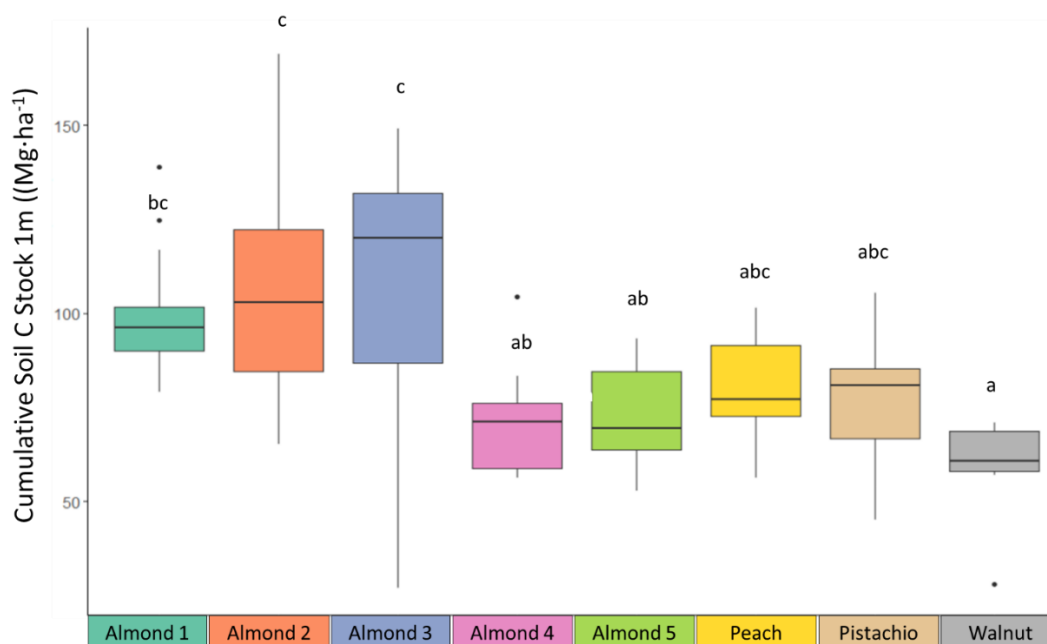


Figure 2-3. Cumulative soil C stocks (Mg/ha) for the 8 sites in the study. The post hoc letters above each bar indicate the results of a post hoc test performed after a linear mixed model to determine significant differences between the groups. Groups that share a letter are not significantly different from one another.

INFLUENCE OF MANAGEMENT ON SOIL C

Irrigation type had a significant effect on cumulative soil C stocks ($p < 0.001$). In post hoc analysis cumulative soil C stocks in flood irrigation management (walnut) were significantly lower than drip sites ($p < 0.01$). As soil and water dynamics are intricately linked, we also found that soil type exerted a significant influence on cumulative soil C stocks ($p < 0.05$). There was a significant difference between Inceptisols (Almonds 1 & 2) and Entisols (Walnuts) ($p = 0.01$ for both soil types), with Inceptisols having higher soil C stocks than Entisols. Soil's clay content had a significant effect on soil C content ($p < 0.05$) and had a significant interaction effect with clay content and depth ($p < 0.05$). The age of orchards ranged from 4 to 30 years and had a significant effect on cumulative soil C ($p = 0.002$). To test the correlation coefficient between age and cumulative soil C, a Kendall rank correlation test was conducted, and found that they were weakly negatively correlated ($T = -0.224$). Tree type had a significant effect on cumulative soil C stocks ($p = 0.007$), where Almonds had a significantly higher stocks than walnuts ($p = 0.02$). Alley and berm locations followed similar trends with depth, and with cumulative soil C stocks. Berms tended to have a slightly higher cumulative soil C stocks content, but it was not a significant difference across most sites, only within the organic sites or Almond 4 and Peach ($p < 0.05$). The effect of organic management did not have a significant effect on soil C stocks compared to conventionally managed sites.

DISCUSSION

CUMULATIVE SOIL C VARIATION ACROSS SITES

The study measured a variable range in cumulative soil C stocks across the orchard sites, spanning from a minimum of $55 \pm 5.2 \text{ Mg C} \cdot \text{ha}^{-1}$ to a maximum of $109 \pm 11.6 \text{ Mg C} \cdot \text{ha}^{-1}$ (Figure 3). Our findings are within the range of other studies reports of orchard soil C stocks (Fenster et al., 2021; Gómez et al., 2022; Sharma et al., 2021). The observed range in cumulative soil C stocks reflects the complexity of orchard systems, where factors such as soil properties, management practices, and regional variations can exert pronounced effects on soil C stocks. While some sites were significantly different, these differences in soil C stocks across sites were not extreme, suggesting some consistency, possibly due to regional or climatic factors mitigating local variability.

DEEP SOIL C

Our study reveals a pertinent finding regarding deep soil C contributions to understanding whole orchard soil C stocks. We found that the average cumulative soil C stocks to a depth of 1 m in Central Valley orchards was $85 \pm 5 \text{ Mg C} \cdot \text{ha}^{-1}$. Importantly, had we solely sampled surface soils (0-30 cm), the estimate of cumulative soil C stocks would have been significantly lower, averaging $34 \pm 1 \text{ Mg C} \cdot \text{ha}^{-1}$. This surface-only estimation would have underestimated the soil C stocks in these orchards by a substantial 60%, equivalent to a difference of $51 \pm 2 \text{ Mg C} \cdot \text{ha}^{-1}$. Although the deeper soil layers contain relatively less soil C compared to the surface, their cumulative contribution to 1 m is crucial for assessing the potential for enhancing soil C sequestration and long-term stocks in perennial orchards. Soil C stocks at deeper depths exhibit greater protection from microbial mineralization due to the complex interactions between physical, chemical, biological, and environmental factors within the soil matrix (Button et al., 2022). Key parts of protection include increased soil aggregation (Blanco-Canqui & Lal, 2004; Ozlu & Arriaga, 2021), soil organo-mineral associations (Vormstein et al., 2020), and the decreased oxygen levels slow down microbial activity and decomposition rates. Consequently, soil C in deeper layers persists longer due to increased stability which relates to reduced turnover rates, significantly impacting long-term soil carbon stocks dynamics (Fontaine et al., 2007; Hicks Pries et al., 2023).

In perennial systems like orchards, there is a significant opportunity to increase deep soil C through various management practices. One such method is the burial of biochar or woody residues during the establishment of the orchard. Biochar, a stable form of carbon produced from the pyrolysis of organic materials, can enhance soil structure, water retention, and nutrient availability while providing a long-term carbon sink (Singh Yadav et al., 2023; Yin et al., 2022). Incorporating woody residues into deeper soil layers can also add to the carbon pool, as these materials decompose more slowly compared to finer organic matter (Gooding, 2023; Zeng & Hausmann, 2022). Additionally, practices such as whole orchard recycling, where pruned branches and other biomass are returned to the soil, can further enhance carbon stocks and nutrient cycling within orchard systems (Jahanzad et al., 2022). These practices not only enhance the C content of deeper soil layers but also improve overall soil health and resilience, providing additional benefits such as increased water infiltration and reduced erosion (Kumi et al., 2021; Singh Yadav

et al., 2023). Despite the potential benefits, there are still many unknowns about the most effective management tools for increasing deep soil C. Research is needed to better understand the interactions between different soil types, climate conditions, and management practices to optimize deep soil carbon sequestration. This area of study holds promise for developing sustainable agricultural practices that contribute to climate mitigation by leveraging the protected nature of deep soil carbon.

INFLUENCES ON SOIL C STOCKS

We examined several management factors, including tree type, orchard age, soil type, and irrigation method, and found most had a significant effect on cumulative soil C stocks. We also tested the difference of alley and berm location on cumulative soil C and soil C stocks by depth. We found that the surface soils of the two locations within an orchard were not significantly different for either soil C concentration or content across sites, but there was a significant difference at deeper depths. The berms had a larger, but not significant, soil C stocks than alleys in the deeper depths across sites, with an average cumulative stock of $23 \pm 5 \text{ Mg C} \cdot \text{ha}^{-1}$ and compared to the alleys $19 \pm 2 \text{ Mg C} \cdot \text{ha}^{-1}$. This increase in soil C stocks in the berms compared to the alleys is likely attributed to root exudation. Research on rates of C allocation from root exudation might explain why we do not observe a large shift of soil C stocks between alley and berm. Trees in unmanaged systems often respond to reduced water supply by modifying their belowground C allocation, and potentially increase root exudation rates (Brunn et al., 2022). If orchards are carefully managed to reduce tree stress from drought through precision irrigation, it is unlikely that they would exhibit the same trends as natural systems. When orchard trees exude carbohydrates from their roots at a rate unaffected by drought effects it could be substantially less than natural systems. Having roots in the berms, compared to the alleys where there are little to no living plants, provides inputs from exudation along with root turnover.

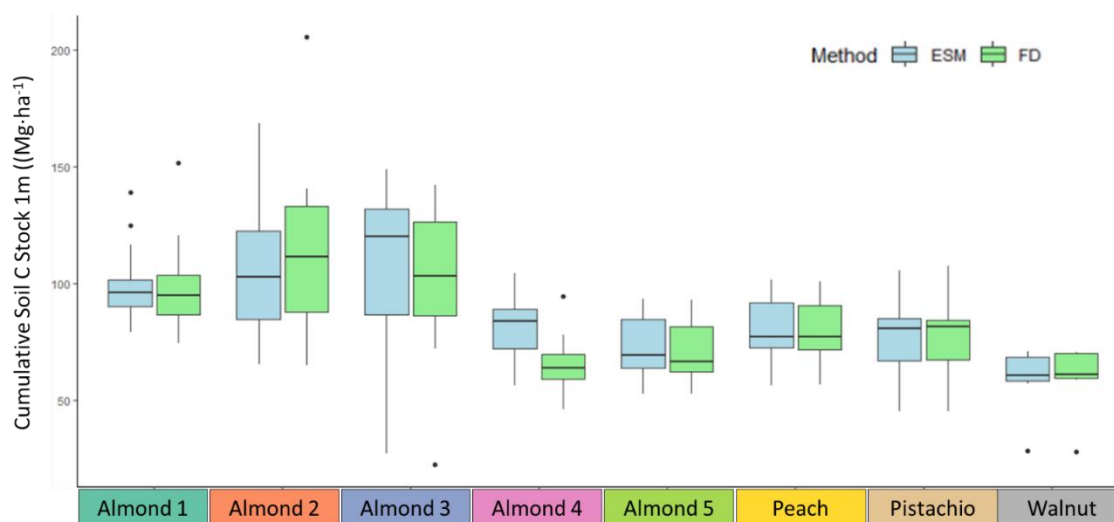


Figure 2-4. Cumulative soil C stocks method comparison between equivalent soil mass and fixed depth methods for the 8 sites in the survey.

METHOD COMPARISON

The ESM and the FD method did not significantly differ in their estimation of cumulative soil C stocks and soil C stocks along the depth profile (Figure 2-4). The FD method can be more labor intensive in the field due to the use of quantitative corers or bulk density pits, and have error associated with the use of bulk density that could over- or underestimate soil C stocks. The ESM method might be prone to errors if the sampler is not careful to exclude soil additions that from the surface into the auger while sampling. Despite these different challenges, both methods estimate similar values soil C stocks. The ESM method would be the preferred choice especially with studies investigating the effect of management changes that could affect the bulk density such as decreased tillage, cover crops, or the addition of exogenous organic material such as compost (von Haden et al., 2020). ESM corrects for extremes in bulk density and uses mineral soil mass to calculate soil C stocks, and as such it would more likely capture changes to soil C that are related to soil C processes (e.g., increased C inputs or mean residence time of C) rather than changes to soil compaction which decreases bulk density. Other studies have found that there are significant differences in the way in which these two methods estimate soil C stocks (Raffeld et al., 2024). These studies found significant differences between the two methods due to temporal effect in the analysis where changes in bulk density due to management have a larger influence (Rovira et al., 2022; Mikha et al., 2013).

COMPARISONS OF ORCHARD SOIL C STOCKS TO OTHER CROPLANDS

In general, systems that have been converted to croplands experience substantial loss of soil C. In a long-term study in Germany, Wiesmeier et al. (2013) found that grasslands stored the most (118 Mg C · ha⁻¹ in the top 45 cm), followed by forest soils (98 Mg C · ha⁻¹ in the top 45 cm), and croplands stored the least (90 Mg C · ha⁻¹ in the top 45 cm). To compare within the state of California, orchard systems store less soil C than rangelands. In California, rangelands are a dominant land use type covering half of available land (Carey et al., 2020). These rangelands are comprised of annual grasses and forbs and are used as forage for cattle grazing. Within these annual systems, soil C stocks averaged 90 Mg C · ha⁻¹ in the top 50 cm (Silver et al., 2010). Compared to our orchard sites for the same depth, perennial orchards store markedly less at 56 ± 1 Mg C · ha⁻¹. The sites with the most recent conversion to a perennial cropping system have the highest soil C stocks, like our Almond 1 and 2 sites (4 years old), which were previously managed as rangelands before almond production, a finding supported by additional research on grassland conversion (Tang et al., 2019). There seems to be a trend that sites with a longer history of orchard production, like the Walnut site (20+ years old), are the most depleted in soil C stocks. Current conventional management practices within orchards do not support soil C sequestration. The depleted status of orchard soils should make them a key target for implementing management practices aimed at enhancing soil C stocks sequestration (Yin et al., 2022).

CARBON DEBT OF ORCHARDS

Agricultural practices have historically incurred a significant global soil C debt, with estimations indicating a loss of 31.2 petagrams of carbon (Pg C) from the upper 30

centimeters of soil over the past 12,000 years (Sanderman et al., 2017). Recent research indicates that this estimation might be conservative, projecting croplands to have a soil C debt ranging between 21 and 186 Pg C (Padarian et al., 2022). Preceding agricultural conversion, California's Central Valley was characterized by extensive biodiverse grasslands interspersed with riparian wetland zones (Garone, 2020). However, the introduction of conventional tillage in the 1930s swiftly transformed these grasslands into croplands, leading to massive losses of soil C due to wind and water erosion and microbial decomposition (Mitchell et al., 2016). On average agricultural soils now contain 25% to 75% less soil C than their pre-agricultural states (Don et al., 2011). Even with a conservative estimate of a 25% loss, the soils in our study might have historically stored 47.47 Mg C·ha⁻¹, potentially losing 11.9 Mg C·ha⁻¹ through agricultural conversion, tillage, and conventional practices. The amount of soil C lost can give a conservative goal for soil C sequestration efforts.

POTENTIAL SOIL C SEQUESTRATION

Understanding this soil C debt is essential for evaluating the potential for soil C stock enhancement through sustainable soil management techniques. Orchards are natural “sponges” for absorbing CO₂, but only under management that promotes the soil environment that fosters increased soil C stocks (Gelaye & Getahun, 2024; Patil & Kumar, 2017; Sharma et al., 2021). If we were to sequester the amount of soil C lost during agricultural conversion, an estimated 56.96 MMT could be stored within the orchard systems in California. However, the rate at which this goal could be achieved remains debatable and contingent upon various factors, including soil texture, management strategies, climate, and initial soil C content. Soils with low saturation exhibit a greater potential for enhancing soil C stocks compared to soils at or near their saturation point (Georgiou et al., 2022). California has set a target of removing 7 MMT CO₂ per year by 2030 (CARB, 2022). California's orchards hold significant potential to contribute to these goals through soil carbon sequestration. A 0.5% increase in soil carbon concentration to the 30 cm depth across 1.3 million hectares of nut orchards (assuming average BD within our study of 1.4 g/cm³) could sequester approximately 27 MMT of carbon, equivalent to 100.19 MMT of CO₂. This represents nearly 26% of California's annual CO₂ emissions of 384 MMT (CARB,2022), surpassing the annual sequestration target.

MANAGEMENT RECOMMENDATIONS FOR INCREASING SOIL C SEQUESTRATION

To enhance soil C sequestration and storage in orchards—and capitalize on their potential to offset a significant portion of California’s CO₂ emissions—it is essential to implement management practices that not only improve soil health but also promote carbon capture through plant photosynthesis. By focusing on the biological, physical, and chemical components of soil health, such practices can facilitate a functioning soil ecosystem while drawing atmospheric CO₂ into the soil via plant growth and organic matter deposition. This dual approach supports long-term carbon storage, providing both climate mitigation benefits and improved agricultural productivity (Lal, 2016). Practices such as compost application, cover cropping, and whole orchard recycling not only promote organic matter decomposition and nutrient cycling but also support long-term

carbon storage and soil fertility. The planting of cover crops in the rainy winter season not only aids in increasing soil C stocks (Peng et al., 2010; Steenwerth & Belina, 2008; Tautges et al., 2019), but also supports native pollinators (Sardiñas et al., 2022). Exogenous inputs of C, like compost, woody biomass, or biochar, can also increase soil C stocks in orchards (**Error! Reference source not found.**). Compost, often surface-applied, acts as a slow-release fertilizer that encourages microbial activity and promotes early soil C stabilization and stocks (Ryals & Silver, 2013; Wong et al., 2023).

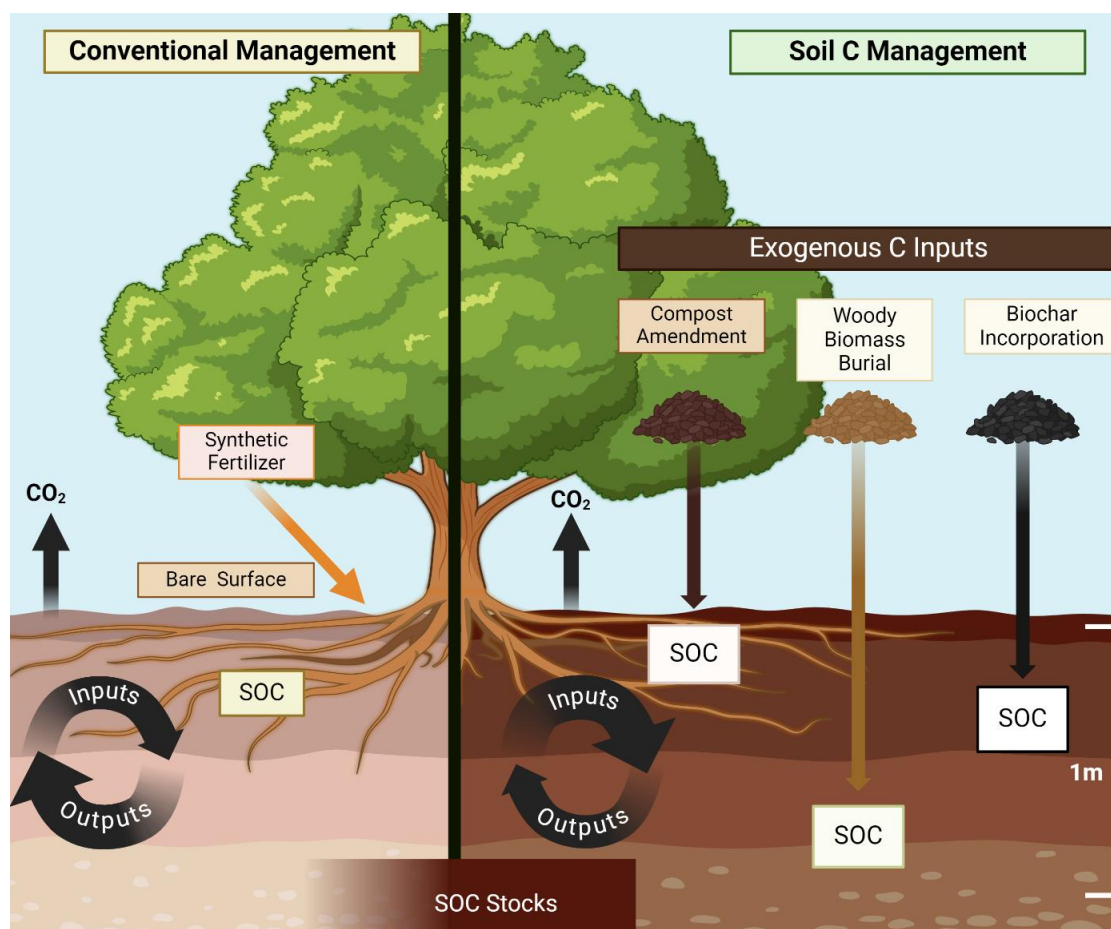


Illustration: 2-2 Conceptual figure of the shift from conventional management to that of soil C management in orchards systems. In both management systems, there is a cycle of inputs and outputs, if outputs (i.e. CO₂ emissions via SOC mineralization) are greater than inputs, SOC storage steadily decreases through time. In the soil C management side of the figure, the three exogenous inputs are demonstrated: compost amendments, woody biomass burial, and biochar incorporation – with arrows indicating where in the soil profile they are often added to (surface, root zone soil, or deep soil). Note: not all are implemented at the same time, but rather often chosen based on soil health or C management goals.

Woody residues, such as those from whole orchard recycling, can be added to deeper soil layers, where they decompose more slowly than finer organic matter, contributing to the C pool (Gooding, 2023; Jahanzad et al., 2022; Zeng & Hausmann,

2022). Biochar, a stable form of C produced from the pyrolysis of organic materials, enhances soil structure, water retention, and nutrient availability while providing a long-term C sink (Singh Yadav et al., 2023; Yin et al., 2022). These practices not only enhance the C content of deeper soil layers but also improve overall soil health and resilience, offering benefits like increased water infiltration and reduced erosion (Kumi et al., 2021). However, research has suggested that the greatest opportunity to increase deep soil C every 20 years, is during orchard replanting (Khalsa & Brown, 2017). This highlights the critical importance of timing interventions to maximize carbon sequestration potential. It also suggests that achieving significant increases in soil carbon stocks may require long-term planning, potentially prolonging the time until measurable benefits are observed. Strategic management during these key periods of orchard lifecycle transition is essential to optimizing soil carbon storage.

Despite these potential benefits, more research is needed to understand the most effective management tools for increasing deep soil C. Understanding the interactions between different soil types, climate conditions, and management practices is crucial to optimizing deep soil C sequestration. This research holds promise for developing sustainable agricultural practices that contribute to climate mitigation by leveraging the protected nature of deep soil C. Careful monitoring of soil C stocks, both spatially and temporally, is essential to accurately assess the impact of these practices. Programs like California's Soil Health Program provide funding support to orchardists for these often-front-loaded expenses (Bradford et al., 2019). These combined efforts can contribute to increased soil C stocks, enhancing orchard productivity and aiding in climate change mitigation through C sequestration.

CONCLUSION

The soil C stocks of orchards across California's Central Valley are relatively low, highlighting the need for management strategies that increase soil C sequestration and build long-term C storage. Our observations of cumulative soil C levels across orchard sites reveal the multifaceted influence of factors such as soil properties, management practices, and regional variations. Comparing methods for assessing soil C stocks, we found both the ESM and FD approaches to be reliable, with the ESM method proving more robust in scenarios where management practices affect soil bulk density. While our study sheds light on the roles of tree type, orchard age, soil type, and irrigation TYPE in shaping cumulative soil C stocks, it is essential to account for additional factors, including historical soil health practices, prior land use, and management actions such as deep tillage for planting. These historical and operational variables likely exert a significant influence on current soil C stocks. Orchards have been historically overlooked as a critical land type for increasing soil C stocks, yet their potential to contribute meaningfully to California's climate goals is undeniable. While achieving the calculated offset of 27% of California's annual CO₂ emissions may pose challenges, it represents a compelling and impactful target. Incremental increases in soil C, achieved through sustained, science-based management practices, offer a tangible pathway to climate mitigation, highlighting the critical role of orchards in these efforts.

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DECLARATION OF GENERATIVE AI AND AI-ASSISTED TECHNOLOGIES IN THE WRITING PROCESS

During the preparation of this work the authors used ChatGPT by OpenAI to increase readability via syntax and fix grammatical errors. After using this tool, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

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CHAPTER 3: THE POSITIVE INFLUENCE OF COMPOST AND COVER CROPS ON KEY SOIL HEALTH INDICATORS IN NUT ORCHARDS

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ABSTRACT

Sustainable management practices in nut orchards are crucial for enhancing soil health and minimizing environmental impacts associated with conventional production. Over three years, this study evaluated the effects of stacked soil health practices in five nut orchards (almonds and walnuts) in California. Experimental plots received annual applications of 19 Mg/ha of yard waste and manure compost, along with cover crops at a rate of 13.8 kg/ha, while conventionally managed plots served as controls. Compost and cover crops significantly improved all soil health indicators by year three. Microbial biomass carbon (MBC) increased significantly by $573 \pm 98\%$ in alleys and $307 \pm 100\%$ in berms, while microbial biomass nitrogen (MBN) showed significant increases of $487 \pm 95\%$ in alleys and $55 \pm 14\%$ in berms. Permanganate oxidizable carbon (POXC) levels rose significantly by $133 \pm 23\%$. Short-term carbon mineralization rates were $70 \pm 5\%$ higher, reflecting enhanced soil biological activity likely from increased POXC. Cation Exchange Capacity (CEC) significantly increased by $25.7 \pm 14.5\%$ in alleys and $48 \pm 16.9\%$ in berms. Soil extractable ammonium (NH_4^+) levels increased rapidly following compost application, remaining significantly elevated for five months before aligning with control plots, whereas soil extractable nitrate (NO_3^-) levels did not significantly change. By the end of the study, the average percent increase in soil organic carbon (SOC) content with the stacked practices was $102 \pm 21\%$, resulting in an average SOC content of $60.7 \text{ Mg C ha}^{-1}$ across sites, with a cumulative soil C storage increase (0-30cm) of . The pH in compost + cover crops plots decreased slightly from 7.1 in 2021 to 6.8 in 2024, with an average change of -0.3 ± 0.3 . Electrical conductivity (EC) increased significantly with an average rise of $445.3 \pm 311.2 \text{ } \mu\text{S/cm}$. Improvements in wet aggregate stability were also significant, with large macroaggregates increasing by $2.1 \pm 0.5\%$, medium macroaggregates by $1.3 \pm 0.2\%$, and small macroaggregates by $8.4 \pm 2\%$. These findings underscore the critical role of compost and cover crops in enhancing soil health within conventional nut orchard management, independently from site factors such as soil type and management. The increase in SOC storage highlights the potential of orchard soils to serve as a significant carbon sink while supporting agricultural sustainability and resilience to climate change.

INTRODUCTION

Ecological-based management of nut orchard systems can play a key role in supporting ecosystem services and climate mitigation (Fenster et al., 2021; Timberlake et al., 2022; Weier et al., 2024). Environmental concerns related to conventional nut orchard production include water use, hazardous pesticides and herbicides, soil compaction, and nitrate leaching to groundwater (Baram et al., 2016; Fulton et al., 2019; Zhan & Zhang, 2014). Since soil health is crucial for supporting ecosystem services that benefit both human and ecological systems, there is an increasing need to quantify the effects of integrated soil health practices. Compost application and the planting of cover crops has been identified as practices with individual and synergistic benefits to soil health. The application of compost provides essential nutrients and organic matter, increasing stable aggregates and water-holding capacity, while cover crops reduce soil erosion, decrease compaction, and provide additional nutrients through biological nitrogen fixation (Hodson et al., 2021; Repullo-Ruibérriz de Torres et al., 2021). Both practices independently have been documented to enhance the biological, chemical, and physical components of soil health in orchards (Kutos et al., 2023; Scavo et al., 2022). However, more research is needed to assess the effectiveness of these applications when integrated as "stacked" practices, along with the continued use of chemical fertilizers and irrigation, in nut orchard systems.

Compost, made from organic materials such as food waste, yard waste, and livestock manure, is a nutrient-rich soil amendment. It contains essential macronutrients, micronutrients, and organic matter all of which are crucial for plant growth and the provisioning of soil services (Ho et al., 2022). In many agricultural ecosystems, compost increases water-holding capacity and enhances soil structure, resulting in improved root growth, nutrient uptake, reduced soil erosion, and better soil moisture retention, which is particularly important in California's dry climate (Goldan et al., 2023; Ho et al., 2022). Compost also boosts biological activity, as the organic matter component serves as an energy source for soil microorganisms, fostering a diverse and active microbiome (Lazcano et al., 2022). This microbial life is essential for nutrient cycling, breaking down organic matter into forms that plants can readily absorb, thus enhancing soil fertility (Gougoulas et al., 2014). The increased biological activity also contributes to disease and pest suppression and promotes a healthy soil food web, supporting beneficial organisms that further improve soil structure and plant health (M. W. Brown & Tworkoski, 2004; van Bruggen & Semenov, 2000). Additionally, compost has been documented to increase C sequestration, thereby aiding in climate mitigation (Just et al., 2023; Kutos et al., 2023; Wang et al., 2022). This is especially important in perennial agricultural systems, as orchards are often grown for over 20 years, extending the time between major soil disturbances which release large quantities of CO₂ (Cai & Chang, 2020). Almond and walnut soils in this region exhibit relatively low soil C stocks, averaging $36 \pm 0.9 \text{ Mg C} \cdot \text{ha}^{-1}$ in the 0-30 cm depth (Cooper et al., In review). The observed low soil C stocks in these orchards present an opportunity for enhanced C storage through soil health management practices such as compost and (Demestihis et al., 2017).

Cover crops, grown between tree rows, provide benefits such as soil erosion control, nutrient cycling, aeration, and weed suppression (Bechara et al., 2018; Haring &

Hanson, 2022; Koudahe et al., 2022a). Incorporating ground cover and minimizing soil disturbances in permanent crop systems such as orchards can increase microbial biomass, which is essential for forming stable, chemically diverse soil organic C and improving soil health (Ingels et al., 2005; Steenwerth & Belina, 2008; Vukicevich et al., 2019; Whitelaw-Weckert et al., 2007; Bastida et al., 2021; McClelland et al., 2021). Some cover crop species can also fix atmospheric nitrogen, into plant-available forms and provide inputs of organic matter via root turnover (Ordóñez-Fernández et al., 2018; Repullo-Ruibérriz de Torres et al., 2021). In orchards, cover crops can be used for pollinator habitat while addressing soil issues like compaction and low infiltration (Mallinger et al., 2019).

Almonds (*Prunus dulcis*) and walnuts (*Juglans regia*) are the two most common and lucrative nut orchard systems in California, where approximately 80% of the world's almonds and 75% of the world's walnuts are produced (CDFA, 2022). Almond orchards account for the majority of land use in California nut orchards, covering 555,633 bearing hectares (ha) and producing 1.27 T of almonds, resulting in the highest-grossing crop at \$3.5 billion per year (California Almond Board, 2020; CDFA, 2023). Walnuts cover 155,800 ha of bearing land, producing 635,029 T and grossing about \$473 million per year (CDFA, 2023). While most research is highly focused on almonds, it is crucial to test regenerative practices on other common nut crops in California, as these practices have been found to be beneficial across crop types (Lazcano et al., 2022; Scavo et al., 2022).

Almond production in California has grown rapidly over the past two decades. This growth has been accompanied by a corresponding increase in the use of chemical fertilizers to provide essential nutrients such as nitrogen, phosphorus (P), and potassium (K) (CDFA Ag Stats, 2022). The most critical of these nutrients is nitrogen, as it plays a crucial role in tree health and productivity. Nitrogen deficiencies can lead to inadequate protein synthesis and reduced photosynthetic capacity, resulting in decreased growth and nut yield (Zayed et al., 2023). Conversely, excess nitrogen can cause dense vegetative growth at the expense of reproductive development, making trees more susceptible to pests and diseases (Sperling et al., 2019). Beyond the orchard, excessive application rates of nitrogen can also lead to nitrate (NO_3^-) contamination of groundwater and surface water, leading to water pollution and adversely affecting drinking water quality (Haynes, 2022). Additionally, NO_3^- pollution can lead to environmental concerns such as the eutrophication of lakes and rivers (Bijay-Singh & Craswell, 2021; Smolders et al., 2010), and contributes to nitrous oxide, a potent greenhouse gas (Liu et al., 2023). Tighter nitrogen cycling within the soil-plant system is essential to maintaining a balance that supports tree health and production while minimizing the ecological impacts of excess reactive nitrogen in the environment (Tully & Ryals, 2017).

Under conventional management, biological and physical soil health indicators are typically negatively impacted by the continuous use of chemical fertilizers, as well as the ground floor management that removes or prevents vegetation and compacts soil. A key issue is that when one or more of the soil health conditions are lacking, a "leaky" nitrogen cycle can occur, where excessive reactive nitrogen is often lost in gaseous and aquatic forms (Fowler et al., 2013). To mitigate nitrogen losses in orchard

agroecosystems, various management practices have been suggested, such as the addition of compost and cover crops to the current fertilizer management protocols (Castellano-Hinojosa et al., 2023; Lawrence & Melgar, 2023). Cover cropping can reduce NO_3^- leaching by minimizing soil erosion, nutrient runoff, and enhancing soil organic matter content, thereby improving water-holding capacity and reducing leaching (Ordóñez-Fernández et al., 2018). Slow-release fertilizers like compost can regulate NO_3^- leaching due to their biochemical characteristics (Hepperly et al., 2009; Kramer et al., 2006; Xu et al., 2020). These practices can enhance nitrogen cycling while buffering trees against nitrogen-related stress (Drinkwater & Snapp, 2007).

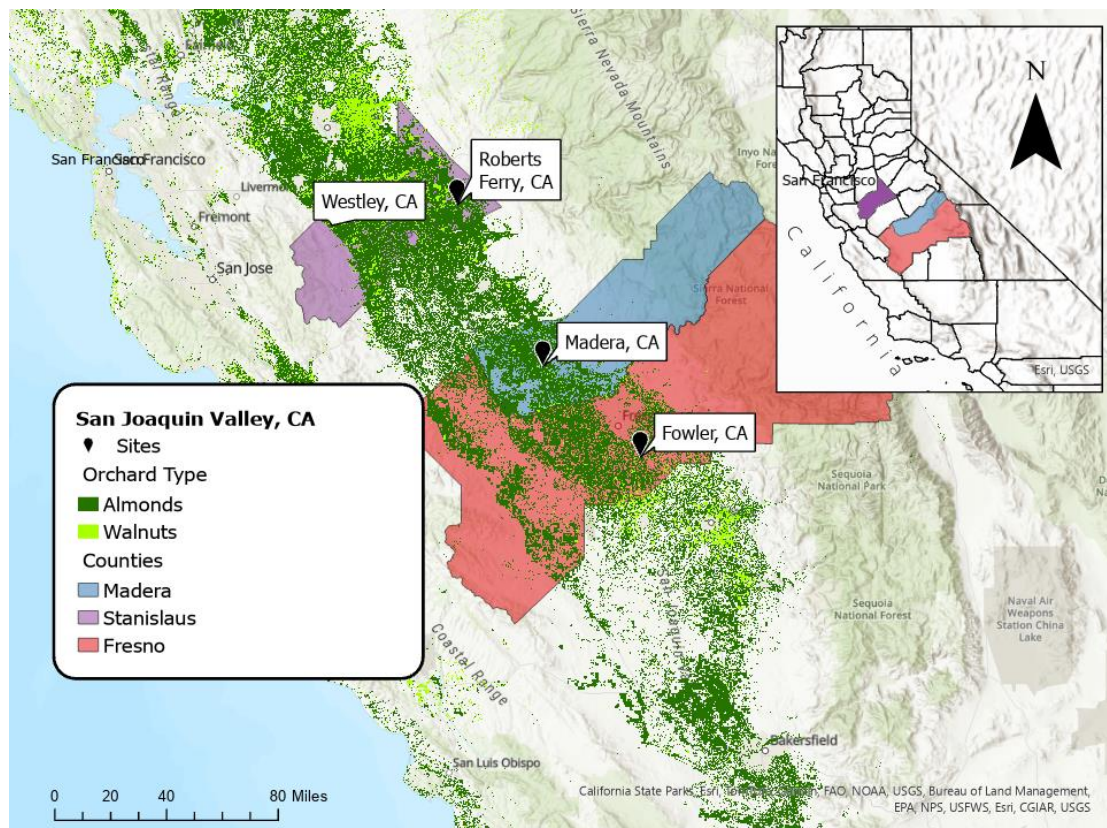


Figure 3-1. Map of site locations, with overlay of current acreage of walnut and almonds along the Central Valley.

This study investigates the combined effects of compost and cover crops on soil health indices in five commercial orchards in California, comprising four almond orchards and one walnut orchard, over four growing seasons (Figure 3-1). We hypothesize that the addition of compost and cover crops will improve the biological, chemical, and physical properties of soil health across these orchards, regardless of soil type or prior management practices.

Given the highly degraded state of soil organic carbon at the start of the study, we expect that organic matter inputs from compost and cover crops will rejuvenate the soil system and enhance soil carbon storage. Furthermore, we propose that integrating these

practices will optimize the soil nitrogen cycle by enhancing key processes such as nutrient cycling, organic matter decomposition, and soil structure formation. This integration is also expected to foster a more favorable soil environment for microbial activity, regulate soil pH, and ultimately improve the overall functionality and health of these highly productive perennial agroecosystems.

MATERIALS AND METHODS

STUDY SITES

To investigate the effects of annual compost application and cover crop seeding to soil health indicators, a four-year study was conducted in four almond and one walnut orchards along California's San Joaquin Valley (Table 3-1), hereafter referred to as “Almond 1”, “Almond 2”, “Almond 3”, “Almond 4”, and “Walnut”. The sites comprised of various soil orders, age, management and irrigation type. The San Joaquin Valley, historically the ancestral homeland of the Tejon, Kitanemuk, Yokuts, and Chamash indigenous peoples of California, is situated south of the Sacramento-San Joaquin River Delta and is drained by the San Joaquin River.

Table 3-1. Site information for the orchards within our study from 2021 to 2024 in the Central Valley of California.

Location	Site	Soil Order	Soil Series	Soil Texture	Variety	Age	Management	Irrigation
Westley, CA	Almond 1	Inceptisol	Zacharias	Fine-loam	Nonpareil & Monterey	6	Conventional	Drip
Westley, CA	Almond 2	Inceptisol	Zacharias	Fine-loam	Nonpareil & Monterey	6	Conventional	Drip
Madera, CA	Almond 3	Mollisols	Visalia	Fine Sandy-loam	Nonpareil & Monterey	12	Conventional	Micro-sprinkler
Roberts Ferry, CA	Almond 4	Entisol	Hanford	Coarse-loam	Nonpareil & Monterey	15	Organic	Micro-sprinkler
Fowler, CA	Walnuts	Entisol	Hanford	Coarse-loam	Chandler & Serr	15	Conventional	Flood

The San Joaquin Valley has a Mediterranean climate characterized by cool, wet winters and hot, dry summers. Mean annual temperatures in this region are approximately 16-14°C. The mean high temperature reaches approximately 30°C, while the mean low temperature is around 12°C (NOAA, 2021). Mean annual precipitation is 13 – 25 cm, with most precipitation occurring between November and April, with typically no rainfall during the summer dry months.

EXPERIMENTAL DESIGN

A replicated field design was employed, with two large plots (approximately 5 ha) established at each of the five orchard sites to represent conventional and compost + cover crop management practices. Within each plot, three replicate transects were designated, and three trees were tagged for sampling within each transect. To account for spatial variability, two sampling locations were selected adjacent to each of the three trees within each replicate: one in the alleyway and one on the berm. The “stacked” soil health management consisted of three annual applications of compost and cover crops. Both conventional and treatment plots were managed the same within each site regarding foliar spray, pesticides, fungicides, irrigation, and in-line fertigation.

Cover crops were seeded in the alleys in November of 2021, 2022, and 2023 at a rate of 13.8 kg/ha. A seed drill was used for the planting of the cover crops across the sites, and drilled the seeds to a max depth of 3 cm. The cover crop mix selected was soil

building mix sourced from Project *Apis m.* This mix contains 30% Triticale (x Triticosecale), 35% Bell Beans, (*Vicia faba*), 28% Peas (*Pisum sativum* or *P. arvense*), 5% Diakon Radish (*Raphanus sativus*), 1% Canola (*Brassica rapa*), and 1% Common Yellow Mustard (*Sinapis alba*). The mix contains brassicas, legumes, and grains to address soil issues such as compaction and erosion while fixing nitrogen and providing weed suppression. This mix also delivers a late source of nectar for pollinators. Weeds and cover crops were mowed within the months of March and April and again in August prior to harvest. No additional water was used to aid in the germination of the cover crops.

An organic-rich compost made from feedstocks of manure and green waste (AllGro® by Synagro, CA, USA) was applied as a surface dressing across berms and alleys at a rate of 19 Mg/ha on a dry weight basis in March of 2022, May of 2023, and April of 2024. Compost timing was dependent on weather and compost availability. One site, Almond 3, delayed compost additions in 2023, and 2024 to November, due to harvest timing. Compost was applied to the alleys and the berms evenly, using a compost/manure spreader. The compost had an average organic C content of $27 \pm 1\%$, and a total organic nitrogen content of $3.3 \pm 0.05\%$ (C:N ratio of 8:1) with a pH of 7.7, and a bulk density of 2.8 g/cm^3 . The amendment of compost added approximately 5.13 Mg C/ha, and 0.5 Mg nitrogen/ha to the soils in each application.

Soils were sampled to 0-10 and 10-30 cm depths to capture the zone of root activity in orchards. A suite of biological, chemical, and physical soil health indicators was measured prior to treatment application and for three years following treatment application. The frequency of sampling varied by indicator type from daily (e.g. soil volumetric water content) to monthly (e.g. soil inorganic nitrogen) to annually (e.g. soil carbon) depending on the temporal variability and projected rate of change of each indicator.

Soil compositing consisted of subsampling 35 g from each replicate at the 0-10 cm depth, generating a composite sample (~100 g) for every replicate transect. Including the alley and berm locations for both control and treatment plots at each site (n=6). The composited samples underwent analysis for soil pH and EC, water holding capacity, microbial respiration, permanganate oxidizable carbon (POXC), and wet aggregate stability. Soil C and nitrogen samples were not composited, and analyses were run on the full replication across sites.

SOIL MOISTURE AND PHYSICAL SOIL PROPERTIES

Soil probes (Drill & Drop, TriScan SDI-12, Sentek, SA, AUS) were installed within each replicate of the plots. These drill-and-drop probes, which utilize capacitance-based technology and site-specific calibrations, were installed and maintained by the agriculture management company SEMIOS (Modesto, CA). The sensors were buried to a depth of 1 meter and measured soil moisture, salinity, and temperature every 10 cm along the depth profile. Readings for these measurements were collected every 5 minutes and averaged to provide daily mean, minimum, maximum, and standard error for each sensor.

Soil texture was determined at each plot via the hydrometer method (Gee & Bauder, 1986) prior to treatment application. Briefly, for each depth increment, 40 g of air-dried 2 mm sieved soil was mixed with 100 mL of sodium hexametaphosphate solution, let sit for 10 minutes, then mixed with an electric mixer for 5 minutes, then added to a glass sedimentation cylinder and brought to up a total volume of 1,000 mL. At the 30 second, 1 minute, 90 minutes, and 24-hour mark, a hydrometer was used to measure the density of the liquid, and the corresponding liquid temperature was recorded. Afterwards, the measurements were then used to calculate the fractions of sand, silt, and clay via equations that were corrected for temperature.

CHEMICAL SOIL HEALTH INDICATORS

Soil pH and electrical conductivity were analyzed prior to treatment and annually for three years of treatment. A 10 ± 0.1 g sieved soil g of soil was mixed with 20 ml of deionized water and then placed on an orbital shaker for 30 min. The soil slurry dilutions (1:2) were let to equilibrate uncapped for 10 minutes, then a calibrated pH meter (Mettler Toledo SS20, OH, USA) and EC meter (Mettler Toledo SS30, OH, USA) were used to measure pH and EC.

Cation exchange capacity (CEC) was analyzed on a subset of samples of alley and berm for sites all years. Soil samples were air-dried, sieved to 2 mm, and analyzed for CEC using the barium chloride method (Rible et al., 1960) at the University of California, Davis Analytical Lab Four deionized water rinses are used to remove excess barium. A known quantity of calcium is then exchanged for barium and excess solution calcium is measured. CEC is determined by the difference in the quantity of the calcium added and the amount found in the resulting solution. The method has a detection limit of approximately 2.0 cmol kg^{-1} .

All samples, and all depths, were analyzed for soil total C and nitrogen. Soil samples were air-dried, sieved at 2 mm, and pulverized by hand with a mortar and pestle. Carbonate presence was based on effervesce after addition of 4 nitrogen HCl to soil. No sites exhibited a response to the test for carbonates, thus all soil C data presented here consist solely of organic C. Soil C and nitrogen concentrations were measured on an Elemental Analyzer (ECS 4010 CHNS-O, Costech, CA, USA) coupled to a continuous flow isotope ratio mass spectrometer (Delta-V Plus, Thermo Fischer Scientific, CA, USA) at the Stable Isotope Ecosystem Laboratory of University of California, Merced.

Soil C content was calculated using the equivalent soil mass method (ESM). The ESM method uses soil mass, volume, and the percent soil C and then uses a cubic spline of reference mass layers that is site-specific (Wendt & Hauser, 2013). The ESM method uses soil mass, volume, and soil C concentration and then uses a cubic spline of reference mass layers that was site specific (Wendt & Hauser, 2013). To calculate soil C content, we used an R script developed by Von Haden et al. (2020). Soil C and nitrogen contents were calculated independently by berm and alley locations. Cumulative soil C and nitrogen content by site was calculated using an area-weighted approach to account for the unequal land coverage of the berms and alleys. As berms account for approximately one-quarter of the area with an orchard, and alleys the remaining three quarters, the soil C

storage measured at each location was multiplied by each fraction of coverage and then summed to get a more accurate account of soil C storage in the orchards.

The potentially available ammonium (NH_4^+) and nitrate (NO_3^-) in the top 0-10cm of soil was assessed monthly in each replicate of control and treatment plots at all sites, except for Almond 4, from January 2022 to August 2024. 20 g of fresh sieved soil samples were weighed, shaken in 75 ml of 2 M potassium chloride (KCl) for 1 h, and filtered through Whatman 1 filter papers. Filtrate was analyzed for NO_3^- and NH_4^+ by microplate-colorimetric techniques using the vanadium-chloride method and salicylate-nitroprusside method, respectively (Mulvaney et al., 1996). The absorbance was read at 540 nm for NO_3^- and 650 nm for NH_4^+ on a microplate reader (BioTek Gen5 Microplate Reader, Agilent Technologies, CA, USA), then corrected to concentration from the standard curve equation.

BIOLOGICAL SOIL HEALTH INDICATORS

To assess microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) content, analyses were conducted on berm and alley locations within three out of nine replicates in both control and treatment plots within the 0-10 cm depth range, which represents the primary zone for microbial activity. Soil samples were taken each year in January and extracted within 24 h of sampling using the chloroform fumigation method (Vance et al., 1987). Extractants were stored at -4°C until ready for analysis. The thawed extractant was diluted to a ratio of 4:1 sample to deionized water. Soils were run with blanks and standards on a total organic carbon analyzer (TOC-L) (Shimadzu Scientific, Japan) in the Environmental Analytical Lab at the University of California, Merced. MBC and MBN were corrected with a standard curve of known concentrations for total nitrogen and total organic C. We used an extraction efficiency factor of (keC) of 0.45 for MBC and 0.55 for MBN (keN) (Beck et al., 1997).

Permanganate Oxidizable C (*POXC*) was analyzed on the composited samples once before treatment, and yearly for three years. We used the method described by (Weil et al., 2023). Briefly, 2.5 ± 0.01 g air-dried sieved soil was added to 20 mL of 0.2 M potassium permanganate (KMnO_4) and deionized water and then were shaken for 2 minutes then let settled for 10 minutes. After settling, 0.5 mL of supernatant was transferred from the centrifuge tube, diluted with 49.5 mL of deionized water, and then 200 μL of the supernatant was read on a microplate reader at 550 nm (Agilent BioTek Gen5 Microplate Reader, Agilent Technologies, Santa Clara, CA, USA). Sample absorbance was then corrected with known concentrations of KMnO_4 , and included the milligrams of C oxidized by 1 mole of MnO_4 (9,000 mg C/mol) and reported as mg KMnO_4 reduced per kg soil.

Short-term C mineralization of soils was measured on composited soil samples before treatment and the three years following. A Picarro multi-gas analyzer and Soil Flux Processor (G2508, Picarro, CA, USA) were used to measure the flux of microbial respiration after a wetting event for 4 days. For the lab study, 30 g air dried, sieved soil was placed into half-pint jars, with specialized chamber lids. The chamber lid is fitted with two 1/4" tube fittings (Swagelok, Solon, OH) each connected to an inlet or outlet 1/4" tube with sample air flowing at a rate of 275 ml min^{-1} . Briefly, the baseline was

determined on the dry fluxes that were measured the day 0 of the study. The next day deionized water was added to achieve 60% water holding capacity (WHC). The maximum WHC of each soil was determined prior to the study for each site by calculating the volume of water retained after 1h (no drips) via a percolation method with a funnel, filter paper, and drainage setup. Concentrations of CO₂, N₂O, and CH₄ were measured for 5-minute intervals for each sample. After taking a measurement, gas concentrations were allowed to return to ambient concentrations before the next measurement. Gas fluxes (nmol m⁻² s⁻¹) were calculated in the Picarro Soil Flux Processor program using the exponential model developed by Hutchinson and Mosier to account for nonlinear changes in headspace concentration, and cumulative flux concentrations were calculated with linear line. The cumulative flux over the four days was calculated by trapezoidal interpolation between measurement dates.

PHYSICAL SOIL HEALTH INDICATORS

Wet aggregate stability was analyzed on composited samples prior to treatment and three years after. Air dried soils were processed by the wet sieving method using a RO-TAP RX-29 mechanical shaker (W.S Tyler, Ohio). 20 gs of air-dried and sieved soil (4.75 mm) was placed onto the topmost of three sequentially arranged sieves of 2.00, 1.00, and 0.25 mm and shaken at 45 revolutions per minute for 5 min. Subsequently, the wet stable aggregate (WSA) fractions (large macroaggregates (4.75–2.00 mm = Lma), medium macroaggregates (2.00–1.00 mm = Mma), small macroaggregates (1.00–0.25 mm = Sma), and microaggregates, including silt and clay (<0.25 mm = Mia), were obtained, weighed, and expressed in a percentage (%) to the initial sample weight.

The bulk density for each soil sample was determined using the dimensions of the custom steel cores for our study. They had a diameter of 8 cm, and the height of the cores was adjusted based on the sampling depth (either 10 cm or 30 cm). The volume of each core was calculated using these dimensions. To account for soil moisture content, a subsample of soil was air-dried, and then used to correct the bag of soil. The moisture-corrected dry soil mass was then divided by the core volume to calculate the bulk density, expressed in grams per cubic centimeter (g/cm³).

STATISTICAL ANALYSIS

All analyses were conducted using R Statistical Software (v4.4.2; R Core Team, 2021). The primary independent variable was management practice, categorized as either Conventional or Compost + Cover Crops, while various soil health indices served as numeric response variables. The study followed a longitudinal design, with repeated measurements collected over three years to capture temporal variations and evaluate the impact of soil health practices. For each response variable, normality was assessed using the Shapiro-Wilk test. If the variable failed to meet normality assumptions, transformations (log, natural log, or square root) were applied. Transformed variables that achieved normality were analyzed using parametric tests, while those that remained non-normal were analyzed with non-parametric approaches. Response variables that met the assumptions of parametric testing (EC, pH, POXC, volumetric water content, and mineralized CO₂) were analyzed using linear mixed models (LMMs). These models included Site as a random effect to account for variability between sites. Fixed effects

included management, location (Alley vs. Berm), soil depth, and year, as well as their interactions. The significance of fixed effects for LMM models was tested using Type II ANOVA (Girden, 1992). For variables that could not be transformed to meet parametric assumptions, generalized linear mixed models (GLMMs) were employed to account for their distribution characteristics. GLMMs were fitted using the `glmer` function from the `lme4` package (Bates et al., 2015), specifying Site as a random effect. The significance of fixed effects was tested using Type II Wald Chi-square tests via the `Anova` function in the `car` package (Fox & Weisberg, 2019). Pairwise comparisons for both models were conducted using the `emmeans` package (Lenth, 2021) to identify significant differences between management practices across years, locations (Alley vs. Berm), and soil depths. The `emmeans` analysis provided contrasts with estimates, standard errors, z-values, and p-values to highlight significant effects.

RESULTS

PHYSICAL

Bulk density at the onset of the study ranged from a low of 1.29 g cm^3 to a high of 1.79 g cm^3 in the 10 cm depth, the 30 cm depth ranged from a low of 1.34 g cm^3 to a high of 1.73 g cm^3 (). By the end of the study, the average bulk density of the compost + cover crops plots in the 10 cm depth were 1.22 g cm^3 , and 1.5 g cm^3 for the 30 cm depth. The analysis found that all factors in the study significantly influence BD, Year ($F= 117, p < 0.01$), Management ($F= 187, p < 0.01$), and Depth ($F= 106, p < 0.01$), and Location i.e. berm or alley ($F = 18, p < 0.01$). There was a significant decrease in BD for the 10 cm depth across sites from the baseline to the final sampling was 0.15 g cm^3 , whereas there was a slight increase in in the 30 cm depth of 0.03 g cm^3 .

Table 3-2 .Physical properties of bulk density (BD) and wet stable aggregates (WSA), organized by management plots of conventional (C) and compost + cover crops (T), and depth (cm) for the location of alley and berm for the four years of sampling.

Site	Plot	Indice	Depth	2021		2022		2023		2024		
				Alley	Berm	Alley	Berm	Alley	Berm	Alley	Berm	
Almond 1	C	BD	10	1.63 ± 0.02	1.63 ± 0.02	1.26 ± 0.06	1.27 ± 0.07	1.26 ± 0.06	1.1 ± 0.05	1.64 ± 0.04	1.4 ± 0.07	
			30	1.47 ± 0.06	1.47 ± 0.04	1.21 ± 0.07	1.17 ± 0.08	1.33 ± 0.04	1.26 ± 0.03	1.64 ± 0.02	1.4 ± 0.02	
			10	1.36 ± 0.02	1.36 ± 0.03	0.94 ± 0.08	1.04 ± 0.05	1.06 ± 0.08	1.04 ± 0.05	1.21 ± 0.03	1.18 ± 0.04	
	T	BD	30	1.34 ± 0.08	1.34 ± 0.02	1.36 ± 0.07	1.27 ± 0.06	1.27 ± 0.04	1.21 ± 0.05	1.71 ± 0.02	1.42 ± 0.01	
			WSA	2 mm	2.4 ± 0.13	2.7 ± 0.33	2.5 ± 0.01	2.3 ± 0.12	3 ± 0.01	3 ± 0.03	3 ± 0.01	3 ± 0.03
				1 mm	0.7 ± 0.13	0.8 ± 0.12	0.4 ± 0.01	0.5 ± 0.03	0.7 ± 0.03	0.3 ± 0.01	0.7 ± 0.03	0.3 ± 0.02
	0.25 mm	0 ± 0		0 ± 0	0 ± 0	0 ± 0	0 ± 0.01	0 ± 0	0 ± 0.01	0 ± 0		
	< 0.25 mm	26.9 ± 0.26		26.5 ± 0.45	27.1 ± 0.02	27.2 ± 0.2	26.3 ± 0.04	26.7 ± 0.04	26.3 ± 0.05	26.7 ± 0.04		
	2 mm	2.3 ± 0.05		2.3 ± 0.19	2.9 ± 0.05	2.8 ± 0.03	3.4 ± 0.03	5.2 ± 0.31	3.4 ± 0.03	5.2 ± 0.31		
	1 mm	0.5 ± 0.05		0.4 ± 0.12	0.7 ± 0.05	0.9 ± 0.03	1 ± 0.03	0.9 ± 0.03	1.1 ± 0.03	1 ± 0.02		
C	WSA	0.25 mm	0 ± 0	0 ± 0	2.3 ± 0.12	0.6 ± 0.04	2.2 ± 0.07	0.3 ± 0.05	2.9 ± 0.13	0.5 ± 0.07		
		< 0.25 mm	27.2 ± 0.01	27.3 ± 0.21	24.1 ± 0.12	25.7 ± 0.08	23.4 ± 0.08	23.7 ± 0.3	22.6 ± 0.11	23.3 ± 0.37		
		10	2.4 ± 0.13	2.7 ± 0.33	2.5 ± 0.01	2.3 ± 0.12	3 ± 0.01	3 ± 0.03	3 ± 0.01	3 ± 0.03		
Almond 2	C	BD	10	1.79 ± 0.02	1.79 ± 0.07	1.36 ± 0.08	1.26 ± 0.13	1.26 ± 0.06	1.1 ± 0.05	1.64 ± 0.02	1.4 ± 0.02	
			30	1.51 ± 0.01	1.51 ± 0.02	1.54 ± 0.06	1.39 ± 0.07	1.64 ± 0.06	1.48 ± 0.06	1.64 ± 0.01	1.4 ± 0.07	
			10	1.29 ± 0.02	1.29 ± 0.04	1.03 ± 0.07	1 ± 0.06	1 ± 0.03	1.06 ± 0.06	1.21 ± 0.05	1.18 ± 0.02	
	T	BD	30	1.55 ± 0.03	1.55 ± 0.02	1.19 ± 0.07	1.09 ± 0.06	1.22 ± 0.05	1.21 ± 0.03	1.71 ± 0.02	1.42 ± 0.04	
			WSA	2 mm	2.4 ± 0.13	2.7 ± 0.34	2.6 ± 0.02	2.3 ± 0.12	3 ± 0.01	3 ± 0.03	3 ± 0.01	3 ± 0.03
				1 mm	0.8 ± 0.18	0.8 ± 0.12	0.4 ± 0.01	0.5 ± 0.03	0.7 ± 0.03	0.3 ± 0.01	0.7 ± 0.03	0.3 ± 0.01
	0.25 mm	0 ± 0		0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0.01	0 ± 0		
	< 0.25 mm	26.8 ± 0.32		26.5 ± 0.45	27 ± 0.03	27.2 ± 0.09	26.3 ± 0.04	26.7 ± 0.04	26.3 ± 0.04	26.7 ± 0.03		
	2 mm	2.3 ± 0.05		2.3 ± 0.19	2.8 ± 0.06	2.8 ± 0.03	3.4 ± 0.03	4.8 ± 0.03	3.4 ± 0.03	4.8 ± 0.03		
	1 mm	0.5 ± 0.07		0.4 ± 0.08	0.7 ± 0.04	0.9 ± 0.03	1 ± 0.03	0.9 ± 0.03	1.1 ± 0.04	1 ± 0.03		
C	WSA	0.25 mm	0 ± 0	0 ± 0	2.2 ± 0.06	0.4 ± 0.09	2.2 ± 0.07	0.3 ± 0.04	2.9 ± 0.14	0.6 ± 0.06		
		< 0.25 mm	27.2 ± 0.03	27.3 ± 0.2	24.3 ± 0.05	25.9 ± 0.11	23.4 ± 0.08	24 ± 0.04	22.6 ± 0.15	23.6 ± 0.06		
		10	2.4 ± 0.13	2.7 ± 0.34	2.6 ± 0.02	2.3 ± 0.12	3 ± 0.01	3 ± 0.03	3 ± 0.01	3 ± 0.03		
Almond 3	C	BD	10	1.3 ± 0.02	1.3 ± 0.02	1.25 ± 0.04	1.15 ± 0.02	1.24 ± 0.02	1.21 ± 0.04	1.3 ± 0.12	1.42 ± 0.03	
			30	1.5 ± 0.02	1.5 ± 0.04	1.42 ± 0.05	1.31 ± 0.06	1.39 ± 0.06	1.27 ± 0.02	1.31 ± 0.03	1.42 ± 0.04	
			10	1.49 ± 0.01	1.49 ± 0.02	1.29 ± 0.2	0.98 ± 0.03	1.01 ± 0.05	0.92 ± 0.04	1.24 ± 0.02	1.2 ± 0.03	
	T	BD	30	1.39 ± 0.02	1.39 ± 0.03	1.2 ± 0.05	1.05 ± 0.02	1.17 ± 0.03	1.05 ± 0.03	1.14 ± 0.03	1.32 ± 0.01	
			WSA	2 mm	5 ± 0.39	4 ± 0.21	4.3 ± 0.02	4 ± 0.01	5.5 ± 0.15	3.8 ± 0.08	4.3 ± 0.02	4 ± 0.01
				1 mm	0.3 ± 0.01	0.8 ± 0.04	1.3 ± 0	0.8 ± 0.01	0.9 ± 0	0.6 ± 0	1.2 ± 0.09	0.8 ± 0.01
	0.25 mm	0 ± 0		0 ± 0	0 ± 0.01	0 ± 0	0 ± 0	0 ± 0	0 ± 0.01	0 ± 0		
	< 0.25 mm	24.7 ± 0.4		25.3 ± 0.23	24.4 ± 0.03	25.1 ± 0.02	23.6 ± 0.15	25.6 ± 0.08	24.5 ± 0.11	25.1 ± 0.02		
	2 mm	7 ± 0.04		6.3 ± 0.04	7.2 ± 0	5.8 ± 0	7.7 ± 0.18	7.6 ± 0	7.2 ± 0	5.8 ± 0		
	1 mm	0.9 ± 0.06		0.5 ± 0.17	0.6 ± 0.01	0.6 ± 0.03	1.2 ± 0.23	1.2 ± 0.07	1.9 ± 0.21	2 ± 0.26		
C	WSA	0.25 mm	0 ± 0	0 ± 0	0 ± 0.02	0.1 ± 0.01	0.9 ± 0.29	1 ± 0.09	0.5 ± 0.24	0.5 ± 0.13		
		< 0.25 mm	22 ± 0.1	23.2 ± 0.14	22.2 ± 0.04	23.5 ± 0.03	20.3 ± 0.68	20.2 ± 0.16	20.5 ± 0.26	21.7 ± 0.38		
		10	1.47 ± 0.09	1.47 ± 0.02	1.49 ± 0.08	1.29 ± 0.08	1.35 ± 0.09	1.45 ± 0.08	1.82 ± 0.02	1.37 ± 0.02		
Almond 4	C	BD	10	1.56 ± 0.12	1.56 ± 0.06	1.46 ± 0.11	1.39 ± 0.13	1.56 ± 0.09	1.54 ± 0.07	1.67 ± 0.08	1.6 ± 0.03	
			30	1.44 ± 0.02	1.44 ± 0.02	1.28 ± 0.09	1.23 ± 0.09	1.21 ± 0.05	1.1 ± 0.03	1.2 ± 0.05	1.33 ± 0.02	
			10	1.44 ± 0.02	1.44 ± 0.02	1.28 ± 0.09	1.23 ± 0.09	1.21 ± 0.05	1.1 ± 0.03	1.2 ± 0.05	1.33 ± 0.02	
	T	BD	30	1.5 ± 0.07	1.5 ± 0.04	1.41 ± 0.09	1.44 ± 0.1	1.42 ± 0.07	1.3 ± 0.07	1.51 ± 0.01	1.44 ± 0.05	
			WSA	2 mm	5 ± 0.39	4 ± 0.21	4.3 ± 0.02	4 ± 0.01	5.5 ± 0.15	3.8 ± 0.08	4.3 ± 0.02	4 ± 0.01
				1 mm	0.3 ± 0.01	0.8 ± 0.04	1.3 ± 0	0.8 ± 0.01	0.9 ± 0	0.6 ± 0	1.2 ± 0.09	0.8 ± 0.01
	0.25 mm	0 ± 0		0 ± 0	0 ± 0.01	0 ± 0	0 ± 0	0 ± 0	0 ± 0.01	0 ± 0		
	< 0.25 mm	24.7 ± 0.4		25.3 ± 0.23	24.4 ± 0.03	25.1 ± 0.02	23.6 ± 0.15	25.6 ± 0.08	24.5 ± 0.11	25.1 ± 0.02		
	2 mm	7 ± 0.04		6.3 ± 0.04	7.2 ± 0	5.8 ± 0	7.7 ± 0.18	7.6 ± 0	7.2 ± 0	5.8 ± 0		
	1 mm	0.9 ± 0.06		0.5 ± 0.17	0.6 ± 0.01	0.6 ± 0.03	1.2 ± 0.23	1.2 ± 0.07	1.9 ± 0.21	2 ± 0.26		
C	WSA	0.25 mm	0 ± 0	0 ± 0	0 ± 0.02	0.1 ± 0.01	0.9 ± 0.29	1 ± 0.09	0.5 ± 0.24	0.5 ± 0.13		
		< 0.25 mm	22 ± 0.1	23.2 ± 0.14	22.2 ± 0.04	23.5 ± 0.03	20.3 ± 0.68	20.2 ± 0.16	20.5 ± 0.26	21.7 ± 0.38		
		10	1.47 ± 0.09	1.47 ± 0.02	1.49 ± 0.08	1.29 ± 0.08	1.35 ± 0.09	1.45 ± 0.08	1.82 ± 0.02	1.37 ± 0.02		
Walnut	C	BD	10	1.58 ± 0.02	1.58 ± 0.06	1.44 ± 0.1	1.59 ± 0.06	1.32 ± 0.07	1.03 ± 0.1	1.34 ± 0.04	1.3 ± 0.02	
			30	1.74 ± 0.07	1.74 ± 0.02	1.65 ± 0.07	1.41 ± 0.07	1.28 ± 0.03	1.38 ± 0.04	1.91 ± 0.02	1.61 ± 0.05	
			10	1.33 ± 0.02	1.33 ± 0.03	1.08 ± 0.04	1.06 ± 0.11	1.06 ± 0.03	1.09 ± 0.05	1.13 ± 0.03	1.36 ± 0.02	
	T	BD	30	1.6 ± 0.01	1.6 ± 0.02	1.44 ± 0.12	1.41 ± 0.15	1.18 ± 0.04	1.13 ± 0.04	1.77 ± 0.02	1.55 ± 0.01	
			WSA	2 mm	9.3 ± 0.33	9.2 ± 0.38	9.5 ± 0	9.2 ± 0.33	9.3 ± 0.33	10.3 ± 0.32	9.3 ± 0.33	10.3 ± 0.32
				1 mm	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0	0.1 ± 0	0.1 ± 0	0.1 ± 0
	0.25 mm	0 ± 0		0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0.01		
	< 0.25 mm	20.6 ± 0.32		20.7 ± 0.37	20.4 ± 0.01	20.7 ± 0.34	20.6 ± 0.33	19.5 ± 0.32	20.6 ± 0.33	19.5 ± 0.32		
	2 mm	9 ± 0.31		9.2 ± 0.22	10.3 ± 0.32	9.9 ± 0.34	10.6 ± 0.07	12.1 ± 0.01	10.6 ± 0.07	12.1 ± 0.01		
	1 mm	0.1 ± 0.02		0.2 ± 0.06	0.4 ± 0.1	0.3 ± 0.03	0 ± 0.01	0.1 ± 0.01	0.1 ± 0.03	0.2 ± 0.01		
C	WSA	0.25 mm	0 ± 0	0 ± 0	0.2 ± 0.03	0.2 ± 0.02	0 ± 0.01	0.1 ± 0.01	0.1 ± 0.02	0.4 ± 0.26		
		< 0.25 mm	20.9 ± 0.32	20.6 ± 0.26	19.2 ± 0.33	19.6 ± 0.35	19.3 ± 0.07	17.7 ± 0.03	19.2 ± 0.07	17.3 ± 0.25		
		10	1.58 ± 0.02	1.58 ± 0.06	1.44 ± 0.1	1.59 ± 0.06	1.32 ± 0.07	1.03 ± 0.1	1.34 ± 0.04	1.3 ± 0.02		

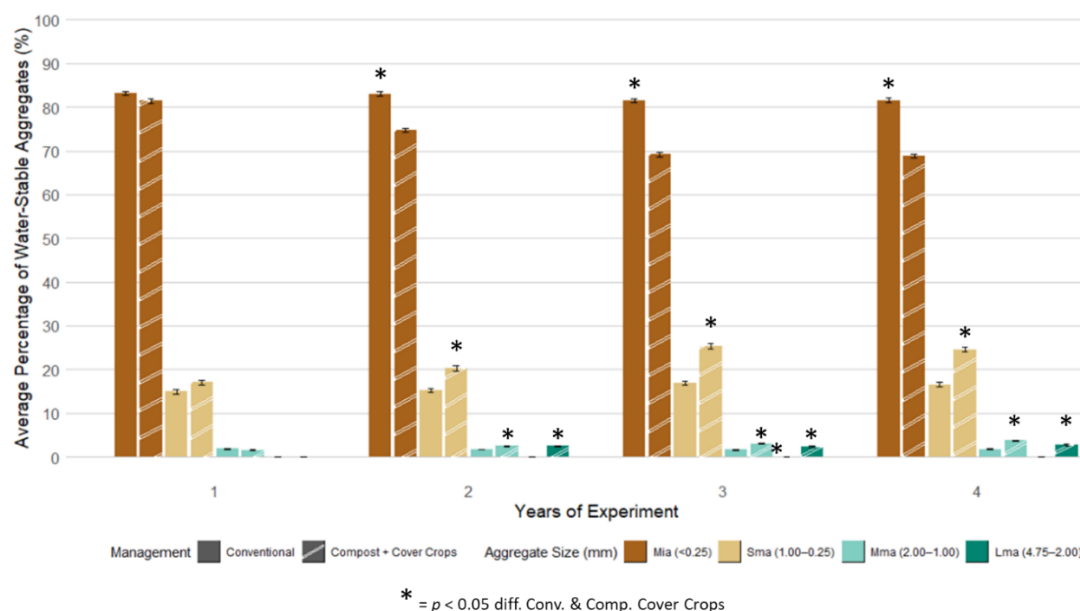


Figure 3-2. The percentage (%) of average wet aggregate stability (WSA) averaged across sites over the four years of the study. Management is denoted by patterns, with plain bars representing conventional plots and crosshatching representing plots with compost and cover crops. This figure displays averages across berm and alley locations. The asterisk (*) indicates a significant difference between conventionally managed sites and those with compost and cover crops.

Wet aggregate stability in the compost + cover crop treated plots increased over time, characterized by a shift towards larger aggregates (ϕ). Specifically, Small Macroaggregates (Sma) increased by 8.4 ± 2 %, followed by gains in Large Macroaggregates (Lma) by 2.1 ± 0.5 % and Medium Macroaggregates (Mma) by 1.3 ± 0.2 %. Conversely, Microaggregates (Mia) decreased by 11.7 % in the treatment plots over the study period. Management, sample location (alley/berm) and year exerted significant influences on these changes ($p < 0.05$), as illustrated in Figure 2. There were significant differences in aggregate stability observed between sample locations (alley vs. berm), with berms having a higher level of stable aggregates across size classes ($p = 0.03$).

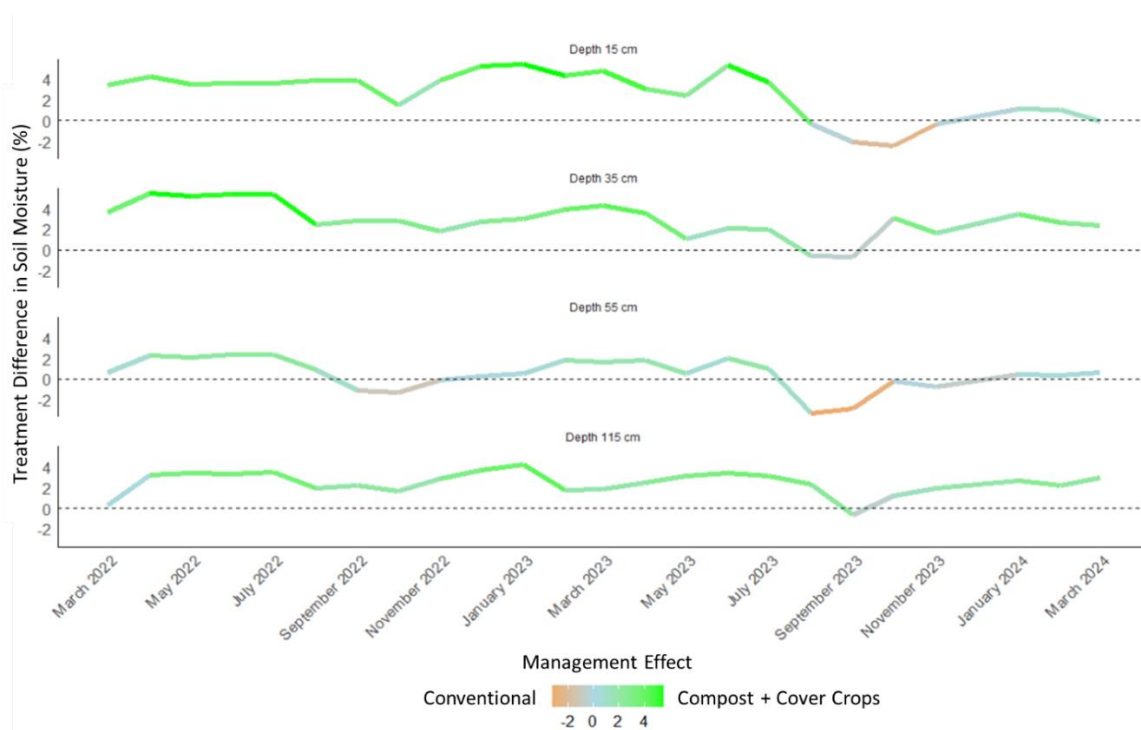


Figure 3-3. Volumetric water content % for the months of August and September across years 2, 3, and 4 for the four sites. The line represents the treatment difference between management plots of compost + cover crops and conventional, across all sites in the study from March 2022 to March 2024. When the line is above zero, it denotes an increase in soil moisture for the compost + cover crops plots.

VOLUMETRIC WATER CONTENT

There was a significant effect of management (Compost + Cover Crops vs Conventional), depth, season (Spring, Summer, Fall, Winter), and site on volumetric water content (VWC) (%). The management approach demonstrated a significant effect on VWC ($F = 19129.639$, $p < 0.001$). Depth also significantly affected VWC ($F = 8550.015$, $p < 0.001$), showing considerable variation across soil depths. Similarly, the season had a significant effect on VWC ($F = 24747.737$, $p < 2.2e-16$), suggesting that VWC varies significantly by season with higher levels in Spring and Winter. The interaction between management and depth was significant ($F = 1489.530$, $p < 2.2e-16$). The trend was that the uppermost of the soil profile, the 0-35 depth, was higher with the Compost + Cover Crops, and the deepest soils i.e. 115cm. The interaction between management and season was significant ($F = 611.271$, $p < 2.2e-16$), suggesting that the management effect on water volume differs across seasons, while the interaction between depth and season was highly significant ($F = 1439.552$, $p < 2.2e-16$), showing that seasonal effects on water content vary by depth. Compost + Cover Crops consistently exhibited higher VWC compared to Control (Conventional Management) across all seasons. In Fall, the estimate was -0.753 ($p < 0.0001$), indicating significantly higher water content in the Treatment. In Spring, the estimate was -1.947 ($p < 0.0001$). In

Summer, the estimate was -1.966 ($p < 0.0001$), and in Winter, the estimate was -1.932 ($p < 0.0001$).

CHEMICAL

PH, EC, SOIL C & N CONCENTRATIONS, AND SOIL C CONTENT RESULTS.

Table 3-3. Chemical soil properties for the four years of the study, organized by alley and berm locations and management plots of conventional (C) and Compost + Cover crops (T).

Site	Plot	Indice	2021		2022		2023		2024	
			Alley	Berm	Alley	Berm	Alley	Berm	Alley	Berm
Almond 1	C	pH	7.2 ± 0	6.39 ± 0.03	7.13 ± 0.06	7.81 ± 0.02	6.88 ± 0.09	5.64 ± 0	7.34 ± 0	6.98 ± 0.02
			T	7.65 ± 0.01	7.07 ± 0	6.91 ± 0.03	6.81 ± 0.04	7.14 ± 0.01	6.78 ± 0	6.9 ± 0.02
	C	EC	269.75 ± 0.38	702.2 ± 6.99	633.75 ± 0.26	654.55 ± 0.26	165.15 ± 0.14	166.25 ± 0.03	188.75 ± 0.09	245.9 ± 0.29
			T	472.6 ± 5.43	467.7 ± 16.57	677.7 ± 0.12	955.9 ± 0.81	315.95 ± 0.26	780.5 ± 0.52	591.25 ± 0.61
	C	CEC	26.2	23.3	24.8	15.9	23.7	23.7	20.3	20.5
T			24.4	23.8	25.5	256	25.1	25.8	26.1	28.6
Almond 2	C	pH	7.08 ± 0.01	7.14 ± 0.01	7.37 ± 0.01	7.29 ± 0.01	7.19 ± 0.01	7.26 ± 0.01	7.53 ± 0.03	7.96 ± 0
			T	7.58 ± 0.02	7.31 ± 0.01	6.89 ± 0.01	7.04 ± 0.01	7.22 ± 0	6.57 ± 0.01	7.07 ± 0.01
	C	EC	486.55 ± 4.13	749.4 ± 9.58	633.23 ± 0.58	654 ± 0.61	165.17 ± 0.15	166.17 ± 0.09	188.57 ± 0.2	245.93 ± 0.29
			T	436.65 ± 6.38	424.15 ± 2.11	676.73 ± 0.97	954.57 ± 1.56	315.73 ± 0.34	780.07 ± 0.68	589.77 ± 1.6
	C	CEC	22.5	24.5	20.7	20.3	21.2	15.4	16.6	20.5
T			24.4	23.8	25.7	25.9	23.8	26.8	25.3	26.8
Almond 3	C	pH	6.43 ± 0.03	6.56 ± 0.06	6.71 ± 0.03	6.64 ± 0	7.32 ± 0.03	7 ± 0.02	6.98 ± 0.02	7.41 ± 0.01
			T	7.39 ± 0.05	6.74 ± 0.03	6.09 ± 0.03	6.24 ± 0	7.09 ± 0	6.58 ± 0.01	6.43 ± 0.01
	C	EC	851.95 ± 8.63	851.95 ± 8.63	235.07 ± 0.23	1122.43 ± 479.27	241.97 ± 3.51	84.98 ± 0.49	216.87 ± 30.66	167.1 ± 0.56
			T	673.4 ± 1.67	2294 ± 0.58	2703.67 ± 5.49	7965 ± 39	3000.13 ± 1.05	2319.67 ± 0.33	1968.33 ± 10.74
	C	CEC	12.3	10.6	10.2	15.3	13.7	11.4	16.1	15.1
T			16.4	13.8	14.4	14.4	16.5	24.9	18.5	26.9
Almond 4	C	pH	6.8 ± 0.03	6.54 ± 0.03	6.55 ± 0.01	7.01 ± 0.01	7 ± 0.01	6.59 ± 0.22	6.8 ± 0.01	7.09 ± 0.08
			T	7.15 ± 0	6.43 ± 0.07	6.78 ± 0.01	5.92 ± 0.01	6.42 ± 0.13	6.85 ± 0.32	7.01 ± 0.08
	C	EC	96.46 ± 0.42	98.25 ± 22.95	223.77 ± 0.13	529.17 ± 396.92	171.2 ± 0.15	107.47 ± 0.03	68.59 ± 0.1	134.7 ± 0.87
			T	68.59 ± 0.1	134.7 ± 0.87	224.83 ± 7.12	300.43 ± 0.44	943.6 ± 1.15	560.37 ± 0.54	326.07 ± 0.73
	C	CEC	8.8	9.1	7.5	6.6	9.7	7.5	9.5	9.7
T			7.8	8.6	9.2	4.8	12	11	14.2	11.1
Walnut	C	pH	7.28 ± 0.01	7.68 ± 0.02	7.18 ± 0.01	7.48 ± 0.04	7.44 ± 0.08	6.75 ± 0.01	6.33 ± 0	6.6 ± 0.02
			T	6.96 ± 0.04	7.16 ± 0.01	6.71 ± 0.01	6.8 ± 0.01	6.38 ± 0.01	7.1 ± 0.01	7.42 ± 0
	C	EC	389.15 ± 1.82	209.65 ± 0.03	258.73 ± 0.35	150.87 ± 0.32	176.57 ± 0.23	80.22 ± 0.22	221.73 ± 0.38	142.77 ± 0.28
			T	433.3 ± 0.46	136.05 ± 0.49	710.57 ± 1.25	262.93 ± 0.49	322.2 ± 0.35	159.83 ± 0.03	622.27 ± 0.15
	C	CEC	12.6	9.3	10	7.1	13.1	8.1	13	7.3
T			12.1	5.3	13.8	6.9	14.1	9	14.9	9.7

The average pH in Conventional plots increased from 6.9 in 2021 to 7.1 in 2024, resulting in an average increase of 0.2 ± 0.3 (Table 3-3). In contrast, the average pH in Compost + Cover Crops plots decreased, from 7.1 in 2021 to 6.8 in 2024, with an average difference of -0.3 ± 0.3 . There was a significant interaction effect of management and year on pH, with year 1 ($p < 0.05$) and year 3 ($p = 0.04$), both having a significant decrease in the management plots compared to control. There was not a significant difference between alley and berm locations. Electrical conductivity (EC) decreased in Conventional plots over time, with an average reduction of 313.6 ± 114.5 $\mu\text{S}/\text{cm}$ from 2021 to 2024. In contrast the compost + cover crops plots had a significant positive effect on EC with an average increase 445.3 ± 311.2 $\mu\text{S}/\text{cm}$ by year three ($p < 0.05$). There was a significant interaction effect of management and year, with year 12 ($p = 0.015$) and year 3 ($p < 0.01$), both having a significant increase in the management plots compared to control. Soil CEC across sites at the beginning of the study was highly variable, ranging from a low of 12 cmol kg^{-1} to a high of 25 cmol kg^{-1} (Table 3-3). The GLMM revealed a significant main effect of year 3 ($Z=0.04$) indicating a significant increase in CEC in 2024 compared to the baseline year. A Type II Wald Chi-square test showed significant

effects of Management ($\chi^2=20.40$, $df=1$, $p<0.001$), Location ($\chi^2=10.09$, $df=1$, $p<0.001$), and Date ($\chi^2=15.73$, $df=3$, $p<0.001$) on CEC. A significant interaction between Management and Date ($\chi^2=9.83$, $df=3$, $p=0.02$) indicated that the effect of Management varied across years with an increase over time with the management of compost + cover crops (Figure 3-4). In year 1, there was no significant difference, however, in year 2, CEC was significantly higher under Compost + Cover Crops compared to Conventional management (estimate = 0.15, $p = 0.0406$).

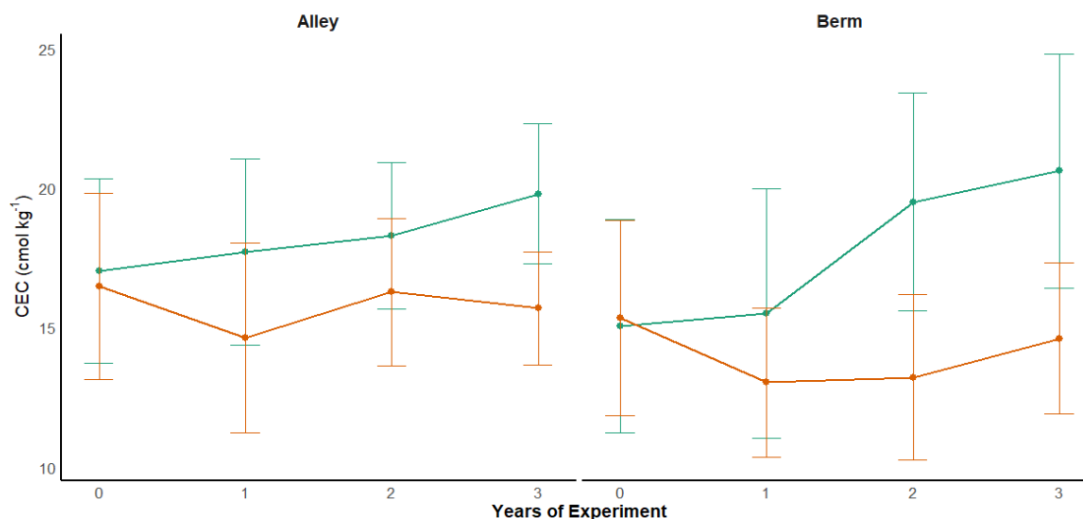


Figure 3-5. Cation Exchange Capacity (CEC) $cmol\ kg^{-1}$ for all sites. Color represents Compost + Cover crops management as green, and conventional as orange.

This trend continued in year 2 (estimate = 0.26, $p < 0.05$) and year 3 (estimate = 0.28, $p < 0.05$), with Compost + Cover Crops consistently showing significantly higher CEC values compared to Conventional management. By year three, there was an average increase of $25.7 \pm 14.5\%$ for the alleys CEC and $48 \pm 16.9\%$ for the berms for plots receiving Compost + Cover Crops, whereas conventional plots saw a small increase in the alleys of $1.3 \pm 9.2\%$ and a decrease in berms of $0.16 \pm 11.7\%$.

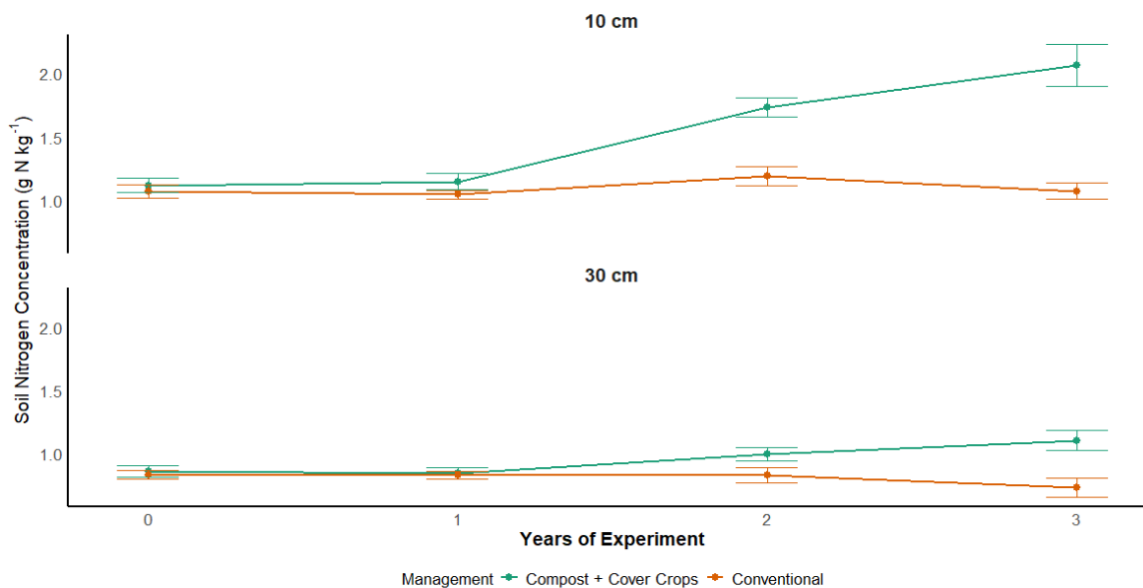


Figure 3-6. Soil N concentration (N / kg dry soil) was averaged across the 5 sites for the baseline and the 3 years of the study. Color represents Compost + Cover Crops management as green and Conventional as orange. Depth is denoted by line type, solid as the 10 cm depth, and dashed as the 30 cm depth. Error bars are the mean \pm the standard error.

POTENTIALLY AVAILABLE NITROGEN

Total soil nitrogen concentration (g N kg^{-1}) was evaluated at baseline across different sites at depths of 10 cm and 30 cm (Figure 3-6). At 10 cm depth, total nitrogen concentration ranged from 0.54% to 1.44%, with an average of $0.89 \pm 0.08\%$. At 30 cm depth, total nitrogen concentration ranged from 0.61% to 1.35%, with an average of $0.89 \pm 0.04\%$. Several factors significantly influenced soil nitrogen concentration throughout the study. Year had a notable effect ($\chi^2=57.7$, $df=3$, $p<0.001$), management ($\chi^2=45.6$, $df=1$, $p<0.001$), and depth ($\chi^2=257.4$, $df=1$, $p<0.001$). Significant interactions were found between year and management ($\chi^2=36.3$, $df=3$, $p<0.001$), year and depth ($\chi^2=32.4$, $df=3$, $p<0.001$). Pairwise comparisons between management practices (Compost + Cover Crops vs. Conventional) were conducted for each year and depth, with results presented on the log-transformed scale. By year 2, Compost + Cover Crops resulted in significantly higher nitrogen content ($p < 0.0001$) and an even greater difference in year 3 ($p < 0.0001$). At 30 cm depth, no significant differences were detected until year 3 ($p < 0.05$). For the Compost + Cover Crops plots, the percent increase in soil nitrogen concentration from 2021 to 2024 was $84.7\% \pm 15.6\%$ at the 10 cm depth and $28.2\% \pm 10.5\%$ at the 30 cm depth. In contrast, for the conventional plots, the percent increase in soil nitrogen concentration from 2021 to 2024 was $0.1\% \pm 7.5\%$ at the 10 cm depth and a decrease of $-11.9\% \pm 9.6\%$ at the 30 cm depth.

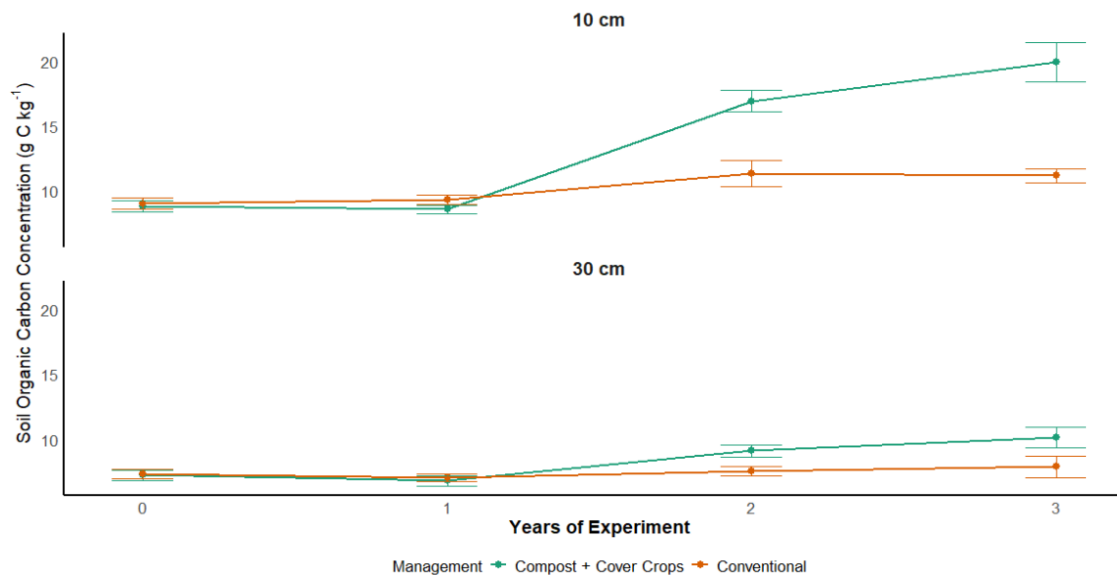


Figure 3-7. Average soil C concentration (g C kg⁻¹) of soils across sites in the study through time. Color represents compost + cover crops management as green, and conventional as orange. Depth is denoted by line type, solid as the 10 cm depth, and dashed as the 30 cm depth. Error bars are the mean \pm the standard error.

SOC CONCENTRATION

Total SOC concentration of soil was evaluated across different sites at depths of 10 cm and 30 cm throughout the study (Figure 3-7). At baseline measurements, the 10 cm depth had soil organic C concentrations (g C kg⁻¹) ranging from 4.6% to 11%, with an average of $7.6 \pm 0.7\%$, see Table 3-4. At the 30 cm depth, soil organic C concentrations ranged from 5.4% to 11%, with an average of $6.5 \pm 0.6\%$. Management practices had a highly significant effect on SOC ($\chi^2 = 16.6$, $p < 0.001$), while the sampling date and soil depth were also significant predictors ($\chi^2 = 155.4$, $p < 0.001$, and $\chi^2 = 233.3$, $p < 0.001$, respectively). Interactions between management and date ($\chi^2 = 38.4$, $p < 0.001$) and between date and depth ($\chi^2 = 48.4$, $p < 0.001$) were significant. The three-way interaction between management, date, and depth approached significance ($\chi^2 = 7.56$, $p = 0.056$), suggesting a potential complex interplay among these factors. By 2023, significant differences emerged, with Compost + Cover Crops showing higher SOC than Conventional management at both 10 cm (estimate = 0.28, $p < 0.0001$) and 30 cm (estimate = 0.12, $p = 0.001$). In 2024, these differences persisted, with significant increases in SOC under Compost + Cover Crops at both 10 cm (estimate = 0.59, $p < 0.0001$) and 30 cm (estimate = 0.24, $p = 0.03$). By 2024, the average percent increase in soil organic C concentration at 10 cm depth was $31 \pm 12\%$ under conventional management. In contrast, the compost + cover crops treatment resulted in a much higher average increase of $149 \pm 60\%$. At the 30 cm depth, conventional management showed a minimal increase of $1 \pm 9\%$, while Compost + Cover Crops resulted in an increase of $63 \pm 49\%$.

Table 3-4. Total C (C Conc.) and N (N Conc.) concentrations (g kg^{-1}), and soil organic C (SOC Cont.) content (Mg ha^{-1}) for the baseline, 2021, and the three years of the study until 2024. Plot represents management and is denoted by the colors used throughout the study, with conventional (C) as orange, and compost + cover crops (T) as green. Depth is measured in cm, and alley and berm represent the location of the soil sample.

Site	Plot	Indice	Depth	2021		2022		2023		2024	
				Alley	Berm	Alley	Berm	Alley	Berm	Alley	Berm
Almond 1	C	N Conc.	10	1.5 ± 0.09	1.4 ± 0.06	1.1 ± 0.1	1 ± 0.05	1.1 ± 0.03	1.1 ± 0.09	1.2 ± 0.05	1 ± 0.09
			30	1.2 ± 0.11	1.1 ± 0.04	0.9 ± 0.07	1.1 ± 0.04	1 ± 0.04	1.1 ± 0.05	1.4 ± 0.42	0.9 ± 0.05
			10	1.6 ± 0.09	1.3 ± 0.1	1 ± 0.08	1 ± 0.07	1.8 ± 0.11	2.3 ± 0.31	1.9 ± 0.29	3.2 ± 0.34
			30	1.1 ± 0.11	1.1 ± 0.03	1.1 ± 0.12	1 ± 0.04	1.2 ± 0.09	1.3 ± 0.07	1.3 ± 0.17	1.4 ± 0.11
	C	C Conc.	10	12.2 ± 1.07	10.5 ± 0.82	9.7 ± 1	9.3 ± 0.52	9.5 ± 0.52	9.1 ± 0.88	12.1 ± 0.68	9.6 ± 1.09
			30	10.4 ± 1.05	8.9 ± 0.47	8.9 ± 1.19	10 ± 0.46	9.4 ± 0.93	10 ± 0.77	15.9 ± 4.99	8.6 ± 0.38
			10	12.2 ± 0.95	9.1 ± 0.78	8.3 ± 0.8	8.4 ± 0.49	17.7 ± 1.4	22.7 ± 4.12	17.3 ± 2.54	28.4 ± 2.82
			30	8.4 ± 0.85	8 ± 0.31	9.3 ± 1.05	8.2 ± 0.46	9.6 ± 0.71	11.4 ± 0.89	15.2 ± 4.65	14 ± 1.49
	C	SOC Cont.	10	15.9 ± 1.5	13.6 ± 1.1	12.5 ± 1.3	12 ± 0.7	15.7 ± 1.1	10.5 ± 0.5	15.7 ± 1.1	12.1 ± 1.7
			30	27.1 ± 1.9	24.6 ± 1.3	23.6 ± 3.5	26.6 ± 1.3	31.4 ± 1.8	27.4 ± 1.1	31.4 ± 1.8	37.5 ± 1.3
			10	15.9 ± 1.3	11.8 ± 1	11 ± 1.1	10.7 ± 0.6	22.1 ± 3.8	34.3 ± 2.7	22.5 ± 3.5	37.3 ± 3.7
			30	23.3 ± 2.9	23 ± 0.5	29.6 ± 4.8	21.4 ± 1.5	35.5 ± 3.7	31.6 ± 5	42.4 ± 12.4	36.3 ± 4.7
Almond 2	C	N Conc.	10	1.3 ± 0.07	1.2 ± 0.05	1.3 ± 0.04	1.3 ± 0.06	1.1 ± 0.03	1.1 ± 0.09	1.1 ± 0.06	1 ± 0.03
			30	1 ± 0.03	1.1 ± 0.05	1.1 ± 0.04	1.1 ± 0.05	0.9 ± 0.06	1 ± 0.02	1 ± 0.03	1 ± 0.11
			10	1.5 ± 0.04	1.3 ± 0.08	1 ± 0.06	1.1 ± 0.07	2.4 ± 0.33	1.9 ± 0.14	1.9 ± 0.29	3.2 ± 0.34
			30	1.1 ± 0.08	1.3 ± 0.07	0.8 ± 0.08	1 ± 0.05	1.7 ± 0.09	1.3 ± 0.09	1.3 ± 0.17	1.4 ± 0.11
	C	C Conc.	10	10.7 ± 0.58	9.6 ± 0.42	11 ± 0.51	12.8 ± 1.6	9.5 ± 0.52	9.1 ± 0.88	13.8 ± 1.24	11.9 ± 0.17
			30	8.3 ± 0.39	9.8 ± 0.74	9.2 ± 0.41	9.8 ± 0.44	8.2 ± 0.59	8.8 ± 0.3	10.3 ± 0.11	9.6 ± 1.09
			10	12.2 ± 0.49	10.8 ± 0.99	8.6 ± 0.59	9.3 ± 0.76	23.6 ± 3.96	19.3 ± 1.89	17.3 ± 2.54	28.4 ± 2.82
			30	9.3 ± 0.81	11.4 ± 0.69	7 ± 0.82	8.4 ± 0.68	15.6 ± 0.98	12.1 ± 0.97	11.9 ± 1.32	13.7 ± 1.16
	C	SOC Cont.	10	14 ± 0.7	11.9 ± 0.5	13.8 ± 0.7	15.9 ± 1.9	11.8 ± 0.6	10.5 ± 0.5	17.6 ± 1.7	15 ± 0.2
			30	26.9 ± 1.5	30.4 ± 2.7	28.2 ± 1.3	27.6 ± 1.8	25.1 ± 1.8	27.4 ± 1.1	35.7 ± 2.5	25.9 ± 1
			10	15.3 ± 0.6	13.6 ± 1.3	10.7 ± 0.6	11.4 ± 0.9	29.7 ± 4.5	23.9 ± 3.2	31.2 ± 6.3	38 ± 2.7
			30	28.9 ± 2.5	33.7 ± 2.4	20.1 ± 4.1	23.7 ± 3.2	37.7 ± 4	29.7 ± 5	34 ± 0.6	37.6 ± 3.6
Almond 3	C	N Conc.	10	0.8 ± 0.15	0.8 ± 0.13	1.4 ± 0.13	1.3 ± 0.09	1.4 ± 0.04	0.9 ± 0.01	1.5 ± 0.28	1.2 ± 0.08
			30	0.8 ± 0.09	0.8 ± 0.14	0.8 ± 0.05	0.9 ± 0.06	0.7 ± 0.02	0.6 ± 0.01	0.7 ± 0.1	0.6 ± 0.08
			10	1 ± 0.13	0.8 ± 0.25	1.5 ± 0.11	2.9 ± 0.29	2.2 ± 0.09	1.3 ± 0.06	1.6 ± 0.11	1.3 ± 0.02
			30	0.7 ± 0.11	0.6 ± 0.11	1 ± 0.05	1.2 ± 0.12	1 ± 0.1	1 ± 0.04	1 ± 0.14	1.5 ± 0.48
	C	C Conc.	10	8.1 ± 1.73	7.8 ± 1.57	10.7 ± 1.04	9.8 ± 0.87	15.5 ± 0.54	8.4 ± 0.1	11.4 ± 0.67	11.5 ± 1.31
			30	7.8 ± 0.72	8 ± 1.53	4.8 ± 0.39	6 ± 0.47	6.3 ± 0.14	5.7 ± 0.13	4.8 ± 0.12	5.9 ± 0.77
			10	8.4 ± 0.65	5.7 ± 1.37	9.7 ± 0.47	8.5 ± 0.37	21.8 ± 1.26	12 ± 0.86	18.9 ± 3.76	10.7 ± 0.3
			30	5.7 ± 1	4.9 ± 0.97	6.2 ± 0.34	7.5 ± 0.21	10 ± 1.12	8.9 ± 0.38	8.8 ± 1.16	9.6 ± 0.12
	C	SOC Cont.	10	10.4 ± 2.3	9.9 ± 2.1	13.7 ± 1.4	12.4 ± 1.1	20 ± 0.7	10.6 ± 0.1	17.8 ± 1.6	15.4 ± 0.2
			30	20.2 ± 1.9	21.1 ± 3.6	13.1 ± 1.1	14.9 ± 1.2	16.2 ± 0.7	14.5 ± 0.4	29.4 ± 2.3	23.4 ± 0.8
			10	10.8 ± 0.9	7.3 ± 1.8	12.3 ± 0.5	10.8 ± 0.5	27.3 ± 1.6	14.3 ± 0.5	31.8 ± 6.3	38.8 ± 2.7
			30	20.2 ± 2.5	14 ± 4	15.2 ± 0.8	18.9 ± 0.7	18.8 ± 3.8	20.2 ± 1.7	23.4 ± 1	34.1 ± 3
Almond 4	C	N Conc.	10	1 ± 0.07	1.2 ± 0.13	1.1 ± 0.1	0.9 ± 0.06	1.6 ± 0.05	1 ± 0.02	1.4 ± 0.3	1 ± 0.07
			30	0.6 ± 0.03	0.7 ± 0.03	0.6 ± 0.08	0.7 ± 0.05	1.2 ± 0.67	0.7 ± 0.03	0.6 ± 0.01	0.4 ± 0.18
			10	0.9 ± 0.09	0.9 ± 0.06	1.1 ± 0.09	0.9 ± 0.07	2 ± 0.14	1.2 ± 0.09	2.8 ± 0.33	1.6 ± 0.25
			30	0.6 ± 0.05	0.6 ± 0.04	0.7 ± 0.09	0.6 ± 0.05	0.8 ± 0.14	0.8 ± 0.07	0.8 ± 0.02	0.8 ± 0.05
	C	C Conc.	10	8.6 ± 0.74	9.8 ± 1.16	10.4 ± 0.95	8.2 ± 0.57	15.1 ± 0.66	10 ± 0.41	14.1 ± 3.47	9.4 ± 1.11
			30	5.1 ± 0.32	5.9 ± 0.29	5.9 ± 0.77	5.8 ± 0.27	7.5 ± 3.07	6.3 ± 0.32	5.4 ± 0.18	4 ± 1.98
			10	7.7 ± 0.95	7.7 ± 0.62	10.5 ± 0.93	7.9 ± 0.65	17.5 ± 1.37	11 ± 0.86	24.9 ± 2.84	15.5 ± 1.49
			30	5 ± 0.42	5.7 ± 0.31	5.5 ± 0.51	5.7 ± 0.43	7.6 ± 1.58	7.2 ± 0.5	7.1 ± 0.16	7.4 ± 0.41
	C	SOC Cont.	10	10.4 ± 0.9	11.9 ± 1.5	13.7 ± 1	10.2 ± 0.7	18.6 ± 0.9	12.1 ± 0.5	18.3 ± 4.8	11.3 ± 1.4
			30	18 ± 3.1	23.1 ± 2	19.2 ± 2.4	19.4 ± 1.5	27.1 ± 12.5	21.6 ± 1.5	24.5 ± 1.8	20.8 ± 0.6
			10	11.2 ± 0.2	9.2 ± 0.8	12.2 ± 1.2	9.3 ± 0.8	21.3 ± 1.9	13.9 ± 1.1	30.4 ± 3.6	19 ± 1.9
			30	12.7 ± 0.2	18.8 ± 1.6	15.1 ± 2	18.5 ± 1.4	23.2 ± 6.3	22.6 ± 1.8	21 ± 0.6	23.5 ± 1.7
Walnut	C	N Conc.	10	0.6 ± 0.05	0.6 ± 0.1	1.1 ± 0.07	0.6 ± 0.06	1 ± 0.14	1.3 ± 0.56	0.8 ± 0.1	0.5 ± 0.03
			30	0.5 ± 0.04	0.4 ± 0.04	0.4 ± 0.06	0.4 ± 0.02	0.5 ± 0.06	0.4 ± 0.01	0.3 ± 0	0.6 ± 0.12
			10	0.6 ± 0.03	0.4 ± 0.01	1.1 ± 0.07	0.6 ± 0.06	1.6 ± 0.15	0.5 ± 0.09	2 ± 0.12	1.4 ± 0.79
			30	1 ± 0.29	0.7 ± 0.12	0.8 ± 0.22	0.5 ± 0.08	0.7 ± 0.09	0.3 ± 0.02	0.8 ± 0.12	0.5 ± 0.22
	C	C Conc.	10	5 ± 0.56	4.9 ± 1.09	10.5 ± 1.09	4.8 ± 0.73	7.8 ± 0.92	15.9 ± 7.37	8.8 ± 1.33	9.2 ± 2.01
			30	4.2 ± 0.43	3.3 ± 0.37	4.3 ± 0.69	3.8 ± 0.22	5.3 ± 0.53	4.3 ± 0.17	7.3 ± 2.56	7.1 ± 1.3
			10	4.8 ± 0.45	3.1 ± 0.25	11 ± 1.05	4.8 ± 0.73	16.8 ± 1.56	5.4 ± 0.98	25.6 ± 4.43	14.5 ± 7.13
			30	9.9 ± 3.37	5.6 ± 1.3	7.5 ± 2.61	3.7 ± 0.67	6.8 ± 0.96	2.4 ± 0.3	7.2 ± 1.24	4.5 ± 1.42
	C	SOC Cont.	10	6.2 ± 0.6	5.4 ± 1.2	6.1 ± 0.5	5.3 ± 1.1	14.7 ± 2.2	19.7 ± 9.9	10.2 ± 1.5	10.7 ± 2.4
			30	13.5 ± 1.5	9.9 ± 0.9	12.3 ± 3.6	10.5 ± 1	8.7 ± 0.2	9.4 ± 2	21.3 ± 6.3	20.6 ± 3.4
			10	5.2 ± 0.6	7 ± 2.2	9.6 ± 2.9	14.6 ± 2	15 ± 0.4	18 ± 0.2	25.3 ± 3.9	26 ± 5.4
			30	10.7 ± 0.2	4.9 ± 1	12 ± 1.7	14 ± 4.1	16 ± 10	16.5 ± 5	17 ± 3	18.8 ± 3.7

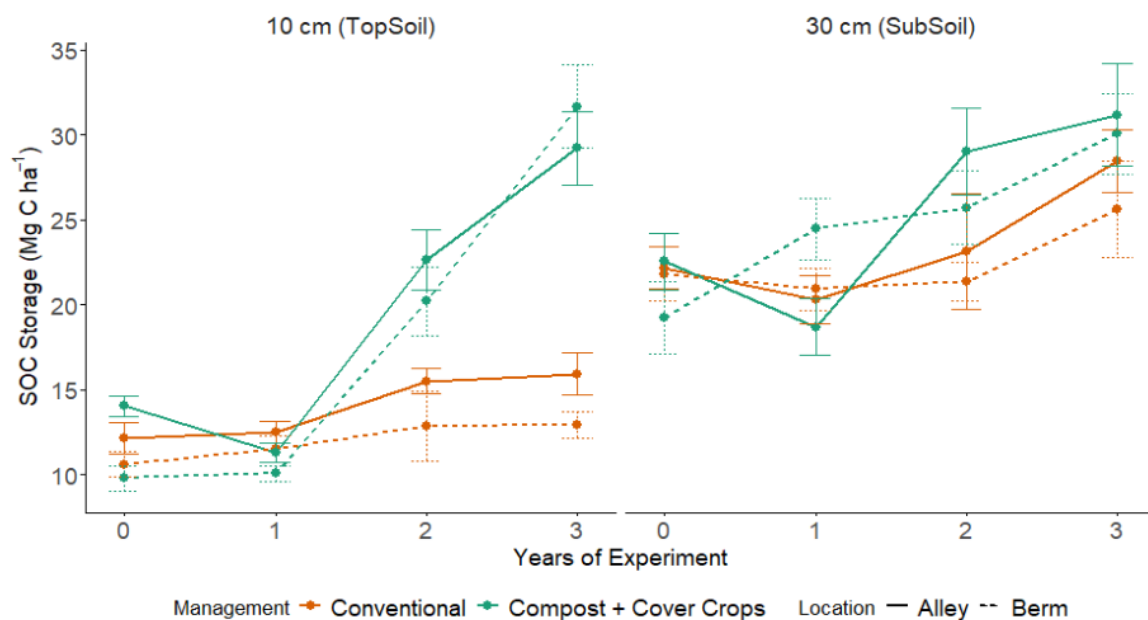


Figure 3-8. Soil organic carbon (SOC) storage by location of alley and berm and depth increments of 10 cm and 30 cm.

Soil organic C storage across sites varied across alley and berm, with lower values for the surface soils than the deeper depth (Figure 3-8). Berms were often lower, with an average of 10 ± 1 Mg C ha⁻¹ for baseline soils in the 10 cm depth, and 20 ± 2 Mg C ha⁻¹ in the 30 cm depth. Alleys were slightly higher than berms, with an average in the 10 cm depth of 13 ± 1 Mg C ha⁻¹, and an average of 22 ± 1 Mg C ha⁻¹. There was a significant effect of management ($\chi^2 = 37.1$, $p < 0.001$), year ($\chi^2 = 206.1$, $p < 0.001$), and depth ($\chi^2 = 371.6$, $p < 0.001$) on SOC storage, along with significant three-way interaction ($\chi^2 = 26.7$, $p < 0.001$). Pairwise comparisons (Comp. + Cover Crops – Conv.) with depth and management across years found that for the surface soils of 10 cm depth, there was a significant increase for year 2 (estimate:0.36, $p < 0.001$) and year 3 (estimate:0.76, $p < 0.001$). As for the deeper soils of 30cm, there was a significant increase for year 1 (estimate:0.13, $p = 0.04$) and year 2 (estimate:0.31, $p < 0.001$). There was a significant effect of compost and cover crops on the 10 cm depth soils across the alleys and berms for year 2 and year 3. As for the 30 cm depth, there was not a significant effect at the 30 cm depth, but there was a trend of increased soil organic C storage in both alley and berm compared to conventional management.

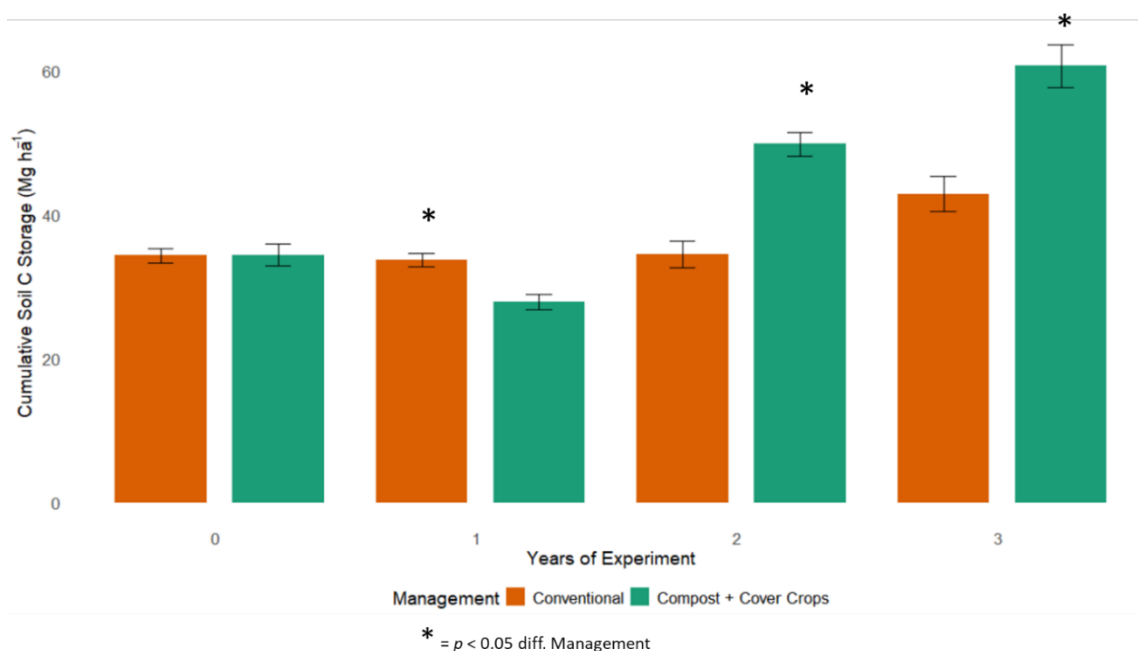


Figure 3-9. Cumulative soil C storage to the 0-30 cm depth, normalized to the ratio of alley and berms. Asterisks (*) indicate significant between management plots $p < 0.05$.

At the beginning of the study, baseline soils cumulative soil C storage to 30 cm ranged from a low of $18 \pm 1 \text{ Mg C ha}^{-1}$ to a high of $41 \pm 2 \text{ Mg C ha}^{-1}$, reflecting high variability across sites and generally soil C depletion. By the end of the study, after three years of applying compost and cover crops, the average percent increase in soil C storage was $102.2\% \pm 21.3\%$, more than double the $37.0\% \pm 9.7\%$ increase observed in the conventional plots (Figure 3-9. Cumulative soil C storage to the 0-30 cm depth, normalized to the ratio of alley and berms. Asterisks (*) indicate significant between management plots $p < 0.05$. Figure 3-9). Treatment plots resulted in an average soil C storage of $60.7 \text{ Mg C ha}^{-1}$, compared to $42.9 \text{ Mg C ha}^{-1}$ in the conventional plots in the cumulative soil profile to 30 cm depth. Over the course of the study, soil C storage was significantly impacted by management ($\chi^2 = 6.4, p = 0.01$), year ($\chi^2 = 187.4, p < 0.001$), and there was also a significant interaction between the two ($\chi^2 = 62.9, p < 0.001$).

Pairwise comparisons found significant differences between Compost + Cover Crops and conventional within after the first year of the study, with a significant decrease in the treatment plots (estimate: $-0.17, p = 0.001$). The trend shifted to positive increases in cumulative SOC storage for year 2 (estimate: $0.32, p < 0.0001$) and year 3 (estimate: $0.41, p < 0.0001$).

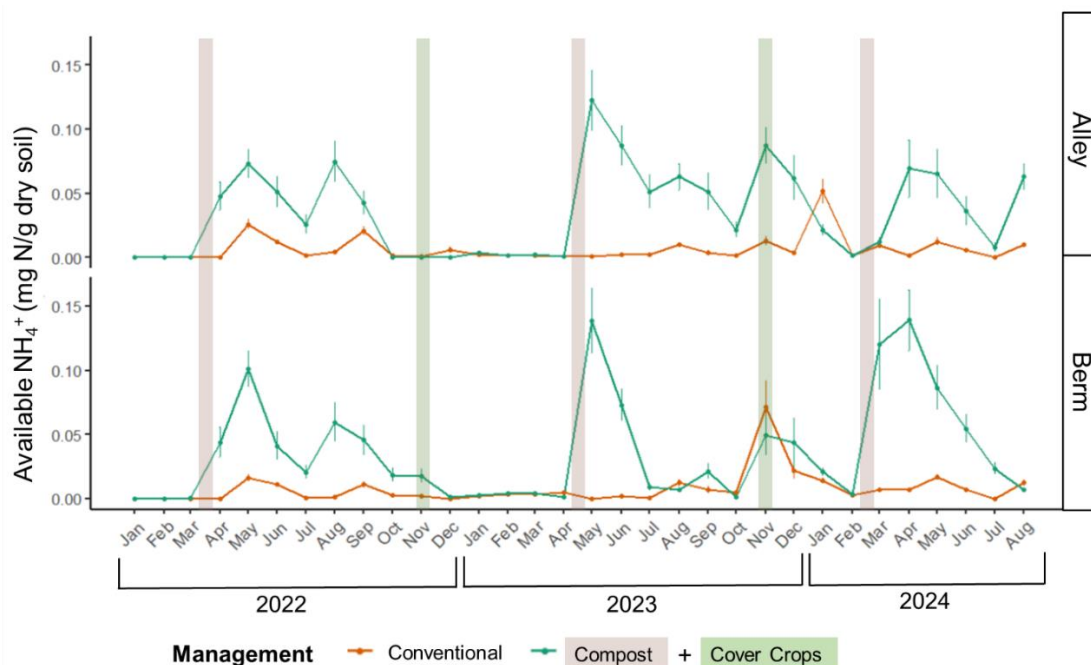


Figure 3-10. Average available ammonium (NH₄⁺) across sites for the months of the study after the application of compost and cover crops. Brown and green highlights represent the application of compost or the planting of cover crops throughout the study

Following the application of compost, the treatment plots showed a rapid increase in NH₄⁺ availability compared to the conventionally managed plots throughout all three years of the study (Figure 3-10). After the first application, the average increase in NH₄⁺ availability was 0.06 (\pm 0.02) mg nitrogen/g dry soil. By the second year, this increase more than doubled to 0.17 (\pm 0.05) mg nitrogen/g dry soil, and in the third year, it stabilized at 0.15 (\pm 0.05) mg nitrogen/g dry soil. A strong trend of increased NH₄⁺ availability was observed in both the berm and alley areas throughout the growing season when compost was applied in the spring. Notably, the alleys sustained this increase longer than the berms, with an average duration of 5 ± 2 months, compared to 3 ± 1 months in the berms. The analysis of deviance for NH₄ concentrations revealed significant effects of several predictors. Management practices had a highly significant influence on NH₄ levels ($X^2 = 284.9$, $p < 0.001$). Sampling dates ($X^2 = 1332.4$, $p < 0.001$) and their interaction with location ($X^2 = 388.21$, $p < 0.001$) were also significant, indicating temporal and spatial variability in NH₄ availability. The interaction between management and date was significant ($X^2 = 1047.43$, $p < 0.001$), suggesting that the effects of management practices varied over time. Additionally, the three-way interaction among management, date, and location was highly significant ($X^2 = 1817.75$, $p < 0.001$), highlighting the combined influence of these factors on NH₄ levels. Over the three years of the study, an increase NH₄⁺ availability was maintained for five months each year in the compost + cover crops plots.

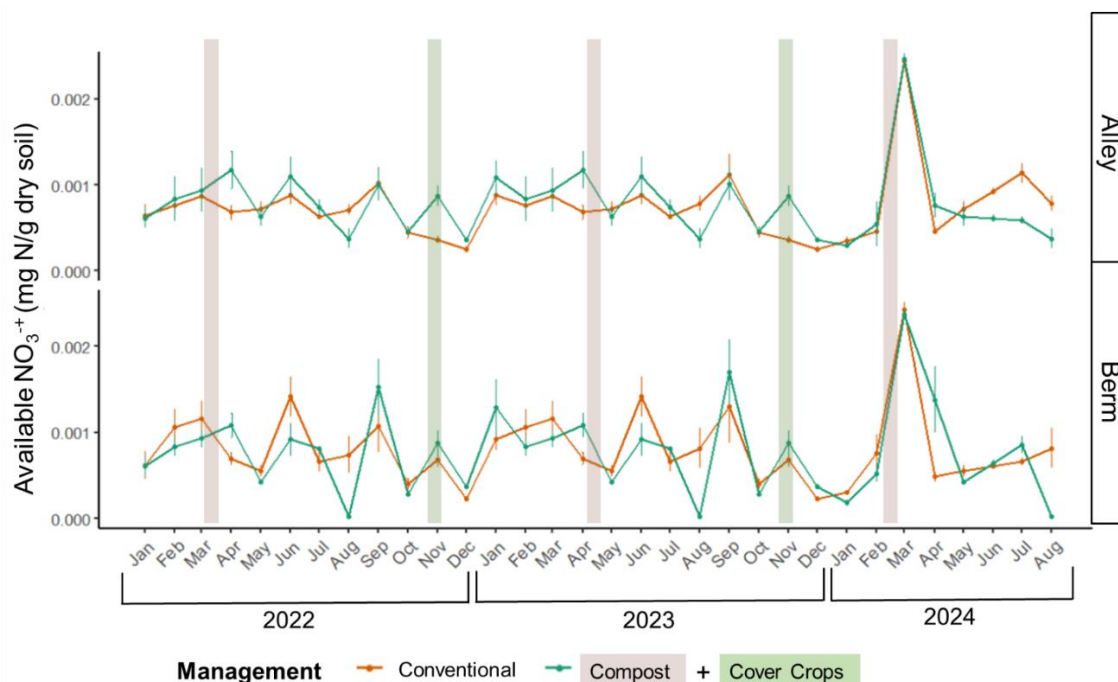


Figure 3-11. Average available nitrate (NO_3^-) content of the soils across sites for the three years after application of compost + cover crops. Brown and green highlights represent the application of compost or the planting of compost throughout the study.

Nitrate (NO_3^-) levels exhibit are highly variable with slight seasonal patterns, with peaks generally occurring in the spring and in the fall around September (Figure 3-11). The analysis of nitrate availability (NO_3) revealed significant effects of date ($\chi^2= 976.2, p < 0.001$), management by date ($\chi^2= 315.9, p < 0.001$), management by location ($\chi^2= 23.3, p < 0.001$), date by location ($\chi^2= 138.29, p < 0.001$), and the three-way interaction of management, date, and location ($\chi^2= 173.58, p < 0.001$). The main effects of management ($\chi^2= 1.95, p = 0.2$) and location ($\chi^2= 2.66, p = 0.1$) were not significant. Over the course of the study, compost + cover crops resulted in significantly higher nitrate levels compared to conventional management in six instances. These occurred primarily in the alley location, with one significant contrast in the berm location. The observed patterns are often results of fertigation events, where both plots increased in nitrate.

BIOLOGICAL

Table 3-5. Biological soil indicators of microbial biomass nitrogen (MBN) in mg N per dry soil, microbial biomass carbon (MBC) in mg C per dry soil, permanganate oxidizable carbon (POXC), and short-term C mineralization cumulative fluxes (CO_2) in $\mu\text{g C/g dry soil}$ for the management plots of conventional (C) and compost + cover crops (T) across the locations of alley and berm for the four years of the study.

Site	Plot	Indice	2021		2022		2023		2024	
			Alley	Berm	Alley	Berm	Alley	Berm	Alley	Berm
Almond 1	C	MBN	0.04 ± 0.01	0.01 ± 0.00	0.04 ± 0.01	0.01 ± 0.00	0.13 ± 0.03	0.01 ± 0.00	0.04 ± 0.00	0.01 ± 0.00
	T		0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.04 ± 0.00	0.01 ± 0.00	0.05 ± 0.00	0.02 ± 0.00
	C	MBC	0.04 ± 0.01	0.02 ± 0.00	0.04 ± 0.01	0.03 ± 0.00	0.13 ± 0.03	0.03 ± 0.00	0.04 ± 0.00	0.01 ± 0.01
	T		0.03 ± 0.01	0.03 ± 0.00	0.08 ± 0.01	0.04 ± 0.01	0.17 ± 0.02	0.04 ± 0.01	0.33 ± 0.02	0.10 ± 0.03
	C	POXC	179 ± 3	197 ± 12	199 ± 48	222 ± 2	309 ± 27	392 ± 22	367 ± 70	232 ± 7
	T		185 ± 48	209 ± 2	263 ± 5	344 ± 3	551 ± 14	551 ± 14	602 ± 57	593 ± 106
	C	CO ₂	5.1 ± 0.8	3 ± 0.43	3.8 ± 0.91	2.8 ± 0.62	2.4 ± 0.3	1.8 ± 0.3	1.8 ± 0.08	2.4 ± 0.13
T	6.7 ± 1.25		1.4 ± 0.14	14.3 ± 3.88	5.2 ± 0.81	7.4 ± 1.5	13.9 ± 6.71	6.7 ± 0.58	3.8 ± 0.08	
Almond 2	C	MBN	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.02 ± 0.00	0.00 ± 0.00	0.02 ± 0.01	0.01 ± 0.00
	T		0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.04 ± 0.00	0.01 ± 0.00	0.06 ± 0.00	0.02 ± 0.00
	C	MBC	0.06 ± 0.01	0.05 ± 0.02	0.06 ± 0.01	0.05 ± 0.02	0.13 ± 0.03	0.03 ± 0.00	0.03 ± 0.00	0.02 ± 0.01
	T		0.05 ± 0.01	0.05 ± 0.02	0.09 ± 0.00	0.04 ± 0.00	0.18 ± 0.03	0.05 ± 0.01	0.31 ± 0.03	0.07 ± 0.03
	C	POXC	155 ± 26	259 ± 43	199 ± 48	222 ± 2	303 ± 25	356 ± 30	309 ± 27	219 ± 7
	T		176 ± 8	225 ± 22	263 ± 5	344 ± 3	575 ± 16	271 ± 29	555 ± 4	437 ± 26
	C	CO ₂	5.1 ± 0.78	4.6 ± 1.56	3.8 ± 0.91	2.8 ± 0.62	2.5 ± 0.27	1.8 ± 0.3	3.5 ± 0	3 ± 0.06
T	8.1 ± 2.34		1.4 ± 0.16	14.3 ± 3.88	5.2 ± 0.81	7.4 ± 1.5	13.9 ± 6.71	10.6 ± 0.14	9.9 ± 0	
Almond 3	C	MBN	0.01 ± 0.00	0.02 ± 0.01	0.01 ± 0.00	0.04 ± 0.02	0.02 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00
	T		0.01 ± 0.00	0.03 ± 0.00	0.05 ± 0.02	0.05 ± 0.00	0.04 ± 0.00	0.03 ± 0.00	0.03 ± 0.01	0.02 ± 0.00
	C	MBC	0.03 ± 0.00	0.01 ± 0.00	0.03 ± 0.00	0.01 ± 0.00	0.09 ± 0.03	0.04 ± 0.01	0.12 ± 0.00	0.05 ± 0.00
	T		0.03 ± 0.00	0.01 ± 0.00	0.07 ± 0.02	0.03 ± 0.00	0.20 ± 0.03	0.21 ± 0.02	0.29 ± 0.02	0.18 ± 0.00
	C	POXC	269 ± 9	242 ± 5	413 ± 6	435 ± 21	495 ± 17	456 ± 2	394 ± 7	304 ± 5
	T		266 ± 6	236 ± 3	455 ± 34	434 ± 25	591 ± 15	436 ± 10	462 ± 90	398 ± 3
	C	CO ₂	3.8 ± 0.82	3.4 ± 0.93	2.6 ± 1.03	1.9 ± 0.52	3.6 ± 1.25	3.5 ± 1.61	4.6 ± 1.03	8.6 ± 0.35
T	3.6 ± 0.23		2 ± 0.28	2.6 ± 0.43	9.5 ± 8.33	6.1 ± 1.95	5.4 ± 3.02	4.5 ± 0.15	16.4 ± 0.59	
Almond 4	C	MBN								
	T									
	C	MBC								
	T									
	C	POXC	147 ± 6	214 ± 30	265 ± 17	129 ± 32	423 ± 7	266 ± 5	350 ± 35	362 ± 19
	T		156 ± 5	258 ± 9	307 ± 22	218 ± 23	528 ± 25	488 ± 4	451 ± 33	473 ± 19
	C	CO ₂	6 ± 0.51	4.5 ± 0.65	3.2 ± 0.57	10.4 ± 5.71	3.2 ± 0.81	15.7 ± 12.18	7.1 ± 0	5.8 ± 0
T	2.7 ± 0.19		3.4 ± 0.53	4.8 ± 0.16	4 ± 0.85	7.5 ± 1.55	5.4 ± 1.13	11.1 ± 1.1	7.5 ± 0	
Walnut	C	MBN	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.02 ± 0.00	0.03 ± 0.01	0.04 ± 0.03	0.02 ± 0.01
	T		0.01 ± 0.00	0.01 ± 0.00	0.02 ± 0.00	0.02 ± 0.01	0.08 ± 0.00	0.02 ± 0.00	0.11 ± 0.01	0.02 ± 0.00
	C	MBC	0.04 ± 0.00	0.01 ± 0.00	0.05 ± 0.00	0.02 ± 0.00	0.08 ± 0.01	0.10 ± 0.01	0.08 ± 0.04	0.07 ± 0.01
	T		0.04 ± 0.00	0.02 ± 0.00	0.06 ± 0.01	0.02 ± 0.00	0.30 ± 0.01	0.12 ± 0.00	0.09 ± 0.00	0.04 ± 0.00
	C	POXC	388 ± 2	153 ± 4	403 ± 0	167 ± 7	365 ± 19	306 ± 3	372 ± 29	280 ± 75
	T		403 ± 0	159 ± 12	531 ± 26	400 ± 6	695 ± 4	340 ± 27	564 ± 12	409 ± 3
	C	CO ₂	2.4 ± 0.31	2.7 ± 1.69	1.2 ± 0.27	0.9 ± 0.49	2.8 ± 0.26	2.4 ± 0.83	6.4 ± 1.04	17.4 ± 4.76
T	5.3 ± 4.2		1.2 ± 0.47	4.2 ± 1.67	9 ± 6.33	5.9 ± 4.54	2.5 ± 1.3	9.3 ± 1.4	6.7 ± 1.06	

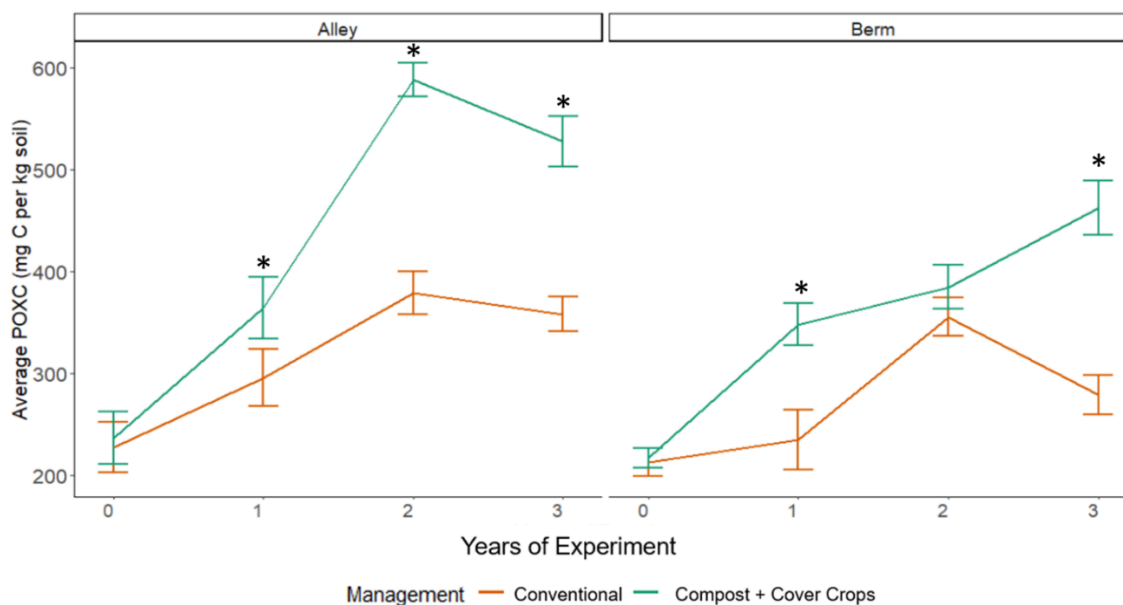


Figure 3-12. Average POXC across sites, with standard error bars of the mean. The orange line represents conventional, while the green represents the treatment plots of compost+ cover crops. Asterisks indicate $p < 0.05$.

PERMANGANATE OXIDIZABLE CARBON

As an indicator for reactive C, the POXC levels for most sites were similar at the beginning of the study with a mean of 223.8 ± 9.7 mg /kg soil (Figure 3-12). Management had a significant effect across sites ($p < 0.05$) on the amount of POXC, with the compost + cover crops plots increasing on average by 81% by year three. Throughout the study, both management plots demonstrated an increase in POXC until year three. By year three there was a significant increase for both alleys and berms across sites between the management of compost + cover crops compared to conventional ($p < 0.05$). There was a significant difference between the conventional alley and compost + cover crops alleys for year 1, 2 and 3 ($p < 0.001$) with treatment plots having 164 ± 29 mg C per kg soil more than conventional plots in year 3 (Table 3-5). There was a significant difference between the conventional berms and compost + cover crops berms for year 1 and year 3 ($p < 0.001$) with treatment plots having 182 ± 32 mg C per kg soil more than conventional plots in year 3.

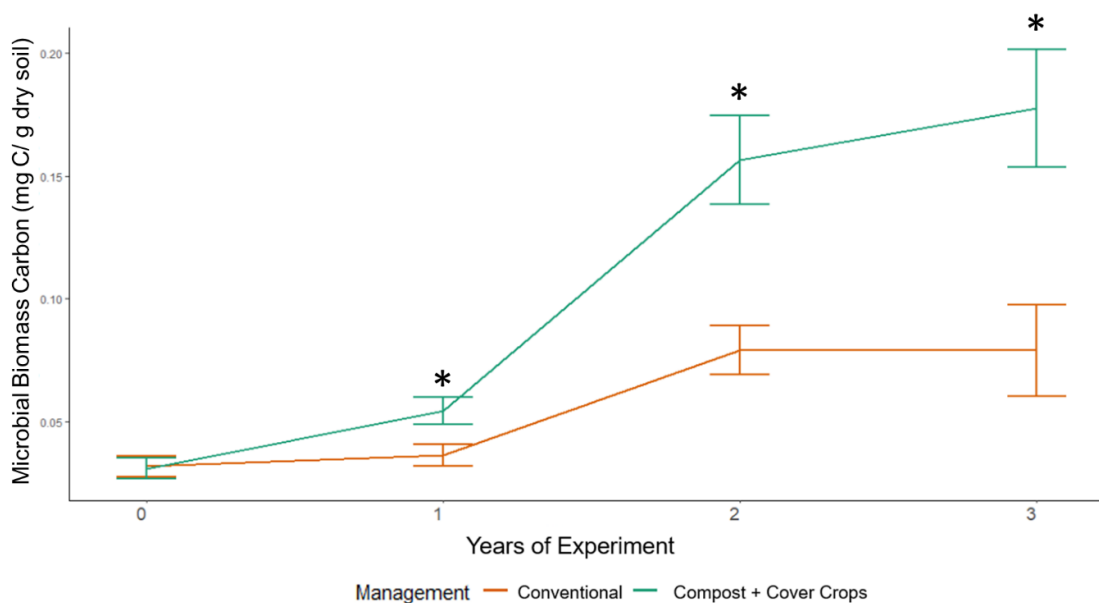


Figure 3-13 Microbial biomass carbon averaged across sites over the four years of the study, combining data from both alley and berm locations. Year 0 corresponds to 2021, and Year 3 corresponds to 2024. Asterisks indicate significance at $p < 0.05$.

The shift in MBC across all sites exhibited a consistent trend of increase in plots managed with compost + cover crops compared to conventional management (Figure 3-13 Microbial biomass carbon averaged across sites over the four years of the study, combining data from both alley and berm locations. Year 0 corresponds to 2021, and Year 3 corresponds to 2024. Asterisks indicate significance at $p < 0.05$). Management practices had a significant influence on MBC levels ($X^2 = 32.1$, $p < 0.0001$), as did location ($X^2 = 81.4$, $p < 0.0001$) and year ($X^2 = 169.5$, $p < 0.0001$). The interaction between management and year was also significant ($X^2 = 16.8$, $p < 0.05$), indicating that the effects of management practices on MBC varied over time. In year 1, MBC was significantly higher in compost + cover crops plots compared to conventional plots, with an estimated difference of 0.36 ± 0.17 ($p=0.03$). This difference increased in year 2, with an estimated difference of 0.69 ± 0.17 ($p<0.0001$), and year 3, with an estimated difference of 0.88 ± 0.17 ($p<0.0001$). Across all management practices, alleys consistently contained approximately twice the amount of MBC compared to berms, with significant differences between these locations ($p < 0.05$). By year three, MBC increased by $0.22 (\pm 0.03)$ mg C/g in alleys and $0.07 (\pm 0.03)$ mg C/g in berms under compost + cover crops management, resulting in percentage increases of $573 \pm 98\%$ for alleys and $307 \pm 100\%$ for berms. In contrast, conventional management practices resulted in increases of $0.08 (\pm 0.03)$ mg C/g in alleys and $0.01 (\pm 0.01)$ mg C/g in berms, with percentage increases of $200 \pm 59\%$ and $59 \pm 21\%$, respectively.

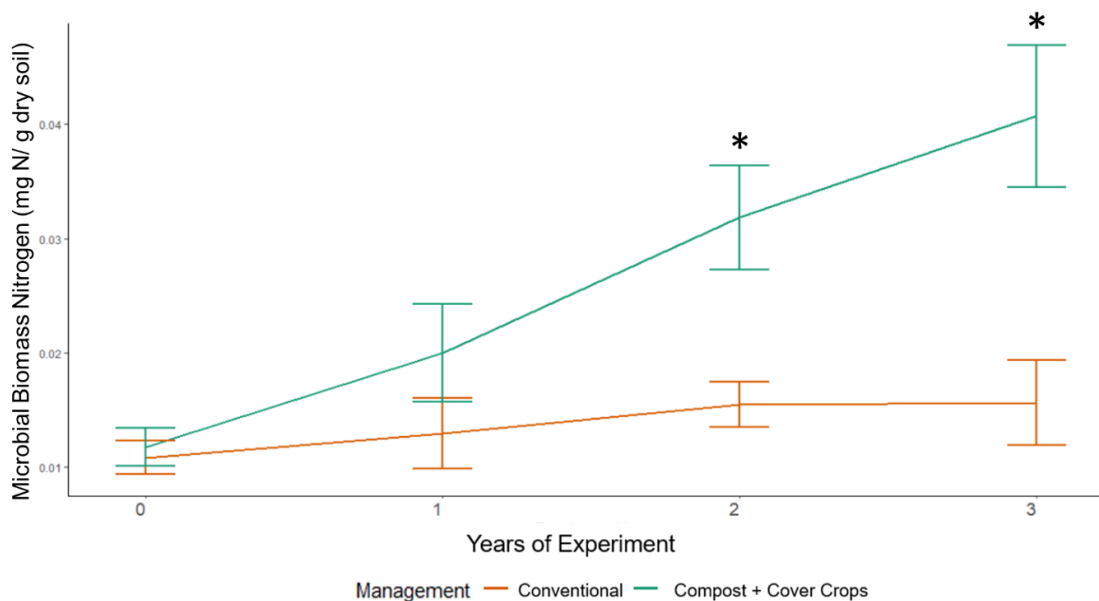


Figure 3-14. Microbial biomass nitrogen averaged across sites over the four years of the study, combining data from both alley and berm locations. Year 0 corresponds to 2021, and Year 3 corresponds to 2024. Asterisks indicate significance at $p < 0.05$.

MBN consistently increased in plots managed with compost + cover crops compared to conventionally managed plots across all sites (Figure 3-14). Management showed a significant effect ($X^2 = 24.9, p < 0.0001$), with compost + cover crops consistently improving MBN compared to conventional management. Location (Alley vs. Berm) also had a significant impact ($X^2 = 34.5, p < 0.0001$), MBN in the alleys was approximately seven times higher than in the berms. In addition, year was a significant effect ($X^2 = 39.8, p < 0.0001$), reflecting temporal variations in MBN levels. Interactions between management and year ($X^2 = 11.8, p = 0.008$) and location and year ($X^2 = 19.7, p < 0.001$) were also significant, indicating that the effects of management practices and spatial differences (alley vs. berm) varied over time. Across locations, MBN was significantly higher in compost + cover crops plots compared to conventional plots, with an estimated difference of 0.6 ± 0.2 ($p = 0.003$). This trend persisted and became more pronounced in Year 3, where compost + cover crops plots exhibited an estimated MBN difference of 0.9 ± 0.2 compared to conventional plots ($p < 0.0001$). These findings highlight the sustained and increasing benefits of compost + cover crops for enhancing MBN over time. By year three, compost + cover crops management led to increases of $487 \pm 95\%$ for alleys and $55 \pm 14\%$ for berms. In contrast, under conventional practices, only led to an increase of $113 \pm 37\%$ in alleys and a decrease of $-21 \pm 7\%$ in berms.

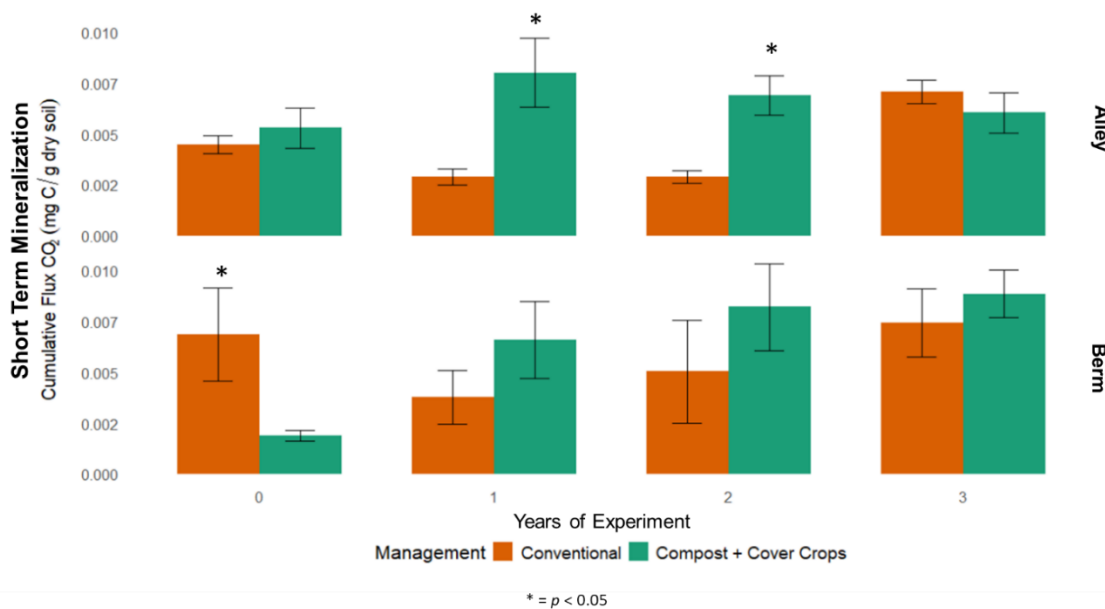


Figure 3-15. Cumulative CO₂ (mg C/dry soil) fluxes averaged across sites that were included in the microbial respiration incubation study conducted over four days. The data spans four years, with 2021 designated as Year 0 (baseline) and 2024 as Year 3 of the study. Cumulative fluxes were calculated based on the respiration measurements taken during the 4-day incubation period. An asterisk (*) indicates a significant difference between the management plots of conventional and compost + cover crops

Short-term C mineralization rates varied across locations of alley and berms through the years, with a compost + cover crops plots demonstrating significantly ($p = 0.04$) higher cumulative soil fluxes for most of the years. Across sites, after the baseline year (2021), the average increase in CO₂ emissions from the compost + cover crops plots was $118 \pm 0.002\%$ for 2022, $90 \pm 0.001\%$ for 2023, $3 \pm 0.001\%$ for 2024. There was a significant interaction effect between management and date ($p < 0.05$). Soils under conventional management exhibited lower microbial respiration rates compared to compost + cover crops management, for all years except 2024 (Table 5). After application, there was a significant increase in cumulative CO₂ emissions in both the alleys and berms, see Figure 3-15. There was not a significant difference between locations and emissions. By year three, there was not a significant difference between CO₂ emissions due to the change in management.

There was not an obvious trend of increased or decrease nitrous oxide (N₂O) and methane (CH₄) with the addition of compost + cover crops across the sites and throughout the study. Management plots did not demonstrate a significant increase in these gases with the addition of compost + cover crops. There was also no significant difference between the alley and berm, as the measurements were highly variable.

DISCUSSION

PHYSICAL INDICATORS

The physical properties of the baseline soils across sites were extremely compacted with higher than ideal bulk density for roots, especially in the alley, and exhibited low aggregation leading to potential erosion via water and wind. These characteristics, often resulting from management practices in conjunction with inherent soil type effects, contributed to the degraded soil quality observed at the start of the study. However, throughout the study period, physical indicators of soil health demonstrated consistent improvement, highlighting the effectiveness of the compost and cover crop management practices in enhancing soil structure and functionality. Compost application to agricultural lands is widely recognized for its effectiveness in enhancing the physical properties of soils, particularly those with poor structure and low organic matter levels (S. Brown & Cotton, 2011; Kranz et al., 2020). Additionally, cover crops have proven effective in reducing soil compaction, increasing aggregation and pore space, particularly when they include a diverse mix of plant species (Adetunji et al., 2020). Our study utilized a "soil builder mix" that included daikon radishes, which are known for their ability to penetrate and alleviate compacted soil layers.

Compacted soils with high bulk density can adversely affect crop health by reducing nutrient infiltration and limiting root growth. Compost has long been recognized for its ability to alleviate soil compaction (Aggelides & Londra, 2000; Curtis & Claassen, 2009), and so has the planting of cover crops (Haruna et al., 2020). By the end of our study, the bulk density of the soils at the 10 cm at the study sites decreased by 0.15 g/cm³. These results are consistent, but higher than, the findings from a study on citrus orchards, where the bulk density at the 0-10 cm depth decreased by 0.07 g/cm³ after two years of annual mulch and cover crop applications (Dung et al., 2022). The response of soil physical properties to compost and cover crops can vary depending on the soil's intrinsic texture characteristics (Siedt et al., 2021). For instance, Chen et al. (2021) observed notable improvements in soil quality from applying straw compost in semi-arid sandy soils. Conversely, Tian et al. (2015) reported a decline in soil quality with the use of composted organic manure in loamy soils.

In agricultural systems, the formation of larger and more stable soil aggregates plays a crucial role in enhancing soil aeration, water infiltration, and root penetration, thereby improving the availability of water and nutrients to plants (Annabi et al., 2007). Enhanced aggregate stability also mitigates soil erosion and compaction risks, preserving soil structure and function over the long term. The reduction in microaggregates and the concurrent increase in larger aggregates suggest that the soil in these plots is becoming more resistant to erosion and compaction. These findings are consistent with research by Scavo et al. (2022), who reported that the application of compost and the use of cover crops significantly enhance soil aggregate stability. These changes occurred relatively quickly, suggesting that added organic matter from both cover crops and compost can rapidly enhance wet aggregate stability. Soil aggregate stability is influenced by intrinsic properties like texture and mineralogy, alongside management-sensitive factors such as soil organic matter (Bissonnais, 1995; Mamedov et al., 2017). Soil clay content is closely

linked to aggregate stability, as clay particles serve as cementing agents that promote aggregate formation (Wagner et al., 2007). Notably, in our high-clay-content sites, we observed the most rapid and significant increases in aggregate size. Our results of improvements to aggregate stability align with other compost studies (Annabi et al., 2007; Whalen et al., 2003). However, some studies suggest that organic amendments may have limited effects on aggregate stability in highly compacted or degraded soils. McClelland et al. (2021) found that in certain soils, the benefits of compost on aggregate stability were not as pronounced unless mechanical interventions such as tillage were also applied. It suggests that while compost and cover crops can significantly improve soil structure, the degree of improvement may depend on the initial soil conditions. In some studies, it is suggested that the addition of organic amendments like compost can enhance the formation of larger aggregates, promoting the stabilization of particulate organic matter (POM) rather than mineral-associated organic matter (MAOM) (Ozlu & Arriaga, 2021; J. Shi et al., 2023; Six et al., 2002) This shift may indicate that the added carbon is contributing to the formation of stable aggregates through improved soil structure, rather than being primarily mineral-associated. Potentially, this stabilization pathway might lead to a labile or less persistent soil carbon pool that is more readily available for microbial decomposition and then release (Poeplau et al., 2021). We demonstrated that compost and cover crops can improve physical soil health indicators relatively quickly, which will have implications for improved soil functioning at the structural level, potentially improving water dynamics while decreasing erosion via wind and rain.

CHEMICAL INDICATORS

The addition of compost and cover crops led to improvements in the chemical indicators of soil health we monitored throughout the study: pH, EC, CEC, plant available nitrogen, soil C content, and storage. This change is likely driven by the direct increase in soil organic matter from compost and the indirect contribution from the decomposition of root biomass from the cover crops.

By enhancing soil organic matter, compost and cover crops buffer soil pH and enhance CEC by providing additional negatively charged sites that facilitate cation retention and exchange (Oyetunji et al., 2022). Additionally, compost helps to moderate electrical conductivity (EC) by supplying balanced nutrients, thus reducing the risk of salinity-induced stress and fostering a more favorable soil chemical environment (Scotti et al., 2016). There was a significant increase in EC across the treatment plots, which relates to the addition of cations and anions to the soil from the compost (Gondek et al., 2020). A significant increase in CEC was observed in the treatment plots, particularly in the alley regions. The significant increase in CEC observed in our orchard sites across alleys and berms was $37 \pm 6\%$, which is lower than a previous study on almonds in the Central Valley. Villa et al (2021) reported an average increase of $58 \pm 22\%$ in CEC across two soil types (loam and sand) following green waste compost amendments (Villa et al., 2021). This indicates an improved ability of the soil to retain and exchange nutrients, which is critical for maintaining soil fertility and plant health. The application of compost is known to enhance CEC by increasing the soil's ability to hold positively

charged ions (cations) such as potassium, calcium, and magnesium. The positive impact of compost on CEC has been well-documented in the literature. The study by Murphy (2015) emphasized that the increase in organic matter from compost leads to higher CEC, which in turn improves nutrient availability and reduces the risk of nutrient leaching. Additionally, Repullo-Ruibérriz de Torres et al. (2021) found that cover crops contribute to increasing CEC by adding organic matter through root turnover and biomass decomposition, further enhancing the soil's nutrient-holding capacity. The findings align with these studies and confirm that the use of compost and cover crops can significantly improve soil fertility through enhanced CEC. Some studies suggest that the impact of compost on CEC may be limited to certain soil types. Rakhsh et al. (2017) reported that in coarser soils with low clay content, the increase in CEC from organic amendments may not be as pronounced. This could explain some of the variability observed in our study, where certain sites exhibited smaller increases in CEC, particularly in the berm areas. This finding suggests that the effectiveness of compost in improving CEC may depend on soil texture and composition. The chemical properties of soil, including pH, EC, and CEC, are interrelated and collectively impact soil fertility and plant nutrient availability.

Compost quickly provides ample amounts of available nitrogen in the form of NH_4^+ , a finding that is well reported across agricultural systems (Goldan et al., 2023; Jain & Kalamdhad, 2020). The application of compost in our study led to rapid increases in NH_4^+ levels in the soil, indicating that compost quickly provides plant-available nitrogen. This is a critical finding for orchard systems where nitrogen is often a limiting nutrient for optimal levels of production. However, NO_3^- levels did not show consistent increases across all sites, suggesting that nitrate is more prone to leaching, particularly during periods of high NH_4^+ levels is consistent with findings from Sullivan et al., (1998) who reported that compost supplies nitrogen in a slow-release form as NH_4^+ , reducing the risk of nitrogen loss and providing a more sustainable source of nutrients. In addition, the use of cover crops has been shown to reduce NO_3^- leaching by increasing water retention, minimizing soil erosion, and enhancing nitrogen cycling (Ordóñez-Fernández et al., 2018). Our study aligns with these results, demonstrating the effectiveness of compost and cover crops in managing nitrogen availability in orchard systems.

However, Wang et al. (2024) noted that compost application can sometimes result in increased NO_3^- levels, especially in wetter climates or under conditions of excessive irrigation. This was observed at one site, Almond 3, where compost was applied in late fall. We documented a significant increase in NO_3^- , likely due to the wetter soil environment along with decrease nutrient demand from trees preparing for dormancy. However, for most of our sites, this contrasts with our findings where NO_3^- levels remained relatively responsive to fertigation events. Some research suggests that compost may not always provide sufficient nitrogen for high-demand crops like almonds, which often require additional inputs of synthetic fertilizers. Bijay-Singh & Craswell (2021) argue that compost alone may not meet the nitrogen needs of certain crops, particularly

during peak growth periods. This highlights the potential need for supplemental nitrogen inputs, especially in high-yield systems like almond and walnut orchards.

The baseline levels of soil C storage were very depleted but the implementation of compost and cover crops resulted in a significant increase in C storage at three of five sites, highlighting the effectiveness of these stacked practices in enhancing soil C levels. At the onset of our study, the average cumulative (30 cm depth) across sites and plots soil C storage was $32 \pm 3 \text{ Mg C ha}^{-1}$, by the end of the study we observed a $91 \pm 10\%$ increase, resulting in an average cumulative soil C storage of $61 \pm 3 \text{ Mg C ha}^{-1}$. The soil C concentration of the study had some interesting but expected trends, with the 10 cm depth demonstrating the most change in soil C concentration compared to the 30 cm depth, however both did increase significantly by year three. These findings are in line with the results of other studies that highlight the effectiveness of compost in boosting SOC, in both the surface and subsoils (Kätterer et al., 2014). Similarly, our results echo findings from orchard systems, where organic amendments like compost significantly increased SOC stocks over time, promoting soil resilience and carbon sequestration (Lepsch et al., 2019; Nichols et al., 2024).

Based on the observed improvements in wet aggregate stability, it is likely that the carbon (C) added through compost played a key role in the formation of particulate organic matter (POM). While POM is typically more labile and readily decomposed, its persistence in soil is governed not merely by its molecular structure but by a combination of factors, including its physical protection within aggregates (Six et al., 2004), chemical associations with soil minerals (Lehmann & Kleber, 2015), and accessibility to microbial decomposers (Schmidt et al., 2011). These mechanisms, alongside environmental and biological controls, collectively determine the long-term stabilization and sequestration of organic matter in soil systems. The persistence of the POM pool will largely depend on microbial activity inhibition, the degree of microbial limitations and carbon use efficiency, and microbial access constraints related to potential occlusion within fine aggregates within larger aggregates (Cotrufo et al., 2019; Cotrufo & Lavelle, 2022). It is also important to note that while our study indicates an increase in SOC, it is difficult to determine whether these changes were entirely the result of carbon inputs of compost. There is likely an increase in root exudation as well, which is a key process in forming or modifying MOAM by attaching organic compounds to the minerals (Keiluweit et al., 2015; Li et al., 2021; Poepflau et al., 2021). Similarly, other research reported that cover crops also contribute to SOC by adding organic matter through root exudates and biomass decomposition, further enhancing soil C storage (Castellano-Hinojosa et al., 2023; Ma et al., 2024). Moreover, the combination of compost and cover crops (a "stacked" practice) has been shown to have synergistic benefits for improving SOC storage. While the permanence and stability of these gains are still not fully understood, this study supports the hypothesis that using both practices together results in greater increases in SOC than using either practice alone.

While our results demonstrate a clear increase in SOC, some studies suggest that chemical fertilizers can also contribute to SOC improvements, particularly in the short term. Khalsa et al. (2020) showed that intensive nitrogen fertilization can increase SOC

by promoting faster plant growth and biomass production. We noted in our conventionally managed plots we do see an increase in SOC, just not to the magnitude of increase we saw with our compost + cover crops plots. This highlights the need for a balanced approach that incorporates organic amendments to conventional fertilization methods to sustain SOC levels over time. More research is needed to understand if this approach increased soil C storage via increased sequestration or via the direct exogenous inputs of C via the compost and cover crops, and whether the increase in soil C will be retained and protected from microbial decomposition.

BIOLOGICAL INDICATORS

We assessed biological soil health through measures of microbial biomass carbon and nitrogen, active carbon (POXC), and short-term C mineralization all followed a similar trend that by year 3, there was a significant difference between the plots that received compost and cover crops compared to conventionally managed.

Compost and cover crops improve microbial activity by providing a steady supply of organic matter, which serves as an energy source for soil microorganisms (Gougoulas et al., 2014). One such food source is permanganate oxidizable carbon (POXC), which represents a portion of soil organic matter that is easily oxidized and potentially biologically active (Duval et al., 2018). POXC is considered a key indicator of soil health because it responds quickly to changes in soil and crop management. As demonstrated in our study, although these changes unfold more slowly, they do materialize, typically showing significant improvements after about two years. This delay is influenced by several factors: the time required for microbes to decompose and utilize new organic matter, the need for soil conditions like moisture and pH to stabilize, and the gradual release of nutrients that microbes need to boost their activity (Wang et al., 2022).

However, it is important to note that some research challenges the reliability of certain carbon availability indicators, such as POXC. According to Margenot et al. (2024), POXC may not accurately measure labile carbon as previously thought but rather oxidizes polyphenols, which can skew results. The changes we saw could reflect polyphenols such as lignin, as permanganate has very high oxidative affinity for phenols (Woodings & Margenot, 2023). Despite this, higher rates of available C can lead to increases in microbial growth and activity (Yang et al., 2024). The overall trends in microbial biomass and activity measured in our study are consistent with findings from other research in orchard systems (Baldi et al., 2018; Bechara et al., 2018; Yao et al., 2005).

We found a significant increase in microbial biomass carbon and microbial biomass nitrogen in treatment plots receiving compost and cover crops by the second and third years. These increases reflect enhanced microbial activity and nutrient cycling in the soil, which are crucial for improving overall soil health (Haruna et al., 2020a; Nicolardot et al., 1994). Significant differences were also observed between the alley and berm areas, with alley regions consistently exhibiting higher microbial biomass. As both areas received compost, but only the alleys were planted with cover crops, it could be indicative that microbial biomass carbon and nitrogen were more readily responding to

the multi-species cover crops inputs of root exudates and root biomass once terminated. A finding supported by Repullo-Ruiberriz de Torres et al., who found that microbial biomass was higher in alley with cover crops compared to soils beneath trees in pecan orchards. (2021). The increases in microbial biomass carbon and nitrogen in treatment plots align with other studies that have demonstrated the beneficial effects of organic amendments, such as compost, on microbial biomass (Bertrand, 2019; Tian et al., 2015). While our results show a clear increase in microbial biomass in treatment plots, some studies suggest that microbial biomass may not always respond as significantly to compost and cover crop treatments under all conditions. For example, Vukicevich et al. (2019) found that microbial biomass can be influenced by soil texture and cover crop species. In cases where soils are compacted or have low organic matter content, microbial biomass may not increase as expected, highlighting the importance of site-specific factors in determining the effectiveness of soil amendments.

The higher rates of short-term C mineralization observed indicate an increase in microbial respiration of CO₂, is a key indicator of soil microbial activity (Koritschoner et al., 2022). The compost and cover crops provide additional organic matter that microbes decompose, releasing CO₂ in the process. This metric combined with POXC are related are measures of active organic matter that may provide early indication of soil C stabilization and mineralization processes. With POXC better reflecting SOM stabilization while short term C mineralization reflects SOM mineralization. In our study, we observed that after the first year, SOC storage across sites decreased, which was initially reflected in an increase in mineralizable C and POXC, indicating an increase in microbial-accessible carbon. This suggests that early on, the addition of compost and cover crops likely enhanced the availability of more labile carbon forms, which stimulated microbial growth and activity (Gentsch et al., 2024; Rath et al., 2022). However, by year three, we noticed a decrease in the trend, with no significant increase in respiration, despite a continued increase in POXC. This transition may indicate that the added compost and cover crops are promoting the formation of organic matter that is less readily available to microbes, potentially contributing to long-term carbon sequestration. The biological component of soil health is a crucial component to measure when assessing changes in management influence on soil health, as these indicators are the underpinning processes to understanding nutrient availability, carbon storage, and the functioning of the soils as a true living system full of microbial activity.

MANAGEMENT IMPLICATIONS

The stacked practices of compost and cover crops in our study demonstrated there were benefits to soil health's chemical, biological, and physical properties (Illustration: 3-1). Compost has demonstrated that it does quickly provide available nitrogen in the form of NH_4^+ . The release of nitrogen from compost depends on factors influencing the rate of mineralization of organic nitrogen to plant-available forms of inorganic nitrogen. These

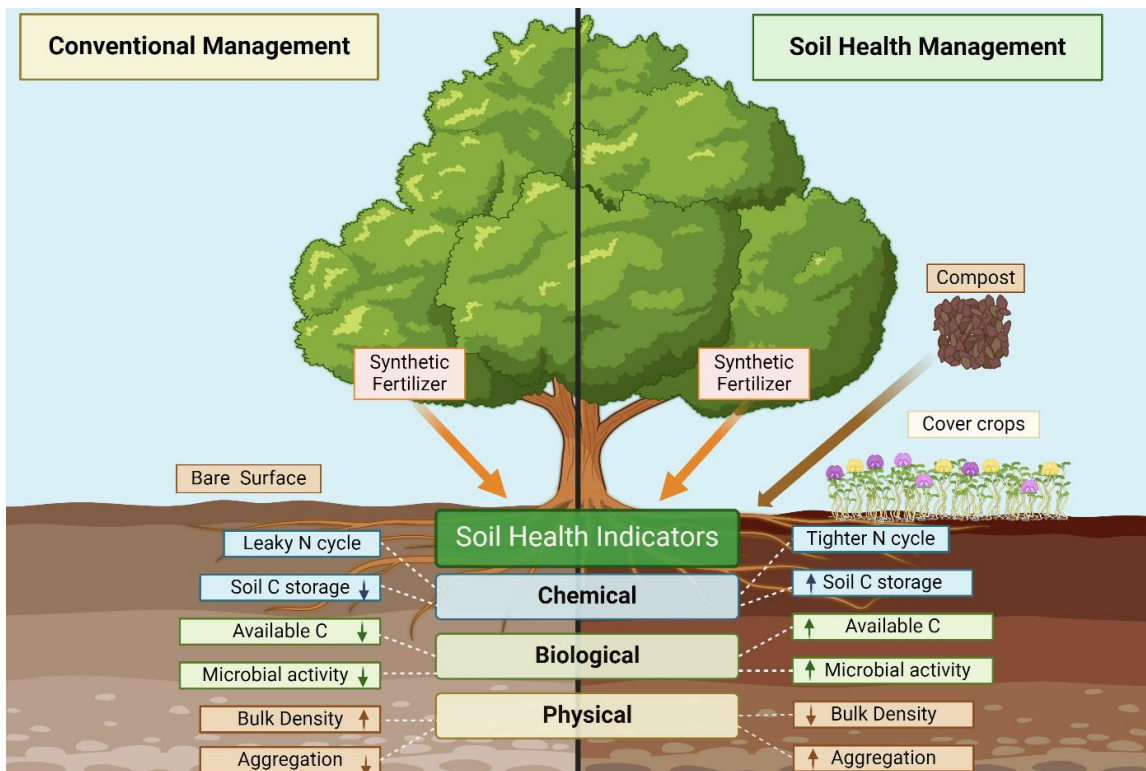


Illustration: 3-1 Comparison of conventional management and soil health management on the soil health indicators organized into chemical, biological, and physical indices that were measured within our study.

factors include soil and management practices such as irrigation, microbial activity, and overall edaphic soil conditions. Whether the amount of inorganic nitrogen provided from compost is sufficient to support almond production without chemical fertilizer isn't quantified in this study. Some studies have already demonstrated that compost can provide the nutrients necessary for farmers to shift wholly over to organic amendments (Hernández et al., 2016). The aim of our study was not to provide data that these practices should be used instead of conventional management, but rather if they have synergistic beneficial effects to current management practices. The addition of compost and implementation of cover crops can help to improve the soil conditions that promote a tighter nitrogen cycle (Rath et al., 2022; Repullo-Ruibérriz de Torres et al., 2021). A more efficient nitrogen cycle relies on the improvement of the soil health indices that we monitored throughout our study, encompassing the biological, chemical, and physical components. Integration of cover cropping into the orchard's current operational system requires careful management to balance benefits with potential competition for water and nutrients, which relies heavily

on timing of planting, and termination. In our system, winter cover crops were the best choice, as they do not require additional water and rely on winter rain events. The timing of termination can mitigate these risks of nutrient competition and ensure optimal performance of both cover crops and nut tree production, our management recommendation was to terminate in March after bees were removed, and before the spreading of compost. Termination methods varied across the study, with most terminating with mowing and some with light tillage. Some research suggests that mowing or roller crimping is a more sound soil health approach as it has less soil disturbance, leaves biomass as mulch on the surface, and promotes slower root decay within the soil (Adetunji et al., 2020). Another challenge to the implementation of both compost and cover crops in almond orchards is that harvesting practices currently use the ground to dry out hulls and to harvest. Most of the orchardists in our study did not report having any issues with the timing or shift of alley floor management to adjust to the use of compost and cover crops. While there are changes in management to integrate these practices, it was not prohibitive.

From an economic standpoint, cover crops along with compost are front-loaded costs, which might take time to see the benefits. Our study demonstrated that the effect was not immediate but became significantly improved by year 3. In states like California, there are programs which can help to cover the implementation costs of these practices, such as the Soil Health Program and connecting with local Resource Conservation Districts to apply for grants. It is difficult to put a price on the functions of a healthy soil system that is more resilient to both abiotic stressors, such as drought, and biotic stressors, such as pests. This is especially crucial as the severity of these stressors is expected to rise with climate change. Healthier soils can act as a buffer, mitigating the impacts of these stressors while potentially increasing both the productivity and longevity of agricultural systems.

CONCLUSION

The combination of compost and cover crops in nut orchards is a powerful management strategy for enhancing soil health, fertility, and nitrogen cycling, which contributes to the transition towards more regenerative nut orchard production. While compost alone may not fully replace chemical fertilizers in nut production, our study demonstrates that integrating compost and cover crops into conventional orchard management offers benefits beyond nitrogen supplementation. These practices positively influence multiple biological, chemical, and physical soil health indicators, including microbial activity, aggregate stability, and carbon storage. All of which demonstrate the power of these practices to restart the ecological soil system services that support nutrient cycling and improved plant/soil interactions. These improvements were observed independently of site factors such as soil type, past management, and irrigation, highlighting the broader applicability of this integrated approach for promoting soil health in diverse orchard systems. This approach not only promotes agroecological resilience to climate change but also supports climate mitigation efforts through the potential of increased soil carbon sequestration.

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DECLARATION OF GENERATIVE AI AND AI-ASSISTED TECHNOLOGIES IN THE WRITING PROCESS

During the final editing of this work the authors used ChatGPT by OpenAI to increase readability via syntax and fix grammatical errors. After using this tool, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

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CHAPTER 4: SOIL MICROBIAL COMMUNITY DIVERSITY IN ORCHARD VARIES BY DEPTH AND MANAGEMENT

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ABSTRACT:

Soil biodiversity is a critical component of soil health, playing a pivotal role in organic carbon turnover, nutrient cycling, and disease suppression. We investigated the effects of depth and management practices (compost + cover crops versus conventional) on soil microbial communities within almond orchards in the San Joaquin Valley of California. Soil samples were taken at four different depth intervals (0-10 cm, 10-30 cm, 30-50 cm, and 50-100 cm) from three orchards with paired management practices (conventional versus cover crops and compost) that were in place for three years. Soil microbial diversity was assessed using 16S and ITS sequencing. We found that bacterial and fungal communities were significantly affected by site, depth, and management practices ($p < 0.05$). Compost and cover crops increased the abundance of beneficial bacterial phyla such as *Actinobacteria*, *Acidobacteria*, and *Chloroflexi*. For fungi, these management practices significantly enhanced the abundance of *Ascomycota*, *Basidiomycota*, and *Mortierellomycota*, phyla consisting of microbes which are involved in organic matter decomposition and nutrient cycling. The depth-dependent variations in microbial communities emphasized the importance of managing both surface and subsurface soils to optimize soil health and microbial-plant interactions. Highlighting that the integration of compost and cover crops fosters a more functionally diverse soil ecosystem crucial for resilient and sustainable orchard management.

INTRODUCTION

Soil biodiversity is a crucial component of soil health in agroecosystems and plays a key role in regulating the turnover of organic carbon, nutrient cycling, and disease suppression (Crowther et al., 2019; Delgado-Baquerizo et al., 2020). Soil health management can have a large influence on soil biodiversity, primarily through the input of organic matter (Robinson et al., 2024; Thiele-Bruhn et al., 2012). Perennial cropping systems like almond orchards are an ideal system to focus sustainability shifts, due to their highly managed status and long growing cycle (~25 years). Almond (*Prunus dulcis*) orchards are one of the most extensive agricultural land use types in California and shifts

to ecological-based management could improve the sustainability and resilience of these agroecosystems (Fenster et al., 2021). Conventional management creates distinct microbial assemblages compared to soil health management (Hartmann et al., 2015). Soil health management focusing on organic matter improvements usually increases bacterial and fungal diversity metrics (Herrmann et al., 2023; K. L. Tully & McAskill, 2020). Management practices, such as the incorporation of compost and cover crops, have been shown to enhance various chemical, physical, and biological properties of soil (Cooper et al., *In Review*; Haruna et al., 2020; Ingels et al., 2005; Koudahe et al., 2022; Lazcano et al., 2022). The effects of soil health practices on the biological component of soil in almond orchards is not well understood and is essential for optimizing plant-soil interactions. In addition to the potential shift from management, there is also a need to understand how microbial communities change with depth in these almond orchards.

Almond orchards represent California's most lucrative crop, generating \$6.09 billion in revenue from 619,169 hectares (CDFA, 2019, 2020). They account for 80% of the global almond supply and nearly 100% of the domestic supply (California Almond Board, 2016). Conventional management practices in almond orchards prioritize chemical interventions over the maintenance of soil health, often leading to long-term ecological imbalances (Wade et al., 2019). Synthetic fungicides and pesticides used in conventional agriculture can cause the loss of non-target species that perform important biochemical processes, such as nitrogen fixation or toxic compound degradation (Beaumelle et al., 2023; Zhan & Zhang, 2014). Conventional soil management practices in orchards, such as maintaining bare alleys through scraping or herbicides often result in dry, compacted soils that are not conducive to microbial and fungal proliferation (Maestre et al., 2015). Almond orchards that are managed conventionally are often bacterially dominated (Camacho-Sanchez et al., 2023). An imbalanced soil system can lead to various issues with nutrient cycling, including reduced availability of essential nutrients like nitrogen and phosphorus, which are critical for plant growth (Adomako et al., 2022).

Soil depth significantly affects microbial communities, with variations in moisture, pH, organic matter, and root interactions influencing microbial abundance and diversity (Naylor et al., 2022). Generally, microbial diversity, for both bacterial and fungal communities, decreases with increasing soil depth, with surface soil having the greatest abundance and diversity (Goyal et al., 2019). However, deeper soil layers can still harbor diverse and active microbial populations, particularly in intensively managed agroecosystems where tillage, irrigation and fertilization influence soil conditions (Nsabimana et al., 2004). Studies have shown that both bacterial and fungal communities exhibit distinct vertical stratification (Fierer et al., 2003; Kluting et al., 2019). Surface soils (0-30 cm) typically contain higher diversity due to greater organic matter inputs and root density (Hao et al., 2020). In contrast, subsoils (30-100 cm) can support unique microbial communities adapted to lower nutrient levels and different moisture regimes. For instance, certain bacterial taxa, such as *Actinobacteria* and *Firmicutes*, are more prevalent in deeper soils due to their ability to degrade complex organic compounds and survive in nutrient-poor conditions (Byers et al., 2023). The relationship between soil depth and microbial communities has not been extensively studied in irrigated and fertilized orchard systems. Given the active root zone for almonds typically extends from

50 cm to 1 m, it is hypothesized that diverse microbial communities are present throughout these depths (Bintarti et al., 2020). Subsoil microbes play critical roles in nutrient cycling, and organic matter decomposition, and may even play a “safety-net” role in enhancing plant resilience to environmental stresses (Beule et al., 2022). Understanding the distribution of microbial communities across soil depths is crucial for improving soil health management practices in almond orchards and promoting sustainable productivity under increased climatic variability.

Shifts towards sustainable practices in almond orchard management are driven by the goal to improve soil health, which is crucial for maintaining orchard productivity. Commercial perennial agriculture, such as almond orchards, is susceptible to declining productivity due to negative plant-soil feedback (Bever, 1994). This occurs when plants foster soil microorganisms that are detrimental to their growth, a process further influenced by management practices such as monocropping. Soil health encompasses chemical, physical, and biological indices, with biological indices—particularly soil microbial abundance and diversity, playing a pivotal role in soil processes and nutrient cycling (Kv et al., 2019). By enhancing these biological aspects, sustainable practices aim to mitigate negative soil feedback and support long-term orchard productivity (Vukicevich et al., 2016). Sustainable practices such as organic amendments and cover cropping are key to enhancing microbial diversity (Nair & Ngouajio, 2012). Organic amendments, like compost, provide essential nutrients and organic matter that serve as substrates for soil microbes (Heisey et al., 2022). Cover cropping, the practice of growing specific crops during the off-season, significantly impacts microbial diversity (Vukicevich et al., 2016). Cover crops, including legumes, grasses, and brassicas, produce diverse root exudates that stimulate various microbial populations (Seitz et al., 2023). Mycorrhizal inoculants included with compost can establish beneficial arbuscular mycorrhizal fungi (AMF) in the soil, which form mutualistic associations with plant roots and enhance nutrient uptake, especially phosphorus (Begum et al., 2019). Nitrogen-fixing bacteria such as *Rhizobium* and *Frankia* are promoted through leguminous cover crops, increasing soil nitrogen availability (Zahran, 1999). Fostering microbial diversity is crucial for the long-term sustainability of almond orchards in the Central Valley (Özbolat et al., 2023).

OUR STUDY

The integration of compost and cover crops into soil management practices shows promise for supporting microbiomes, leading to a healthier soil ecosystem. This study investigates the effects of these practices on bacterial and fungal community dynamics within Central Valley almond orchards, focusing on how they influence indices of abundance and diversity, as well as the role of soil depth in shaping these communities. Understanding bacterial and fungal diversity at different soil depths is essential for developing targeted agricultural practices that can optimize almond production under the unique environmental conditions of the Central Valley. The findings of this study will offer valuable insights into sustainable soil management practices, potentially improving orchard productivity and agroecological functioning. By employing advanced molecular techniques and high-throughput sequencing, this study aims to provide a comprehensive

characterization of microbial communities in relation to soil characteristics, such as texture and depth, within Central Valley almond orchards. This research will contribute to a deeper understanding of soil microbial ecology and inform the development of sustainable agricultural practices for this region.

MATERIALS AND METHODS

STUDY SITES

The study was conducted in three commercial almond orchards located within the San Joaquin Valley of California (Table 4-1), with various soil types, orchard ages, and irrigation management. This region, historically the ancestral homeland of the Tejon, Kitanemuk, Yokuts, and Chamash indigenous peoples, is situated south of the Sacramento-San Joaquin River Delta and is drained by the San Joaquin River. The San Joaquin Valley experiences a Mediterranean climate characterized by cool, wet winters and hot, dry summers. Mean annual temperatures in this region are approximately 14-16°C, with mean high temperatures reaching around 36°C and mean low temperatures hovering around 12°C (NOAA, 2021). Mean annual precipitation ranges from 13 to 25 cm, primarily occurring between November and April, with typically no rainfall during the summer months.

Table 4-1. Site information for the orchards within our study in the Central Valley of California

Location	Site	Soil Order	Soil Series	Soil Texture	Tree Types	Orchard Age	Irrigation
Westley, CA	Almond 1	Inceptisol	<i>Zacharias</i>	Fine-loam	Nonpareil & Monterey	6	Drip
Madera, CA	Almond 3	Mollisols	<i>Visalia</i>	Fine Sandy Loam	Nonpareil & Monterey	12	Drip & Flood
Roberts Ferry, CA	Almond 4	Entisol	<i>Hanford</i>	Coarse-loam	Nonpareil & Monterey	15	Sprinkler

SOIL HEALTH PRACTICES

Cover crops were seeded in November of 2021, 2022, and 2023 at a rate of 13.8 kg/ha in the alleys, between almond tree rows. A seed drill (Brillion Landscape Seeder LSS-6, GE, US) was used for the planting of the cover crops across the sites, at a depth of ranging 0.65 -1.27 cm. The cover crop mix selected was soil building mix sourced from Project Apis m. This mix contains 30 % Triticale (x *Triticosecale*), 35% Bell Beans, (*Vicia faba*), 28% Peas (*Pisum sativum* or *P. arvense*), 1% Canola (*Brassica rapa*), 1% Common Yellow Mustard (*Sinapis alba*), and 5% Daikon Radish (*Raphanus sativus*). The mix contains brassicas, legumes, and grains to address soil issues such as compaction and erosion, while fixing N, and providing weed suppression. This mix also delivers a late source of nectar for pollinators. Weeds and cover crops were mowed within the months of March and April and again August prior to harvest. No additional water was used to aid in the germination of the cover crops.

An organic rich compost made from feedstocks of from manure and green waste (AllGro by Synagro) was applied as a surface dressing across berms and alleys at a rate of 19 Mg/ha in March of 2022 and May of 2023. Compost timing was dependent on weather and compost availability, which differed each year. The compost has an organic

C content of 27%, and a total organic N content of 3.3% (C:N ratio of 8:1) with a pH of 7.7, and a bulk density of 2.8 g/cm³. The amendment of compost added approximately 5.13 Mg C/ha, and 0.5 Mg N/ha to the soils in each application.

SAMPLING PROCEDURE

To assess the impact of compost and cover crop management on the bacterial and fungal distribution, diversity, and abundance, three almond orchards were selected. Soils were sampled across all sites in November 2023, after two years of compost and cover crop implementation. At each almond orchard, two plots were designated: one under conventional practices and the other implementing compost + cover crop management. Within each plot, three replicates (n=3) were sampled using a drill press soil auger (Geoprobe, MC5 Soil Sampling System (LWCR), KS, USA) to a depth of 1 meter. The soil cores were divided into four segments (0-10 cm, 10-30 cm, 30-50 cm, and 50-100 cm). Samples (n=74) were immediately placed on dry ice and stored at -20°C until DNA extraction using a DNeasy PowerSoil Pro kit (QIAGEN, MD, USA). DNA concentration was determined by measuring the absorbance at 260 nm with a spectrometer using a quartz cuvette.

16S AND ITS SEQUENCING LIBRARY PREPARATION

The rRNA 16S V4 region 515F and 806R primers (Parada et al., 2015 and Apprill et al., 2015) were modified to include the Illumina platform adaptor sequences (Illumina, 2013). Briefly, 0.1 uL uL⁻¹ of sample DNA was added to 1X of Kapa HIFI Hotstart Readymix (Roche, Indianapolis, IN, USA) and 200 nm. of the modified primers. The reactions were incubated for 3 minutes at 95 °C, followed by 25 cycles of 95 °C for 30 seconds, 55 °C for 30 seconds, and 72 °C for 30 seconds, and extended for 5 minutes at 72 °C.

Similarly, the ITS2 region 5.8S-Fun and ITS4-Fun (Taylor et al., 2016) primer sequences were appended to Illumina adapters sequences. DNA was amplified in 1X of Phusion High-Fidelity Mastermix (ThermoFisher, Waltham, MA, USA), 200 nm. of primers, 3% DMSO, 0.5 ug/uL of BSA, and 0.1 uL uL⁻¹ of DNA. The following cycling conditions used: 3 minutes at 98 °C, 25 cycles of 98 °C for 10 seconds, 55 °C for 30 seconds, and 72 °C for 30 seconds, with a final extension for 10 minutes at 72 °C.

The 16S and ITS amplicons were purified with 0.8X Ampure XP magnetic beads (Beckman Coulter, Brea, CA, USA) and underwent an additional round of amplification to add Dual Nextera XT indexes (Illumina Inc., Santa Clara, CA, USA) to the amplicons. These reactions were carried out in 1X Kapa HIFI HotStart ReadyMix and 0.1 uL uL⁻¹ indexes at 95 °C for 3 minutes, followed by 8 cycles of 95 °C for 30 seconds, 55 °C for 30 seconds, and 72 °C for 30 seconds, and extended at 72 °C for 5 minutes. As second round of bead purification was performed with 1.2X magnetic beads and the final libraries were pooled in equimolar ratios. The pool was denatured in NaOH and diluted to 8 pM. 15% of phiX was added to the library and the mix was run on the Illumina MiSeq Sequencer as paired end for 2X 250 cycles.

Paired reads (2x301 base pair) were processed with DADA2 (v1.6.0; Callhan et al., 2016) for filtering (parameters 205 maxN = 0, maxEE = c(2, 2), truncQ = 2). For the

16S reads, the forward and reverse reads were 206 trimmed to 15-230 and 20-190, respectively. For the 18S data, both the forward and reverse 207 reads were trimmed from 5-275. Chimeric sequences were predicted de novo and removed with 208 the `removeBimeraDenovo()` function in DADA2 using the “consensus” method. 16S and 18S 209 amplicon sequence variants (ASVs) were assigned taxonomy with Silva (v132; Quast et al., 2013), and the 16S 210 ASVs were further assigned with the RDP classifier v2.11 against training set 16 (Lan et al., 2012). The 211 taxonomy datasets were filtered to remove ASVs with a “chloroplast” or “mitochondria” family 212 assignment from the 16S dataset, or an “arthropoda” phylum assignment from the 18S dataset 213 (one of the samples was contaminated with a spider 18S sequence). Data are presented as 214 relative abundances (normalized to total read number for each sample).

Statistical Analysis

The study design creates the independent variables of site and depth, and response variables of microbial taxa and counts. For alpha diversity metrics, three indexes were selected: Shannon, Simpson Inverse and Chao. These were used to quantify the community diversity at depth, between management, and across sites. Non-Metric Dimensional Scaling (NMDS) was performed on OTU data using Bray-Curtis and Jaccard distance metrics to visualize sample similarities in two dimensions. The `metaMDS` function from the ‘vegan’ package (version 2.6-6.1) was used to compute NMDS coordinates, with stress values calculated to assess the fit of the reduced-dimensional representation. NMDS results were combined with metadata for further analysis, and visualizations were created with `ggplot2`, highlighting patterns by site and management type. The plots included confidence ellipses to represent group variability and were customized for clarity. Differential abundance analysis of microbial bacterial and fungal taxa was conducted using Analysis of Compositions of Microbiomes with Bias Correction (ANCOM-BC) to identify taxa that differed significantly across sites (Almond 1, Almond 3, and Almond 4). Phylogenetic data were processed using the `phyloseq` package in R, where sequences with zero abundance across all samples were removed prior to analysis. For each site samples were independently subset and analyzed using the `ancombc2()` function. The analysis was conducted at the phylum level, with site-specific covariates, including depth and management practices, incorporated into the fixed effects model. The p-values were adjusted for multiple comparisons using the Holm method. Differentially abundant taxa were identified with a significance threshold of $p < 0.05$, and log fold changes were calculated to assess the magnitude of these differences. Before analysis, data was tested for normality using a Shapiro-Wilkes test and non-normal data will be log-transformed. All statistical analyses were performed using RStudio (R Core Team, 2013, R package version 2.6-6.1).

RESULTS AND DISCUSSION

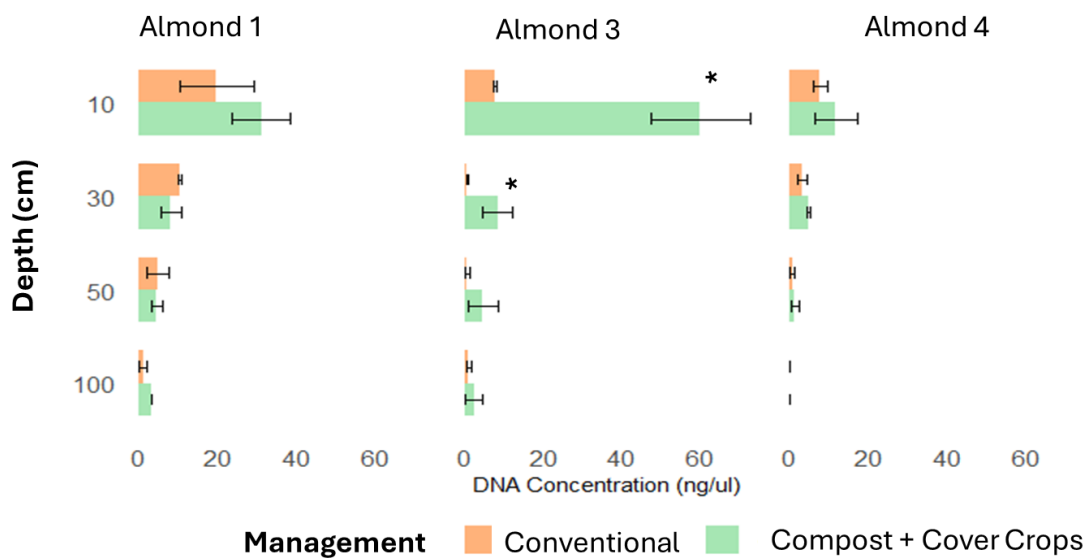


Figure 4-1. DNA concentration with depth across all sites, the color of the bars represents management plots of Conventional and Compost + Cover Crops. The error bars represent means ± 1 standard error.

DNA concentrations demonstrate that the effect of Compost + Cover Crops after two years does results in an increase compared to conventional management (Figure 4-1). The strongest effect is at the surface soils (0-10), where the compost and cover crops were applied. There was only a significant effect of compost + CC for GG 0-10 and 10-30 depth ($p < 0.05$), but there was a general trend of increase in across sites. These results are mirrored within other studies on the influence of various compost types on DNA concentration, with municipal solid waste compost (Bouzaiane et al., 2007; Srivastava et al., 2016). An increase in DNA concentration doesn't directly relate to an increase in community diversity but might be a measure reflecting the differences in total bacterial/fungal abundance between the sites.

BACTERIA

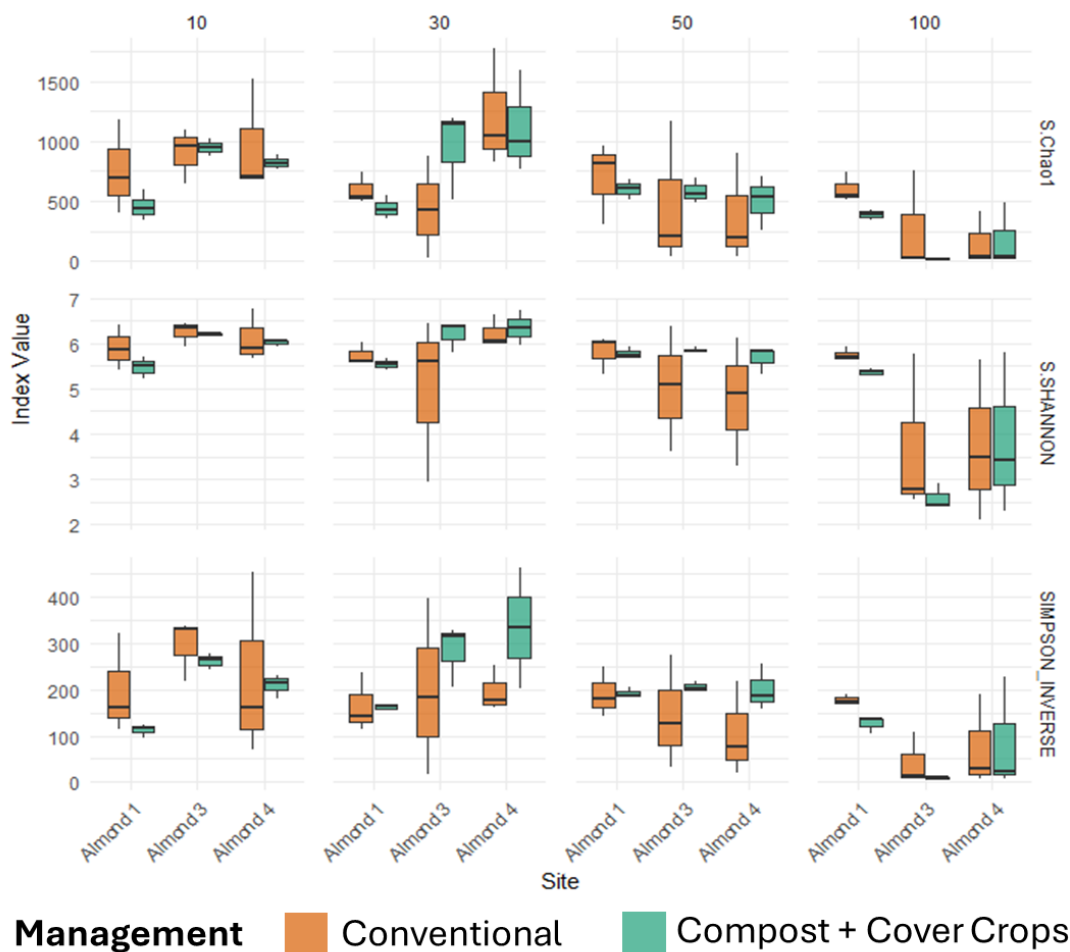


Figure 4-2. Bacterial alpha diversity indices of Simpson Inverse (dominance), S. Shannon (diversity), and S. Chao 1 (richness) across the three sites, box plots are median, with standard error.

Alpha-diversity, which measures the internal diversity of each sample, was calculated as a measurement of Chao 1 index (richness), Simpson Inverse index (dominance), and Shannon index (diversity). There were no significant differences in richness between plots across sites and depths. There was a trend of Compost + Cover Crops plots supporting higher richness at the 30 cm and 50 cm depths, as indicated by the S. Chao1 index. The Shannon diversity index shows that Compost + Cover Crops promotes slightly higher diversity with a more even distribution of species, while Conventional management exhibits slightly lower diversity, particularly in the 50 cm depth. The index of Simpson's Inverse values tends to decrease with increasing soil depth, indicating the community abundance shifted to fewer but more dominant members at greater depths for both management practices across all sites (Figure 4-2). In

the Almond 3 site, diversity is relatively higher at shallower depths (10-30 cm), with both Conventional and Compost + Cover Crops plots showing similar trends, though the Compost + Cover Crops plot exhibits slightly higher diversity in the 10-30 cm depth range (Figure 2, 3). At the Almond 1 site, there was a significant difference between surface soil's 0-10 depth diversity ($p < 0.05$), with the Compost + Cover Crops plots decreasing in diversity compared to conventional. After 30 cm the patterns are more uniform, with minimal differences between the two management practices, showing consistent diversity values across depths. In the Almond 4 site, diversity is higher at shallower depths, particularly in the 30-50 cm range, with the Compost + Cover Crops management showing a significantly higher diversity compared to the Conventional management ($p < 0.05$). Significant differences between the management practices are not apparent at deeper soil layers (below 50 cm), where all indices' levels out.

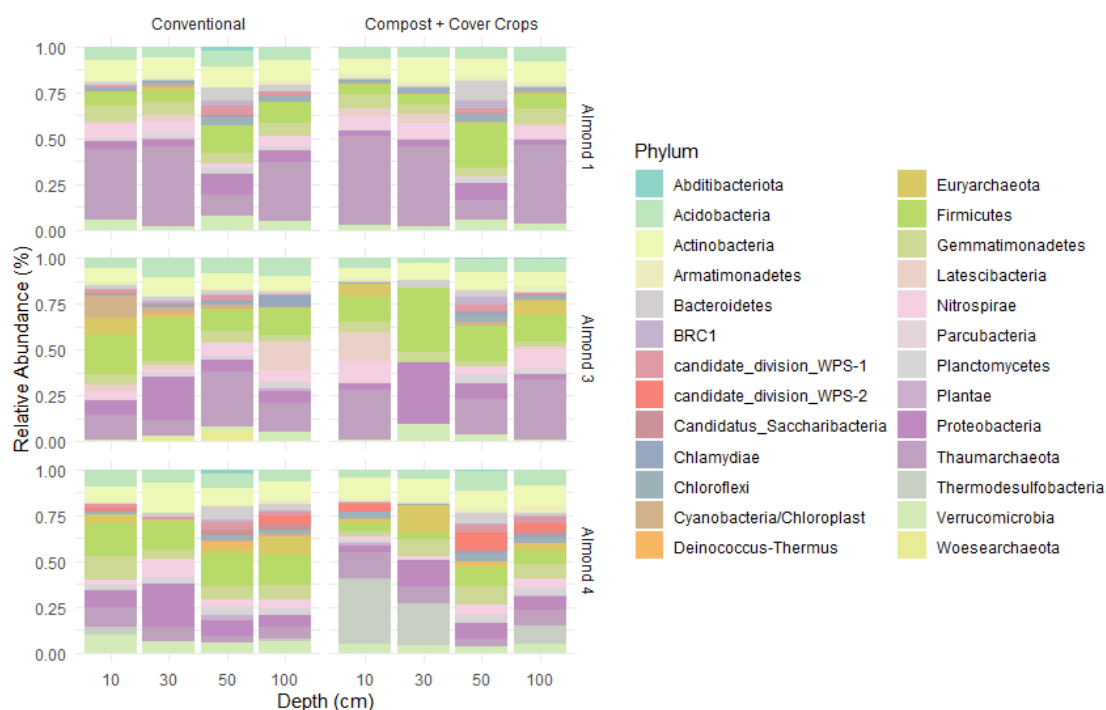


Figure 4-3. Relative abundance (%) of various phylum across the 3 sites, depths and management of Compost + Cover Crops and Conventional

The relative abundance of bacterial phyla varies with depth, with noticeable shifts in the composition of dominant phyla as soil depth increases (Figure 4-3). The average number of phyla represented across sites at the 0-10 cm depth is 17.5 ± 0.43 , while at the 100 cm depth, it is 9.5 ± 0.43 . This indicates a significant reduction in microbial diversity as soil depth increases, with nearly half the number of phyla present at deeper levels compared to the surface. In contrast, almonds grown in the Mediterranean climate of Spain under organic, rainfed conditions—without the use of fertilizers, pesticides, or other inputs—exhibited lower microbial diversity, with only seven observed phyla. This difference may be influenced by management practices or soil conditions (Özbolat et al., 2023). Despite the difference of diversity, a key similarity between the two studies is that

some of the most common phyla were *Actinobacteria* and *Acidobacterium*. *Actinobacteria* enhance nutrient and mineral availability, synthesize plant growth regulators, and are particularly effective in inhibiting plant pathogens (Siddharthan et al., 2022). They play diverse roles, including nitrogen fixation, phosphate solubilization, and siderophore production. The strong negative correlation between *Acidobacteria* abundance and soil organic carbon concentration suggests that some, if not all, members of this phylum may be oligotrophic, thriving in low-nutrient environments (Kielak et al., 2016; Fierer et al., 2007). Many members from these two phyla can produce a variety of cellulolytic and amylolytic enzymes to degrade fresh litter in the soil (Béguin and Aubert, 1994; Pandey et al., 2000).

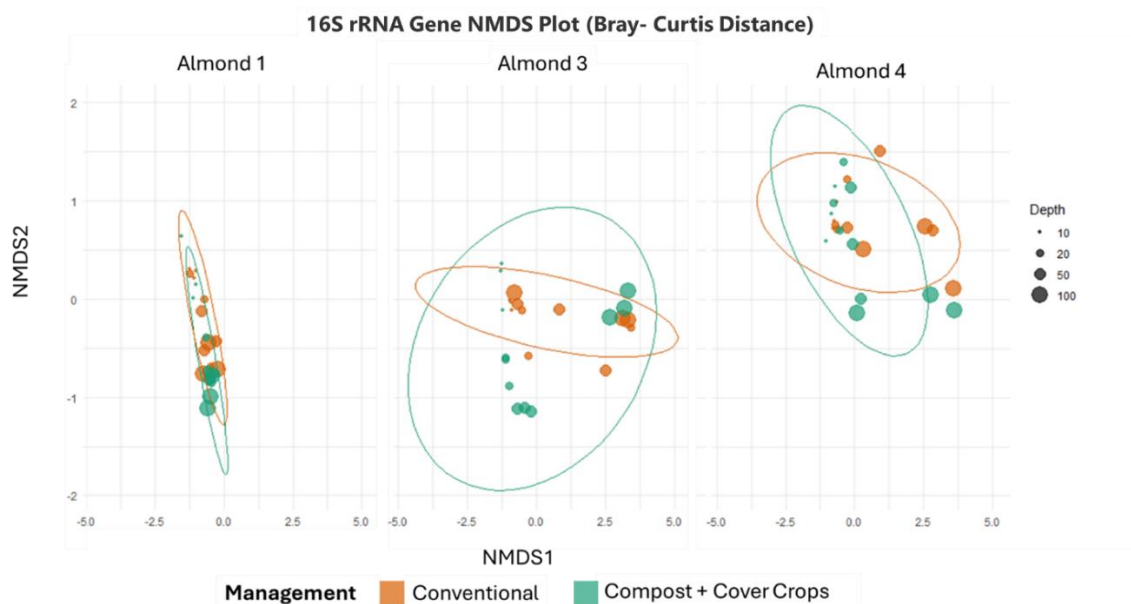


Figure 4-4. NMDS ordination displaying the bacterial communities across the three sites of Almond 1, Almond 3, and Almond 4

A non-metric multidimensional scaling (NMDS) analysis was used to visualize the dissimilarities in community composition between the samples of different management practices (Figure 4-4). As a measure of beta-diversity, NMDS plots in this study were obtained based on Bray-Curtis distances, which resulted in a stress of 0.12. The stress value of the soil bacterial community ordination of less than 0.2 suggests that the NMDS results had certain reliability (Taguchi & Oono, 2005). To support the visual analysis, a PERMANOVA was conducted to assess the effects of site, depth, and management (compost + cover crops and conventional) interactions on the Bray-Curtis dissimilarity of bacterial communities. The management practices and depths showed significant differences with noticeable separation between management plots, which was supported by the PERMANOVA results that management had a significant effect ($R^2 = 0.02$, $F = 2.7$, $p < 0.001$). Overall, samples from each site (Almond 1, Almond 3, Almond 4) demonstrated significantly different community structures ($R^2 = 0.15$, $F = 8.3$, $p < 0.001$). Specifically, the Almond 3 and Almond 4 sites showed moderate overlap between the management practices, indicating some distinction in microbial composition,

while the Almond 1 site exhibited less separation, suggesting very little effect of management on microbial diversity. Depth also influenced community structure ($R^2 = 0.13$, $F = 4.7$, $p < 0.001$), with deeper samples (larger points) often positioned separately from shallower ones, particularly in the Almond 3 and Almond 4 sites, indicating stratification in microbial communities with soil depth (Hao et al., 2020). There were also significant interaction effects with depth of plot ($R^2 = 0.03$, $F = 1.4$, $p = 0.04$) and site ($R^2 = 0.12$, $F = 2.3$, $p < 0.001$). Soil rhizosphere bacterial community structure was influenced by management, as seen in other studies (P. Wang et al., 2022)

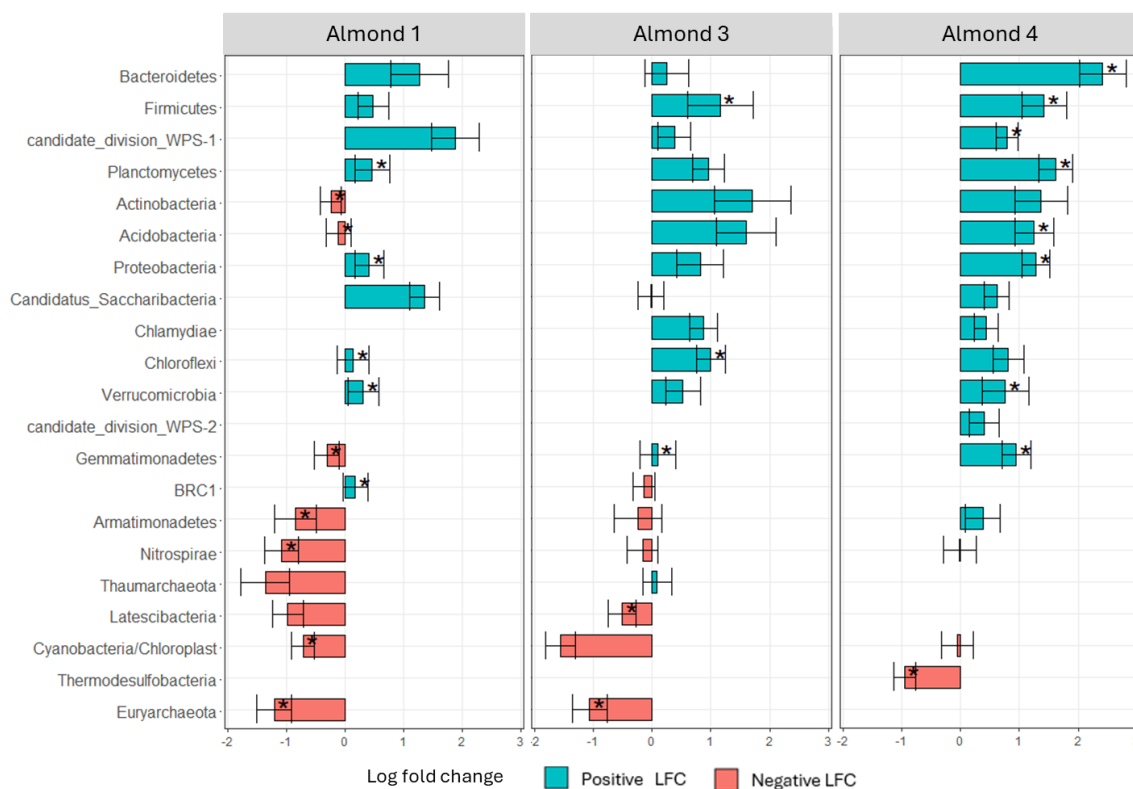


Figure 4-5. 16S community abundance analysis using ANCOMBC, highlighting the effect of soil depth on bacterial abundances. The analysis compares the differences in bacterial communities between topsoil (0-30 cm) and subsoil (30-100 cm). Blue Bars (Positive LFC) represent that the taxa have higher relative abundances in the topsoil compared to the subsoil. Red Bars (Negative LFC) represent that the taxa have higher relative abundances in the subsoil compared to the topsoil. Asterisk (*) denotes significant difference between soil depths.

The analysis of microbial communities across different soil depths reveals significant shifts in bacterial composition, emphasizing the impact of soil stratification on microbial diversity and ecological function (Figure 4-5). The results indicate that key phyla, such as *Actinobacteria*, *Acidobacteria*, *Firmicutes*, and *Proteobacteria*, are significantly ($p < 0.05$) more abundant in topsoil layers across all study sites. *Actinobacteria* are known for their role in decomposing complex organic matter and producing antibiotics, which help regulate soil microbial communities (Manikkam et al., 2019). *Acidobacteria*, abundant in nutrient-poor and acidic environments, contribute to

carbon cycling by degrading plant-derived organic compounds (Ivanova et al., 2020). *Firmicutes*, including many spore-forming species, are involved in the breakdown of organic material and play a role in the nitrogen cycle. Proteobacteria, which include numerous nitrogen-fixing bacteria, enhance nutrient availability and promote plant growth (Fang et al., 2019).

The higher abundance of these phyla in topsoil suggests that these layers harbor highly active microbial communities that facilitate essential ecological processes, such as carbon turnover through the degradation of complex carbohydrates, nitrogen fixation that supports plant growth, and phosphorus solubilization that enhances nutrient availability. The nitrogen cycle, which includes the denitrification process, plays a crucial role in returning fixed nitrogen back to the atmosphere from soil and water. Denitrification is facilitated by a range of soil microbiota, including members of the phyla *Proteobacteria*, *Actinobacteria*, and *Firmicutes*, as well as other soil eukaryotes (Gupta et al., 2016). *Verrucomicrobia* was also found to be more predominant in surface soils is often associated with the degradation of plant saccharides, monosaccharides and polysaccharides, in addition to metabolizing methane and methane oxidation (Baliyarsingh et al., 2022). These findings underscore the importance of topsoil microbial communities in maintaining nutrient cycling, enhancing soil health, and promoting ecological resilience.

Conversely, phyla such as *Cyanobacteria/Chloroplast* and *Euryarchaeota* exhibit higher abundance in subsoil environments, likely due to their adaptation to the anaerobic and nutrient-poor conditions that are more prevalent at greater soil depths (B. Frey et al., 2021). The presence of Cyanobacteria in subsoil layers may play a significant role in nitrogen fixation and the degradation of organic matter under low-light, oxygen-limited conditions, thereby influencing nutrient cycling and soil organic matter content even in deeper soil horizons (Álvarez et al., 2023). The phylum *Euryarchaeota*, a group of Archaea with notable methanogenic capabilities, was more pronounced in subsoils (Bintrim et al., 1997). This presence is significant because methanogenic Archaea contributes to methane production in anaerobic subsoil environments. The production of methane in these areas can impact greenhouse gas emissions and alter soil redox conditions (Conrad, 2020). The distinct microbial compositions observed between topsoil and subsoil highlight the complexity and stratification of soil ecosystems, emphasizing the need for land management practices that promote microbial diversity and resilience, which are crucial for sustaining soil fertility and enhancing long-term ecological sustainability.

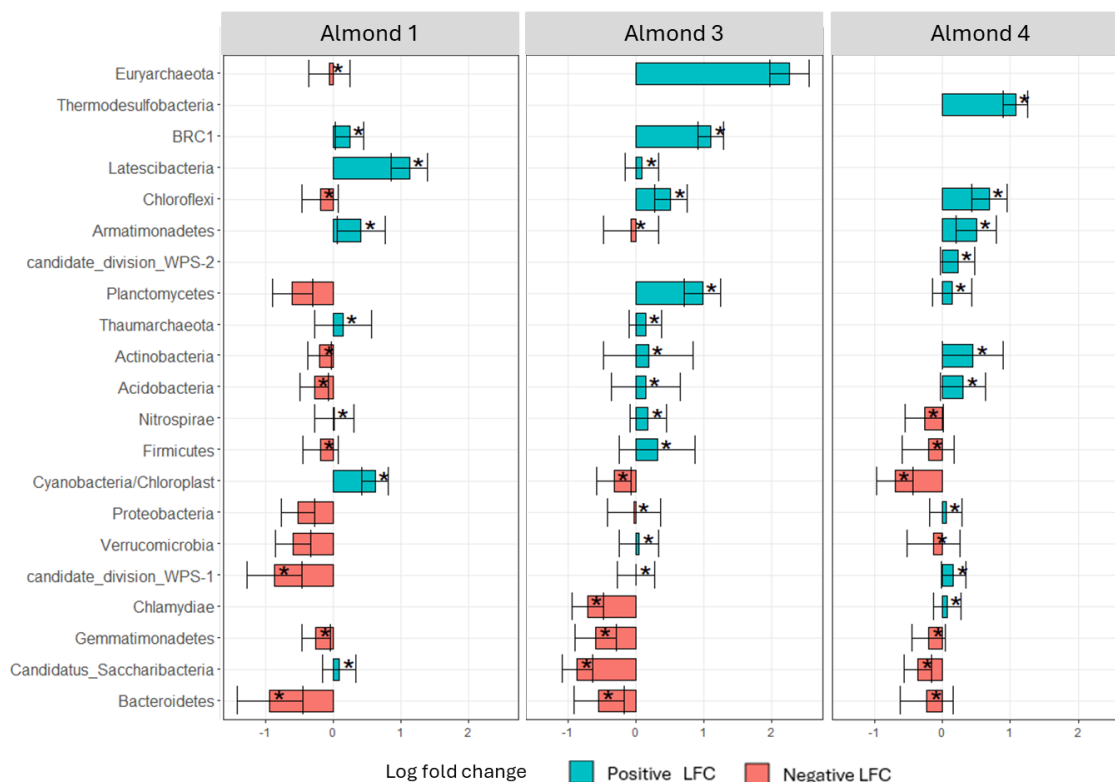


Figure 4-6. The relative abundance of various bacteria phyla in the whole soil profile (Almond 1, Almond 3, Almond 4) under different management practices (compost + cover crops vs. conventional). Log fold changes are used to quantify the differences between the two management regimes and are derived from the ANCOMBC, asterisks (*) denote statistically significant differences between the two management practices ($p < 0.05$)

The increase in certain microbial taxa under compost and cover crops management practices indicates significant benefits to soil health and fertility (Figure 4-6). Specifically, phyla such as *Chloroflexi*, *Thaumarchaeota*, *Actinobacteria*, and *Acidobacteria*, are more abundant under compost + cover crops management for most sites. These groups play crucial roles in nutrient cycling, organic matter decomposition, and nitrogen fixation (Bandopadhyay & Shade, 2024; Choi et al., 2017). For instance, *Chloroflexi* are involved in breaking down complex organic materials, enhancing nutrient availability and soil structure (Dai et al., 2018; Trivedi et al., 2016). *Thaumarchaeota* are crucial contributors to the nitrogen cycle, especially in the process of ammonia oxidation. They transform ammonia (NH_3) into nitrite (NO_2^-), a vital step in nitrification (He et al., 2012). This conversion directly impacts the availability of nitrogen in the soil, making it accessible for plant absorption and utilization by other microbes (Zhang et al., 2021). *Actinobacteria* improve soil health by decomposing organic material and producing secondary metabolites that can inhibit harmful microbes (Schlatter et al., 2022). Collectively, the increased presence of these beneficial microbes underscores the importance of compost and cover crops in fostering a more resilient and fertile soil.

ecosystem, supporting sustainable agricultural practice. Some phyla were consistently more abundant under conventional management across most sites (e.g., *Bacteroidetes*, *Gemmatimonadete*, *Cyanobacteria/Chloroplast*), which may indicate that these microbes either thrive under conventional practices or that the compost management does not favor their growth. The differences in response across sites highlight that microbial community changes depend not only on management practices but also on site-specific factors.

FUNGI

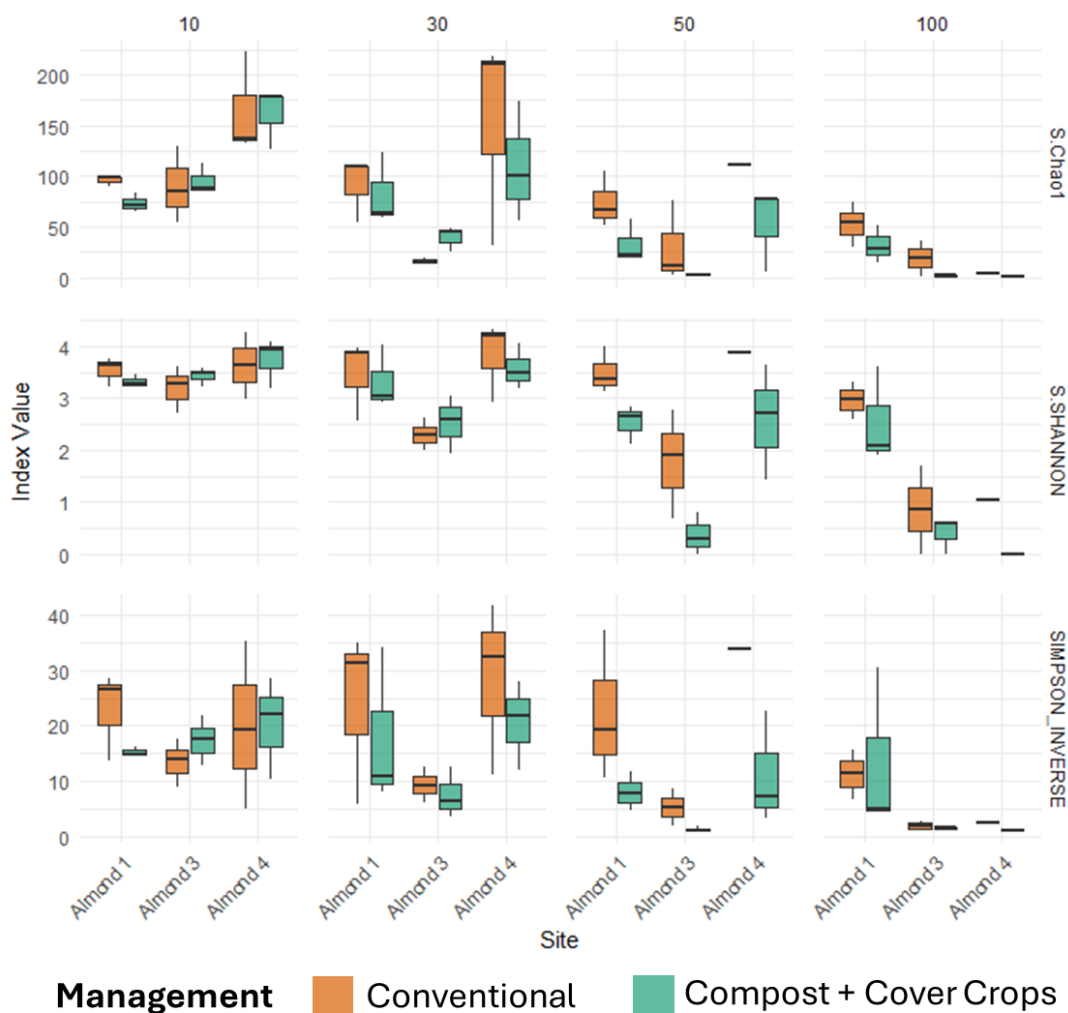


Figure 4-7. Alpha diversity indices of S. Chao, Shannon, and Simpson Inverse for the four depth increments of 10, 30, 50, and 100 across the three sites of the study for fungi. Box plots represent the median, with whiskers demonstrating the standard error.

Across all sites, the fungal diversity appears to generally decrease with increasing depth (Figure 4-7). The shallower depths (10 and 30 cm) tend to have higher diversity compared to deeper layers (50 and 100 cm). For fungi, the Conventional treatment tends to support greater fungal richness, particularly at 30 cm and 50 cm depths, as indicated by

the S. Chao1 index, whereas Compost + Cover Crops shows lower richness at deeper depths. The Shannon index reveals that Conventional management generally leads to greater fungal diversity and more even species distribution, especially at 30 cm and 50 cm, while Compost + Cover Crops displays more variable diversity across different depths and sites. With the Simpson inverse, a lower value represents higher diversity, whereas a higher value represents more dominance by a specific species. According to the Simpson Inverse index, Compost + Cover Crops promotes a more balanced fungal community at most depths, while Conventional treatment shows higher dominance of specific fungal species, particularly at 30 cm in Almond 3 and Almond 4. There was no significant difference between the management practices across the depths and sites.

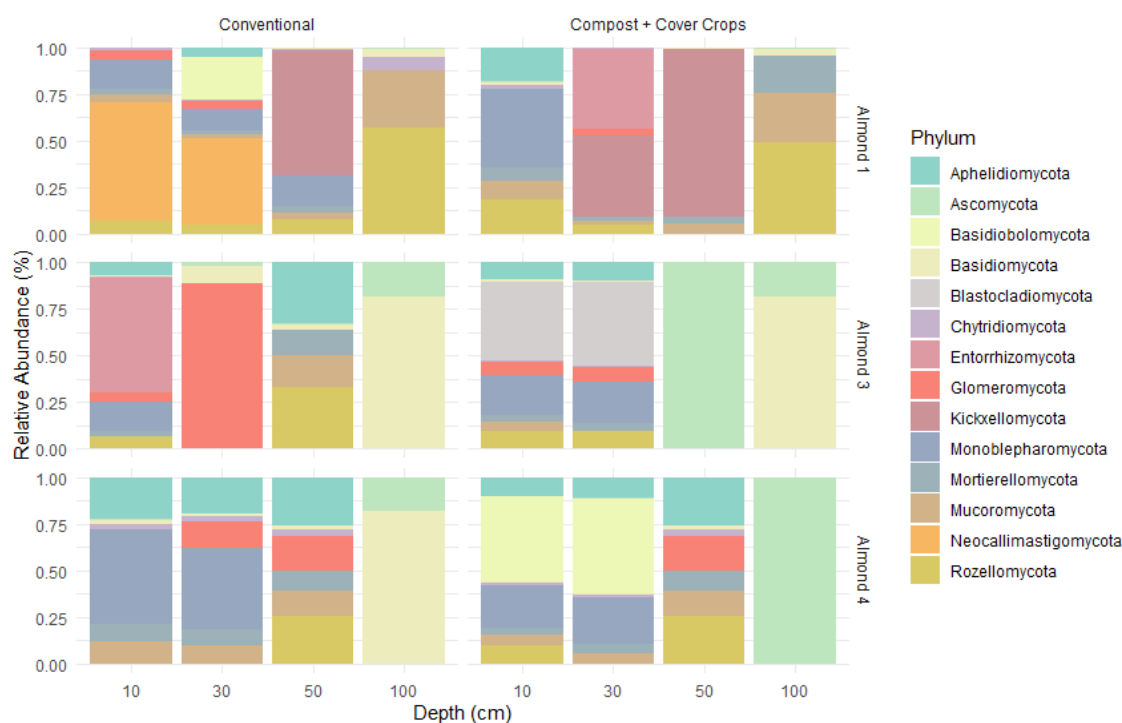


Figure 4-8. Relative abundance (%) of fungal phyla across the three sites and the four depth intervals.

Relative fungal abundance and community composition were assessed across three sites (Almond 1, Almond 3, and Almond 4) and the four depth intervals in Compost + Cover Crops and conventional soils. A total of 14 phyla were identified, with *Ascomycota*, *Basidiomycota*, and *Mortierellomycota* being the most dominant (Figure 4-8). Overall, fungal abundance composition varied significantly among sites ($p < 0.01$) and depths ($p < 0.01$), and with management ($p < 0.05$). Compost + cover crop soils tended to have higher overall fungal diversity, especially in terms of the representation of minor phyla. Conventional soils often show a more dominant presence of *Ascomycota*, with lower diversity overall. *Ascomycota* was the most dominant phylum at all sites, management, and depths, followed by *Basidiomycota* and *Mortierellomycota*. However, the relative abundance of these and other phyla varied considerably across sites and depths. Almond 3 exhibited lower fungal diversity compared to Almond 1 and Almond 4,

with Ascomycota being the predominant phylum at all depths. Fungal diversity generally increased with depth, particularly in Almond 1 and Almond 4. In Almond 3, deeper layers showed a slight increase in diversity, but it remained relatively low compared to the other sites. The relative abundance of various phyla varied across depths, generally decreasing with increasing depth, a pattern consistent with findings in other studies on depth-related influences (Bebber & Chaloner, 2022; Fierer et al., 2003; Hao et al., 2020). In a multi-year regional-scale survey involving 1,251 plots, it was found that soil pH had the most significant impact on fungal diversity. The effects of pH were generally unimodal, affecting fungal diversity both directly and indirectly through interactions with tree species, soil nutrients, and mold abundance (Tedersoo et al., 2020). While edaphic soil conditions might be a strong driver in diversity, some studies have found that competitive avoidance with other phyla also shapes community structure along the soil profile (Mujic et al., 2016). This suggests that microbial communities are not only structured by depth but also by the interactions among phyla, with some groups thriving niches while others avoid competition by occupying different soil layers.

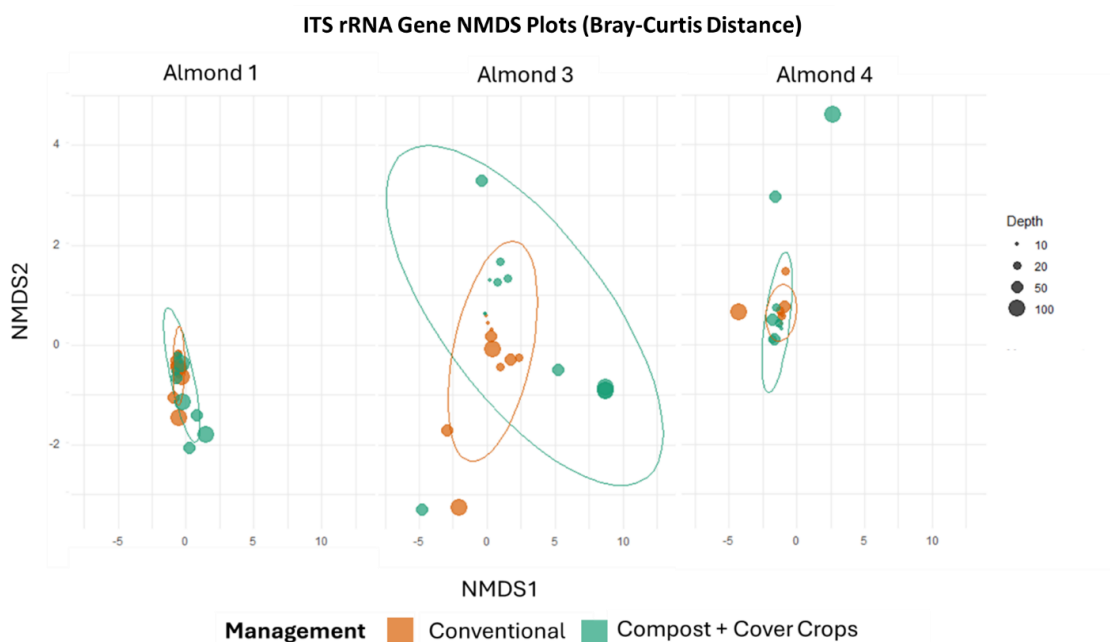


Figure 9. NMDS ordination displaying the fungal communities across the three sites of Almond 1, Almond 3, and Almond 4 along with depth, and management (Conventional + cover crops).

The NMDS ordination found that the communities of fungi across sites were significantly different from one another, and there was a slight effect of management on the communities (Figure 9). The permutation test for homogeneity of multivariate dispersions was conducted to assess whether the dispersions among groups were homogeneous. The results of the test indicated a significant difference in dispersion between the groups ($F = 9.26$, $p = 0.003$). This suggests that the variability within the conventional and compost + cover crops plots is significantly different. The PERMANOVA analysis was conducted to assess the effects of site, depth, and management (compost + cover crops and control) interactions on the Bray-Curtis

dissimilarity of fungal communities. The results showed significant effects for management ($R^2 = 0.03$, $F = 2.7$, $p < 0.001$), site ($R^2 = 0.12$, $F = 4.8$, $p < 0.001$), depth ($R^2 = 0.07$, $F = 2.02$, $p < 0.001$), and the interactions plot ($R^2 = 0.06$, $F = 2.29$, $p < 0.001$) and site ($R^2 = 0.11$, $F = 1.47$, $p < 0.001$). Fungal communities have been shown to be significantly influenced by compost additions (Goyer et al., 2022), with cover crop species mixes having an even greater impact on the composition of fungal types (Cloutier et al., 2020).

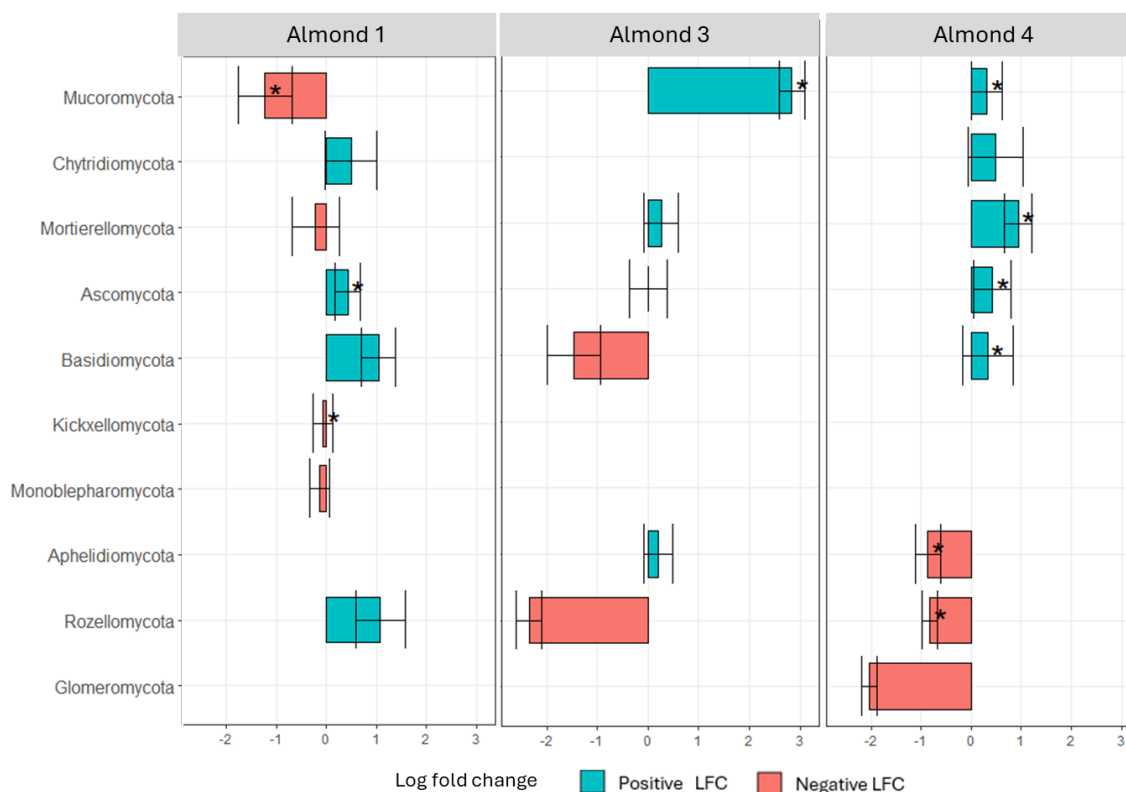


Figure 4-9. ITS fungal community abundance analysis using ANCOMBC, highlighting the effect of soil depth on fungal abundances. The analysis compares the differences in fungal communities between topsoil (0-30 cm) and subsoil (30-100 cm). Blue Bars (Positive LFC) represent that the taxa have higher relative abundances in the topsoil compared to the subsoil. Red Bars (Negative LFC) represent that the taxa have higher relative abundances in the subsoil compared to the topsoil. Asterisk (*) denotes significant difference between soil depths .

There were several key patterns emerge in fungal community composition across the three sites and depth intervals (Figure 10). *Ascomycota*, a phylum known for its role in plant decomposition and nutrient cycling, consistently exhibits significant enrichment in topsoil across all sites, suggesting its preference for the upper soil layer. *Basidiomycota*, often associated with wood decay and mycorrhizal symbiosis, shows more variation in its distribution, with significant enrichment in topsoil at some sites and subsoil at others. This suggests that *Basidiomycota* may have a broader ecological niche compared to *Ascomycota*, adapting to both topsoil and subsoil environments. *Mortierellomycota*, another group of fungi that can play a role in organic matter

decomposition and nutrient mineralization, is also significantly enriched in topsoil at Almond 3 and Almond 4. *Chytridiomycota*, known for its role in decomposing organic matter, is enriched in topsoil at Almond 1, suggesting a potential preference for nutrient-rich conditions. In contrast, the phylum's of *Kickxellomycota*, *Aphelidiomycota*, and *Rozellomycota* are predominantly saprotrophs, and are more often found in subsoil and associated with decomposing plant matter (Nguyen et al., 2016). These fungi play a crucial role in the decomposition process, breaking down organic matter and releasing nutrients into the soil (Várnai et al., 2014). These findings suggest that these phyla may have specific adaptations to the conditions found in deeper soil layers, which is corroborated by a deep soil layer study in a drought exposed forest system by Frey et al., (2021). As soil depth increases, levels of organic carbon, nitrogen, and clay content decrease significantly (Zhou et al., 2019). This decline is accompanied by reductions in fine root biomass, microbial biomass, fungal abundance, all of which are strongly correlated with these physicochemical change (Frey et al., 2019).

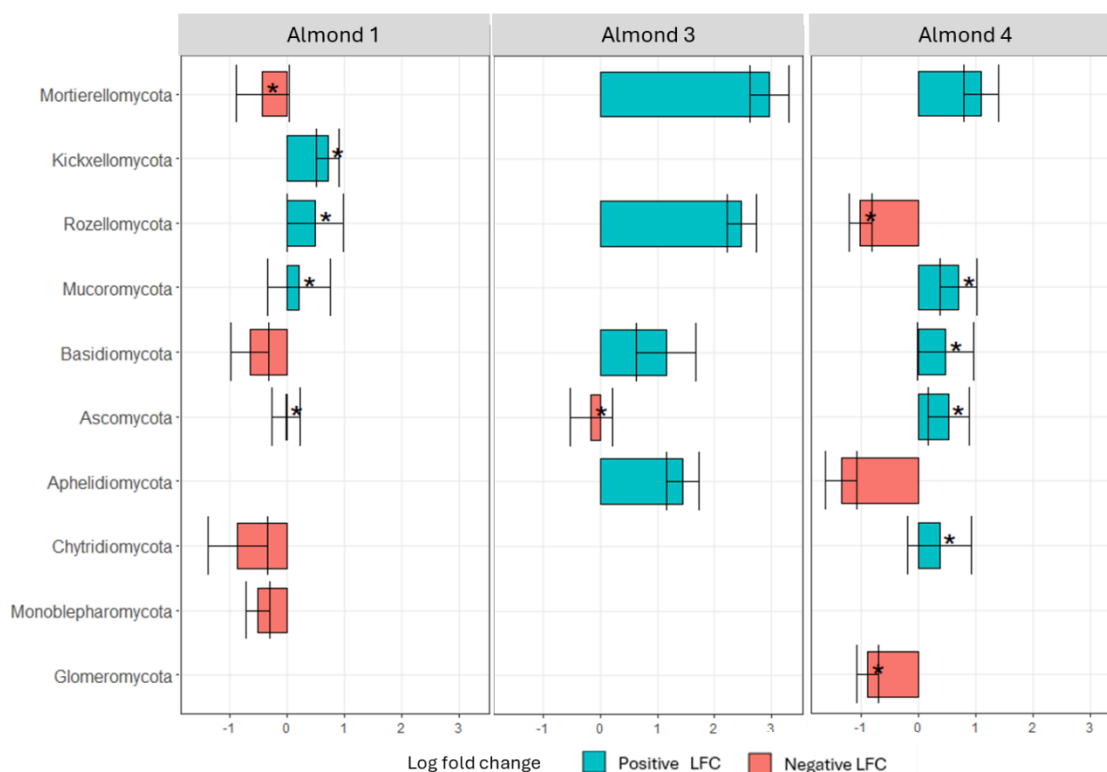


Figure 4-10. The relative abundance of various fungal phyla in the entire soil profile (GG, NAV, WS) under different management practices (compost + cover crops vs. conventional). Log fold changes are used to quantify the differences between the two management regimes and are derived from the ANCOMBC, Blue Bars (Positive LFC) represent that the taxa have higher relative abundances in the compost soils, while red bars (Negative LFC) represent that the taxa have higher relative abundances in the conventional plots soil. Asterisks (*) denotes that phyla that were statistically significant different between the two management practices ($p < 0.05$).

The analysis of fungal community composition under different management practices revealed distinct patterns across sites and with management (Figure 11). Notably, *Mucoromycota* and *Ascomycota*, key players in organic matter decomposition and nutrient cycling, demonstrated significant increases in abundance under compost + cover crop management at multiple sites (Leifheit et al., 2024). This suggests that these practices may create favorable conditions for these beneficial fungi with the input of simple and complex plant materials and organic matter, which can stimulate fungal activity (Dang et al., 2021). While *Mortierellomycota* did not exhibit statistically significant changes at all sites, it showed substantial shifts in abundance at GG, highlighting its responsiveness to management practices (Muneer et al., 2021). *Rozellomycota* and *Basidiomycota* also experienced notable changes in abundance at GG, although these effects were not statistically significant. These findings underscore the complex interplay between management practices, site conditions, and fungal community dynamics and suggest that even subtle changes in land use can have significant impacts on soil microbial communities.

MICROBIOME DYNAMICS

When organic matter is introduced into the soil, bacteria and fungi interact in a complex interplay to decompose the heterogenous material. Soil organic matter contains up to 25% cellulose, hemicellulose, and soluble sugar (Stevenson, 1994). A large percentage of cellulose is present as ligno-cellulosic complex, and is an intricate arrangement of polysaccharides, lignin, and protein (Isikgor & Becer, 2015). Fungi often specialize in breaking down complex compounds, such as lignin, while bacteria target simpler substrates like root exudates (Koo et al., 2005; Ullah et al., 2023). In our study, we observed significant increases in both fungi and bacteria involved in organic matter decomposition and nutrient cycling. The decomposition of fresh organic matter from compost and cover crops likely followed a sequential process, with fungi acting as primary decomposers. Fungi, particularly saprotrophic species within the phyla *Ascomycota* or *Basidiomycota*, released extracellular enzymes such as lignin peroxidases and cellulases to break down complex organic compounds like lignin and cellulose, which are resistant to degradation (Woodward & Boddy, 2008). Once fungi had reduced these polymers into smaller, more accessible compounds, bacteria, including copiotrophic species, likely followed by producing enzymes such as amylases, proteases, and lipases to further break down simpler organic molecules like sugars, peptides, and lipids (Stone et al., 2023). This cooperative decomposition process facilitated the biodegradation of resistant organic matter and promoted the release of nutrients like nitrogen, phosphorus, and carbon, which are essential for both plant uptake and microbial activity. While we do not have a time series of community shifts, that is what would be needed to properly capture the potential succession of decomposition within these soil systems.

The complementary metabolic capabilities of fungi and bacteria underscore the importance of microbial succession in organic matter processing, where the enzymatic actions of one group create more favorable conditions for the next. This cooperative relationship enhances overall soil health and contributes to nutrient availability in the system (Baćmaga et al., 2023; Rao et al., 2014). However, this interaction can also involve competition for specific nutrients depending on the type of organic matter and

soil conditions such as soil acidification and drought which can increase fungal competitiveness (C. Wang & Kuzyakov, 2024). After the addition of fresh organic matter, specialized microorganisms that decompose new material proliferate rapidly, potentially leading to a priming effect where competition for energy and nutrients enhances the decomposition of existing soil organic matter (R. Chen et al., 2014). Understanding these dynamics is crucial for optimizing soil health and fertility through targeted management practices.

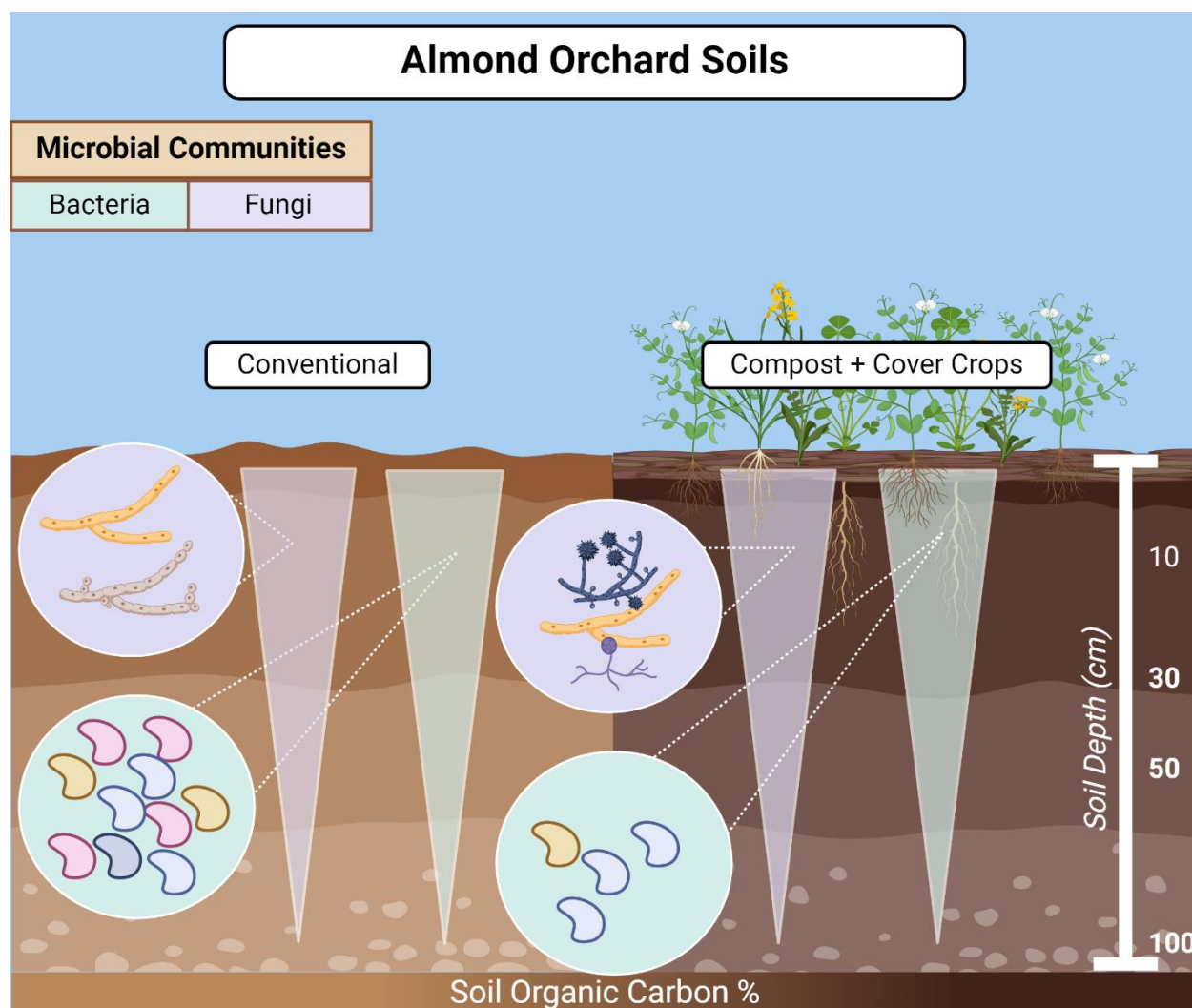


Illustration: 4-1 Hypothesized impact of compost and cover crops on almond orchard soils microbial communities of bacteria and fungi. In both systems, there is a significant decrease in community abundance and diversity with depth. Within the compost + cover crop system, we expect less diversity but greater abundance for both fungi and bacteria, a shift towards phyla that facilitates organic matter decomposition and mineralization.

MANAGEMENT IMPLICATIONS

In almond orchards, the strategic application of compost and cover crops has significantly influenced microbial communities within orchard alleys. Within two years of implementing these practices, we observed a notable shift in microbial community composition, marked by an increase in taxa specialized in decomposing organic matter (Figure 12). This shift often resulted in a decrease in overall microbial diversity, linked to a transition toward fewer but more specialized species of both bacteria and fungi. This was counterintuitive to our original hypothesis that compost and cover crops would increase diversity, as was reported in other studies (Farrell et al., 2009; Heisey et al., 2022; Xu et al., 2023). Targeted management practices, such as the use of compost and cover crops, alter the soil environment by introducing fresh organic matter. This can foster a more functionally focused and robust microbial community, enhancing soil health and addressing key challenges in orchard management. By promoting microbial communities that enhance soil organic matter processing and nutrient cycling, orchards can achieve improved sustainability, productivity, and resilience, underscoring the importance of integrating soil microbiome management into broader orchard management strategies.

CONCLUSION

Our study revealed distinct microbial assemblages across almond orchard sites, with a marked trend of decreased microbial diversity with increasing soil depth, particularly pronounced in fungal communities compared to bacterial ones. Conventional management plots exhibited surprisingly high bacterial diversity. Shifts in both bacterial and fungal phyla in response to management changes, especially with the application of compost and cover crops, suggest an enhancement in microbial balance and activity. Bacterial communities were diverse and predominantly beneficial, significantly contributing to nutrient cycling with minimal adverse effects. Although fungal diversity did not significantly increase under compost and cover crop treatments, there was a notable rise in the abundance of specific phyla often involved in organic matter decomposition, reflecting the impact of management changes on nutrient cycling. These findings emphasize the ability of soil management to influence microbial communities, prompting more questions about the functions and balance of the community. The observed variations in microbial diversity across soil depths and management practices underscore the need for more refined microbial sequencing in the Central Valley to better understand and manage the complex microbial ecosystems within almond orchards. Future research using quantitative PCR could provide deeper insights into species-specific roles within these systems. This enhanced resolution is crucial for developing sustainable orchard management strategies and optimizing soil health.

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CHAPTER 5: CONCLUSION

This dissertation demonstrates the potential of soil health practices, such as compost application and cover cropping, to significantly enhance the sustainability of

almond and walnut orchard systems in California's Central Valley. Key findings reveal that these practices enhance soil organic carbon (SOC) storage, improve soil structure, and promote fungal and bacterial phyla related to nutrient cycling, critical for long-term orchard productivity and soil health. Importantly, these practices also offer a path toward greater climate resilience and mitigation, with enhanced carbon sequestration playing a vital role (Fenster et al., 2021; Khangura et al., 2023).

A pivotal outcome of this research is the significant increase in SOC within the management plots of Compost and Cover Crops, particularly by the third year, demonstrating that the benefits of these interventions take time to manifest fully (Maharjan et al., 2024). Promoting and sustaining soil health management presents significant challenges due to its complex, interdependent components and iterative nature. This research highlights the improvement of physical soil health, particularly in wet aggregate stability, which enhances water retention and reduces erosion risks—crucial attributes in the face of increasing intense weather events and potential flooding. These physical improvements are complemented by shifts in microbial communities, which play a vital role in nutrient cycling and organic matter decomposition, emphasizing the biological advantages of adopting these practices.

A distinctive aspect of this dissertation is its depth-focused approach, with soil measurements extending to one meter. This comprehensive perspective revealed the deeper impacts of compost and cover crop practices, offering novel insights into soil dynamics within almond orchards in California—a system with limited data on deeper SOC storage. Deep soil carbon storage is an important and often overlooked part of soil carbon dynamics. Over 70% of soil carbon is stored below 20 cm within the soil system (Hicks Pries et al., 2023). By incorporating depth-oriented analyses, this study uncovered significant variations in microbial activity and carbon storage across the soil profile, demonstrating that the benefits of these practices extend well beyond surface soils. These findings emphasize the importance of considering the whole soil profile in soil health interventions, highlighting the potential for long-term carbon sequestration and enhanced ecosystem functioning. This work contributes valuable data to the field, advancing our understanding of the depth-specific impacts of soil health practices and their implications for sustainable orchard management.

Despite these promising outcomes, variability in responses across orchard sites and soil types highlights the influence of underlying site-specific mechanisms on the rate at which soil health improves. Some soils responded more rapidly to changes in management, while others required more time or exhibited subtler improvements. This variability underscores the critical roles of factors such as soil type, initial soil conditions, climate, and management history in shaping the trajectory of soil health recovery (Cambardella & Elliott, 1992; Six et al., 2002). For instance, soils with higher clay content tend to stabilize organic carbon more effectively, potentially accelerating the benefits of compost and cover crops in those systems (Lützow et al., 2006). Conversely, sandy soils may require longer periods to exhibit noticeable improvements due to lower cation exchange capacity and organic matter retention (Lehmann & Kleber, 2015). Nevertheless, the consistent improvements observed across sites affirm the broad potential of these practices to enhance soil health and ecosystem resilience

Another challenge is the uncertainty surrounding the long-term persistence of carbon added through soil health practices, particularly regarding its stabilization in deeper soil horizons and the role of root exudation in enhancing carbon sequestration. While SOC levels significantly increased in this study, questions remain about the mechanisms governing the retention and stabilization of this carbon, especially under varying environmental conditions and management regimes (Schmidt et al., 2011). Carbon inputs from compost and cover crops are subject to microbial decomposition, and their eventual stabilization depends on interactions with soil minerals and the formation of stable aggregates (Lehmann & Kleber, 2015).

The potential for increased carbon sequestration through root exudation is particularly intriguing but remains poorly quantified. Root exudates contribute labile organic carbon to the rhizosphere, fostering microbial activity and promoting soil aggregation (Kuzyakov & Domanski, 2000; Li et al., 2024). However, the extent to which these processes lead to long-term carbon storage, as opposed to rapid mineralization and CO₂ release, is still uncertain. Research on root-derived carbon highlights its potential to form stable mineral-associated organic matter (MAOM), but its persistence varies widely depending on soil type and microbial community composition (Cotrufo et al., 2013). Within our study, we hypothesized that the increase in SOC storage is directly related to the application of compost and is likely within the labile pool of carbon in the soils. In this way, we are not directly increasing soil C sequestration.

Future research should focus on tracking carbon dynamics over time, integrating advanced methods such as isotope tracing and spectroscopy to differentiate between stabilized carbon pools and transient carbon fractions. Research suggests that the primary controlling factor controlling the breakdown of soil organic matter is how easily microorganisms can access it, rather than the inherent resistance of the organic matter itself (Dungait et al., 2012). Long-term experiments are needed to evaluate whether the observed gains in SOC contribute to sustained climate mitigation efforts or are subject to eventual loss through microbial respiration. Understanding the balance between carbon inputs, stabilization, and losses will be critical for designing soil health practices that maximize their contribution to carbon sequestration and long-term ecosystem sustainability.

This dissertation also opens new avenues for research into how soil health practices affect the nutritional quality of almonds and walnuts. Improved soil health may enhance crop quality by increasing nutrient availability and reducing water stress during critical growth periods. Understanding how these practices impact secondary metabolites, antioxidants, or other quality markers could link soil management to marketable improvements in crop value.

In conclusion, this research provides strong evidence that compost and cover cropping can significantly improve orchard soil health, contributing to both climate resilience and climate mitigation. However, the complexity of soil systems and variability in responses across sites emphasize the need for site-specific strategies and longer-term studies. This work contributes valuable insights to the growing field of sustainable soil

management, offering guidance for future practices, policies, and research aimed at improving soil health and productivity in California's orchard landscapes.

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