EXPLORING THE EFFECTS OF VINEYARD CANOPY MANAGEMENT PRACTICES ON SOIL MICROBIAL ACTIVITY

By

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EXPLORING THE EFFECTS OF VINEYARD CANOPY MANAGEMENT PRACTICES ON SOIL MICROBIAL ACTIVITY

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Title of Study: Exploring the Effects of Vineyard Canopy Management Practices on Soil Microbial Activity

Major Field: International Agriculture

Abstract:

The purpose of the study was to investigate soil fertility and microbial decomposition associated with vineyard canopy management practices which introduce plant litter into the soil. The study specifically focused on the mineralization and immobilization of carbon and nitrogen. In addition, the study also addressed how vineyard fungal disease management influences soil microbial activity. The use of fungicide to limit the presence of fungal disease in the canopy may suppress the vineyard soil's microbial activity and thus may alter the release of nutrients from the plant litter back into the soil. In addition, soil microbial activity may be increased through residue management.

For the study, five plant residue types obtained from a Vitis vinifera hybrid 'Chambourcin' from the Cimarron Valley Research Station in Perkins, Oklahoma, were added to vineyard soil. The plant litter treatments consisted of grapevine pruning (i.e., woody plant material from the dormant period of the grapevine), grapevine leaves and shoots, fruit, and aboveground tissues from two cover crops, wheat, and clover. Additionally, half of the incubation treatments received Mancozeb, a commonly used fungicide in vineyards, to examine the potential effect of viticultural disease management practices on soil microbial populations. Wheat, the highest quality litter in terms of carbon to nitrogen ratio, had the highest amounts of ammonium and nitrate, microbial biomass carbon and cumulative CO_2 respiration. Fungicide additions suppressed microbial activity and initially prevented the conversion of nitrate to ammonium. Targeted canopy management in vineyard systems may thus improve soil fertility and provide pathways to more sustainable fertilizer use.

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CHAPTER I

INTRODUCTION

To achieve global food security, global food production must increase by 70% by 2050 (Food and Agriculture, 2009). While the current practices rely heavily on inorganic fertilizers to increase food production, these inorganic fertilizers come with a high price in terms of cost and environmental degradation. Therefore, increasing soil fertility and decreasing fertilizer cost are central to the sustainable and ecological management of agricultural systems. A possible solution to increase soil fertility while decreasing reliance on fertilizers is the use of organic amendments and plant litter residue. Farmers are currently looking at ways to increase soil fertility through targeted management of plant residues generated during the growing season. For instance, in vineyard systems, nitrogen is the most limiting element for grapevines, and annually a range of 30-100 lbs. of nitrogen is applied, mostly in the form of inorganic fertilizers. (Kurtural et al.) The reliance on inorganic nitrogen fertilizer may be reduced when plant litter residues are added to and retained within the soil. The plant litter additions may come from management practices already in place for the cropping systems. Vineyard management systems provide many opportunities for residues to be retained in the plant-soil system, but there is uncertainty around how various plant litter additions impact the soil nitrogen and carbon cycle in vineyards. For this reason, this study aims to shed light on the specific effects of shoot thinning, dormant pruning, fruit removal, and cover cropping on the nitrogen and carbon cycles, and ultimately, to evaluate potential implications for soil fertility and health.

Canopy management is important for promoting fruit quality and yield; these practices likely influence soil health and fertility by introducing plant litter into the soil. Throughout the year, different vineyard management practices incorporate different types and quantities of litter into the soil. For instance, in late winter and early spring, while the vines are in dormancy, the vines are pruned to remove the dormant wood from last year's growth and promote fruit production in the upcoming year. Once the dormant wood is removed, the dormant wood may be shredded and tilled into the soil profile. A few months later, the vines receive fungicide treatments via an air blast sprayed at the canopy. While the main goal of the fungicide treatments is to prevent fungal infections in the canopy, a certain volume of fungicide reaches the soil profile. When summer begins, the vines may be shoot thinned, removing the green vegetative growth of the nonfruit bearing shoots to open the canopy. Also, during the summer, the vines may be fruit thinned, removing the small fruit clusters higher in acid and low in sugar content to promote nutrient synthesis in the remaining clusters, to increase their sugar content, and lower the acidity.

These practices introduce other forms of plant litter that differ in chemical composition and nutrient content. In late summer into the fall, the vineyards are harvested and some of the fruit may enter the soil profile, either as spillage or to deter disease from infecting the fruit that could not be harvested. The decomposition of these plant residues entering the soil profile in the vineyard throughout the year is impacted by the complex set of soil processes involving chemical, physical, and biological agents. The litter decomposition is also influenced by the litter chemical composition and environmental conditions in the soil (e.g., pH, moisture, temperature) (Feng et al., 2011). Despite the complexity involved in the decomposition process, a key way of understanding litter decomposition is by examining the carbon and nitrogen (C/N) ratio of the litter (Walse, 2008; Gul et al., 2012; Berg, and Sverdrup, 1998). The C/N ratio of the plant litter acts as a control on the decomposition process and aids in predicting the number of

nutrients released from the plant litter and returned to the plant-soil systems through soil microbial activity. As soil microbes decompose plant litter releasing nutrients stored in the plant biomass, the microbes convert other nutrients in the soil profile from organic into inorganic forms, in particular nitrogen, a main nutrient used to enhance plant growth and yields. For this reason, soil microbes are a critical agent for promoting soil health and fertility (Kumar and Verma, 2019). The process of plant litter degradation by soil microbes has been studied across a range of experimental contexts that vary in factors such as climate, plant species, and management. Since most studies have been conducted on fruit trees, forestry, and their respective litter addition, the effects of residue management on soil fertility in vineyard soils are not well understood. However, these residue managements may promote soil health and fertility in vineyards congruent with the results found in the fruit tree and forestry studies.

CHAPTER II

LITERATURE REVIEW

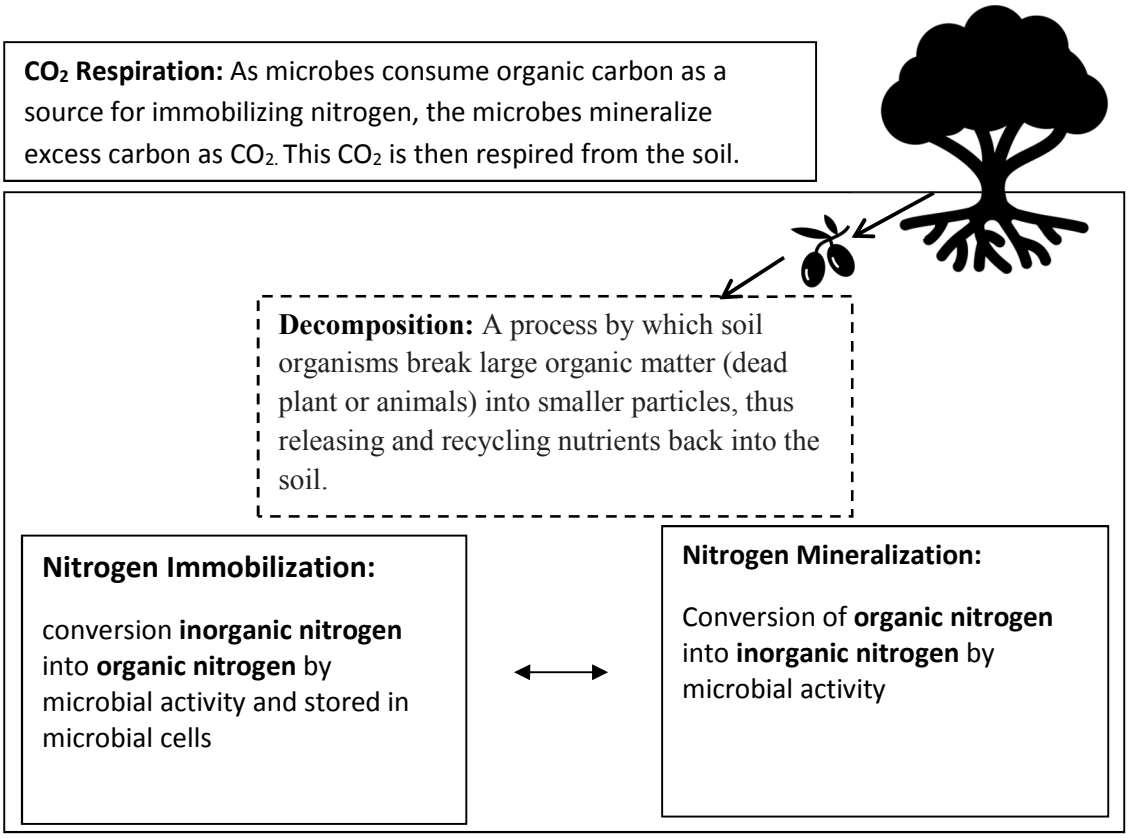

Figure 1: Illustration and definitions of the decomposition, immobilization, mineralization, and respiration processes. The CO_2 respiration may be referred to as CO_2 mineralization. CO_2 mineralization is the process of soil organic carbon converting to CO_2.

Soil microbes and plants are key players in nutrient cycling. As soil microbes grow, they promote nutrient cycling by decomposing dead plants and fallen plant litter. Throughout decomposition the microbes release nutrients back into the soil which can then become available to plants. The plants then take up the nutrients for their growth until the plant dies, decays, and the plant litter returns to the soil. However, not all plant litter will stimulate microbial activity. The microbial response often depends on the quantity and quality of the litter.

Most agricultural and forestry practices influence the amount of plant litter entering the soil. For instance, Smolander et al. (2010) demonstrated removing litter from logging altered the decomposition, carbon and nitrogen cycling processes, and the quality of soil organic matter. Their study found the carbon mineralization rate was lower after ten years when all the living branches, dead branches, and needles were removed compared to when only a portion of the logging material was removed. Additionally, the rate of net nitrogen mineralization and the amount of carbon and nitrogen in the microbial biomass was lower in the complete removal of logging material than when only a portion of the logging material was removed. This provides evidence that removing plant litter from the soil influences microbial activities, nitrogen mineralization, and amounts of carbon and nitrogen present (Smolander et al., 2010).

The results from this study are congruent with findings from other studies which focused on different types of fallen litter. Fallen litter from nearby plants and trees is a

critical component of nutrient biogeochemical cycles in natural and agroforestry systems. A study by Bai et al. (2022) showed the nutrient inputs in the soil changed as a response to litter from various plant types. In their study they used fallen litter of two shade tree species, *Gliricidia sepium* and *Canarium indium*, and a cocoa tree, *Theobroma cocoa*. The litter from G. *sepium* shade tree had more litter mass loss than the cocoa and indium trees. Additionally, the litter from the G. *sepium* showed a higher average in total nitrogen concentration; however, this litter showed lower carbon to nitrogen ratio and nitrogen release, which suggests nitrogen mineralization occurs more in G. *sepium* litter than in the other two tree litters. Their study highlights the importance of understanding how different tree litter affects the total nitrogen concentration, carbon to nitrogen ratio, and nitrogen release in soil treated with the same plant litter from different plant species. Yokobe (2020) further explored this concept in a natural forest ecosystem, highlighting the effect of differing litter types and quantities on microbial biomass and nitrogen mineralization. The study used coarse litter composed of woody material from large roots and fine litter. The fine litter was composed of leaves and small roots. Therefore, litter had varying degrees of carbon to nitrogen ratios, which was the best predictor of microbial biomass accumulation among the coarse and fine litter treatments (Yokobe, 2020). This study shed light on how the different quantities of coarse or fine litter may be linked to soil microbial abundance and nitrogen mineralization.

The chemical composition of litter inputs also influences soil microbial biomass and nitrogen mineralization. A study conducted by McClaugherty et al. (1985) demonstrated the decomposition processes and changes in the nitrogen and organic chemical content of six types of forest litter. They found the nitrogen mineralization rates

do not affect initial decomposition rates. However, the chemical composition of litter affected decomposition rates and patterns (McClaugherty et al., 1985). Similar to studies conducted in tropical agroecosystems, high-quality litter based on the carbon to nitrogen ratio is required to increase soil organic matter turnover and improve crop production. In tropical agroecosystems, the nitrogen concentrations and the polyphenol to nitrogen ratios controlled nitrogen release into the soil. Both the findings of Yokobe (2020) and the results from Seneviratne (2000) showed that carbon and nitrogen ratio is the best determinant of nitrogen release on a wide range of residue nitrogen concentrations. This study also ties into the findings of McClaugherty et al. (1985) by demonstrating how critical levels of carbon and plant nutrients act as limiting agents in the enzyme production of microbial decomposers. The two studies found the microbial activity to be essential determinants in nutrient release from plant litter (Seneviratne, 2000).

Litter decomposition plays an essential role in nutrient cycling and ecosystem functions. Therefore, it is crucial to explore the different controlling factors of nitrogen release and immobilization during the different stages of decomposition (Pei et al., 2019). In a study by Pei et al. (2019), litter that initially had higher nitrogen concentrations and lower initial lignin concentrations were found to have a higher gross nitrogen release rate, consistent with the effects on litter mass loss. This demonstrates various chemical compositions and ratios may impact nitrogen release and litter decomposition. Additionally, the initial carbon and nitrogen ratio was observed to be the most crucial determinant of gross nitrogen immobilization rates. The gross nitrogen release was highly regulated by the nitrogen concentration of the litter during the initial stages of decomposition but was later regulated by the lignin concentration. This concept

demonstrates how different litter compositions release nitrogen and highlights macromolecules affecting litter decomposition. Additionally, Pei et al. (2019) concluded that gross nitrogen immobilization was positively correlated to both the nitrogen concentrations during the initial stage of decomposition and the carbon-nitrogen ratio during the later stages. Litter with higher gross nitrogen release rates are a main contributor to the soil nitrogen pool in the litter-soil system.

Increasing the nitrogen pool in the soil from litter decomposition and nutrient mineralization is crucial to maintaining soil fertility and promoting plant growth. Processes that govern the soil microbial activity, such as agricultural management, may significantly alter microbial decomposition rates and, as a result, the amount of carbon and nitrogen in the soil. In a study by Martínez-García et al. (2021), litter carbon loss was found to be higher in soil under organic management compared to conventional management irrespective of litter quality (higher nitrogen release from low carbon to nitrogen ratio litter) and not affected by the agricultural management. However, the soil under organic management had higher concentrations of dissolved organic carbon, mineral nitrogen, and organic nitrogen than in the conventionally managed soil, suggesting stimulated microbial activity and, therefore, litter decomposition. Thus, initial litter quality (namely, the carbon and nitrogen ratio) was the main driver of litter nitrogen release, whereas soil management was the main driver of decomposing carbon loss.

The soil respiration rate can provide insight on how carbon and nitrogen ratios control the gross immobilization and mineralization rates, through the stimulation of microbial activity. As shown in deciduous forests, differences in gross nitrogen immobilization and mineralization rates in soil are related to the soil respiration rate. The

respiration rate showed a direct relationship with cumulative mineralization and immobilization (Bengtsson and Månsson, 2003), which highlights how both mineralization and immobilization physiologically linked to soil respiration.

Conclusions and objectives

The carbon and nitrogen ratio of decomposing plant litter can be an indicator of broader soil health, as the carbon and nitrogen ratio controls microbial activity and the decomposition and release of nutrients within the plant litter. The soil respiration rate is used to track the activity of soil microbes. In agricultural systems, many factors influence the soil microbial activity, including the types of litter, management practices, and time passed since crop establishment. Studies have found high-quality litter can improve soil health as high-quality litter increases the soil organic matter and releases more nutrients than low-quality litter. There is contradicting information involving the effects of macromolecules such as lignin on the decomposition of plant litter. There are also very few studies comparing the effects of vineyard management on the microbial community several years after establishment. This study aims to explore soil fertility in vineyards and the specific effect of contrasting plant litter additions and fungicide applications derived from the viticultural practices. The overall objective of this research was to determine the extent to which plant litter additions from viticulture practices influence carbon and nitrogen mineralization process.

CHAPTER III

Materials and Methods

Vineyard Conditions at Time of Soil Sampling

At the time of soil and plant tissue sampling, the vineyard management practices included one application of the herbicide, Gamoxone, at a rate of 2.5 pints per acre applied under the vine rows. Four applications of fungicides included two applications of the Mancozeb at a rate of 1.5 lb per acre per application, an application of azoxystrobin at a rate of 10 fl oz per acre, and an application of Myclobutanil at a rate of 18 oz per acre. The vineyard had not received any irrigation. The vineyard had also been pruned on June 7, 2021; the vineyard was pruned for dormant and green pruning, which is not consistent with most practices (Syvantek, 2021).

Soil Collection

The soil was collected in August 2021 from the Cimarron Valley Research Station in Perkins, Oklahoma (Lat. 33.996772 N, Long -97.040544 W). According to Web Soil Survey, the vineyard was established on a Teller fine loam and fine sand loam; mixed, active, thermic, ustic haplustalf. The A/E horizon consists of fine loamy sand, and the BT horizon consists of sandy clay loam.

Five kilograms of soil were collected and composited from a depth of 0-30cm using a 1in-diameter soil probe. Before sampling the soil, the above-ground plant litter on the soil surface was removed from the site before sampling. The soil was sampled using a soil probe down to a depth of 0-30cm. After collection, the soil was transported and stored at 4°C before being sieved through a 2mm sieve and analyzed for the total carbon and nitrogen, moisture content, pH, EC, and microbial biomass, prior to incubation.

Soil Analysis Prior to Incubation

Total soil carbon and nitrogen were measured via dry combustion analysis by grinding the soil using an 8000 M Mixer/Mill ball grinder, which was run for 5mins, then placing 200 ± 0.02 mg of the soil samples into tin foil cones prior to analysis on a LECO CN628 series combustion analyzer (Buchanan et al., 2020). The moisture content was determined by weighing out 10 grams of soil, then placing the 10 grams of soil into an oven at 65°C for 24 hours, and then reweighing before subtracting the dry weight of the soil from the original weight of the soil. The pH and EC were found using a 1:1 ratio of soil to nanopure water.

The chloroform fumigation extraction method was used to determine microbial biomass carbon and nitrogen (Vance et al., 1987). The 10 grams of soil were placed into two sets of 100ml sterile specimen containers for the chloroform fumigation process. One set of the 100ml sterile specimen containers were placed into a fumigation chamber with 20ml of chloroform, while the other set was placed into a control desiccator without

chloroform. A vacuum pump fitted with a three-way vacuum line allowing the control desiccator and chloroform desiccator to undergo the same vacuum pressure was used to boil the chloroform. The chloroform boiled vigorously for 1 min then was left to rest for 1 min. After repeating the process three times, the desiccators were capped off, covered, and kept in the dark for 24 hours. After 24 hours, the vacuum was released, and the soil samples were extracted with 50ml of potassium chloride. The potassium chloride was added to the 10 grams of soil in the sterile specimen containers and shaken for 30 min, then let rest. After the rest period, the potassium chloride was filtered from the soil using a number 42 Whatman filter paper. After the filtration process, the filtrate was analyzed for total organic carbon and total nitrogen using a TOC carbon-nitrogen analyzer (Elementar Vario Select, Germany).

Evaluating Water Holding Capacity

Before incubation, the soil was adjusted to 40% of water holding capacity. The soil was then moistened to field capacity by placing 10 grams of soil into a #42 Whatman filter paper inside of a funnel with a sterile specimen container placed at the bottom of the funnel to catch the leachate as it passed through the soil. Then 50ml of water was allowed to pass through the soil. The soil was then let to rest for 24hrs, before the soil was weighed and then placed into an oven at 65 °C for 24 hrs. The field capacity and dry weight were used to determine the amount of water to add to bring the soil to 40% of water holding capacity. The target soil moisture was maintained throughout the

incubation period, and the soil was weighed to determine the need for additional water to maintain the 40% of water holding capacity. After setting the water holding capacity, the soil was preincubated at 24°C for one week.

Carbon and Nitrogen Analysis of Plant Litter

During the preincubation period, the plant material (dormant prunings, green plant material consisting of grapevine leaves and shoots, fruit, wheat, and clover) used for the treatments, which were collected at the same time as the soil, were air-dried on the day of collection and then oven-dried for 24 hours at 65°C before being ground. The plant litter was ground using an 8000 M Mixer/Mill ball grinder (SPEX Sample Prep, Metuchen, New Jersey), which was run for 5 mins. After grinding, the plant litter was packed into tin foil cones at a sample weight of 100±0.02mg. (Buchanan et al., 2020) The tin foil cones were then sent to the Soil Water and Forage Analytical Lab at Oklahoma State University to determine the carbon to nitrogen ratio of the plant material.

Plant Litter and Fungicide Additions

50 gram subsamples of the soil were placed into 568ml wide mouth mason jar with the mason jar lid fitted with a septum for CO_2 sampling. After adding 50g of soil into the mason jars, the jars were then divided among the six plant litter treatments and a fungicide treatment with and without fungicide. The treatments consisted of soil, dormant pruning, green plant material (consisting of shoots and leaves), fruit, and cover crops

wheat and clover, each treatment had 16 replicates of the 16 replicates half received fungicide. The jars were placed in an incubation chamber set at approximately 25°C for 1 week allowing the soil to settle and microbial activity to return to the original rate. During the settling period, the plant litters was cut to a length of approximately 1 cm. Then after the settling period, 5 mg of each plant litter type was added to the soils, with a set of 16 jars with only soil not receiving any litter addition serving as a control treatment. The five plant litter additions and bare soil (16 jars per treatment) were then split in half, with half receiving fungicide and the other half not receiving a fungicide treatment. The fungicide treatments are at rates of 0.03ml/50mg soil using 1ml of deionized water as a carrier agent for the Bonide Mancozeb Flowable with Zinc Concentrate and adding the one ml of deionized water into the moisture calculations to not go over the 40% water holding capacity. These rates were based on the recommended surface area rates for the mancozeb product in vineyards east of the Rocky Mountains and other studies (Tortella et al., 2013; Chen et al., 2001; Baćmaga et al., 2015; Zhang, 2019). After the plant litter and fungicide additions were made the mason jars were place in an incubation chamber set at 24°C.

CO_2 Respiration Measurements

CO_2 measurements were taken from the 568 ml wide mouth mason jar by inserting a needle and syringe into the septum fitted on the lid. The syringe was filled with 5ml of air from the jar the sampled CO_2 was injected into an infrared gas analyzer

system (LI-COR LI-850, Lincoln, Nebraska) for analysis (Kunito et al., 2018; Lavallee et al., 2018; Cordova et al., 2018; Yan et al., 2020). After the sample was run through the infrared gas analyzer, the jar lids were removed to reset the jar back to ambient air. CO_2 measurements were initially taken every 4-12 hours during the first week and then every 19 hours during the 15-day experiment. The experiment was ended at 15 days as this was when the CO_2 measurement began decreasing, and the treatment's CO_2 respiration rate was equal to the control and remained stable for three days.

Microbial Biomass and Total Soil Carbon and Nitrogen Analysis

After the first week, half of the treatments were randomly selected, removed, and analyzed for microbial biomass carbon and nitrogen, total organic carbon and total nitrogen, and total inorganic nitrogen. At the end of the experiment, after the CO_2 had declined and remained stable for three days, the remaining half of the treatments were analyzed for the microbial biomass carbon and nitrogen, total organic carbon, and total nitrogen, total inorganic, total carbon and total nitrogen, and pH/EC as previously described in the preincubation analysis section.

Statistical Analysis

Data analysis was performed in R studio (2022.02.2+485). A two-way ANOVA with a confidence interval of $\alpha=0.05$ was conducted to determine the differences in

nitrate, ammonia, microbial biomass carbon, pH, total soil carbon, and total soil nitrogen among the treatments and time points on day 7 and day 15. A Tukey Post-Hoc analysis was also run in R studio to evaluate differences in treatment means and across time points.

CHAPTER IV

RESULTS

During the study, most of the differences in soil chemistry and microbial activity were largest after the first time point. The initial pH values of the soil had an average of 6. The ending pH of the soil after 15 days of the litter treatments had values ranging from 5.5-7.5, with wheat and clover with fungicide additions having the highest pH values, as shown in Figure 5.

Throughout the experiment, the total soil carbon remained not significantly different, although on day 15, the total soil carbon had a wide range of measures compared to day 7. The total nitrogen in the soil was significantly different and at day 7 had the highest range of measures, with clover and wheat treatments both with and without fungicide having the highest amount of nitrogen while the green plant material, dormant prunings, and fruit had the lowest amount of total nitrogen (Figure 3 and 4). On day 15, the treatments that received fungicide additions had the highest total nitrogen in the soil, as shown in Figure 4.

On day 7, the wheat litter addition treatments had the highest amount of NH_4^+, followed by the clover without fungicide; the rest of the treatments remained significantly different from one another but had a relatively small range of measures (Figure 7). At day 15, all the treatments were not significantly different except for wheat with fungicide, which was significantly higher and almost 3x the amount on day 7 (Figure 6).

On day 7, even though clover without fungicide had one of the lowest amounts of nitrate, the clover with fungicide had the highest amount of nitrate, followed by the wheat treatments, and then soil with fungicide (Figure 7). At day 15, wheat without fungicide had the highest amount of nitrate followed by wheat with fungicide and clover without fungicide (Figure 8). The nitrate concentration in the wheat without fungicide treatment almost doubled from day 7 to day 15, while the wheat with fungicide stayed relatively the same.

While there were significant differences in the amount of microbial biomass on day 7 (Figure 9), there was no significant difference on day 15 (figure 10). On day 7, the fruit without fungicide, soil, and clover with fungicide had the highest microbial biomass, but only the fruit without fungicide was significantly different from the other treatments.

The fruit without fungicide had a higher initial respiration rate for approximately 100 hours before having a significant decrease, while the fruit with fungicide had a lower initial respiration rate. The fruit with fungicide had a higher and more consistent respiration rate. Additionally, the wheat with fungicide had a higher initial respiration rate but decreased quickly and was approximately the same as the wheat without fungicide after the first 100 hours (Figure 11).

The cumulative CO_2 was higher in the fruit treatments than in the other litter treatments on days 7 and 15 (Figures 12 and 13). The cumulative CO_2 matched the carbon to nitrogen ratio of the litter as the higher quality litter treatments clover and wheat had the higher cumulative CO_2, respectively than the other litter treatments green plant material, dormant prunings, and the control soil.

Percent of carbon and nitrogen in soil and litter additions and amount of carbon added to soil.

Litter Treatments	% Nitrogen	% Carbon	Litter C/N ratio	mg carbon added per gram of litter addition per gram of soil	mg carbon in fungicide addition per gram of soil	mg carbon added per litter addition with fungicide per gram of soil
Wheat	3.239	39.727	12.2643	0.00794	0.021	0.02894
Clover	2.351	39.988	17.011	0.00799	0.021	0.02899
Fruit	1.588	42.241	21.602	0.00844	0.021	0.02944
Dormant Prunings	1.919	41.462	62.918	0.0008292	0.021	0.02929
Green Plant Material (Shoots and leaves combined)	0.682	42.927	26.593	0.00858	0.021	0.02958
Soil	0.125	1.7	13.627	0	0.021	0.021

Table 1. Percent carbon and nitrogen found in litter treatments, the carbon to nitrogen ratio of the litter, the amount of carbon added per 5 grams of litter treatments with and without fungicide, and the amount of carbon added per fungicide treatment.

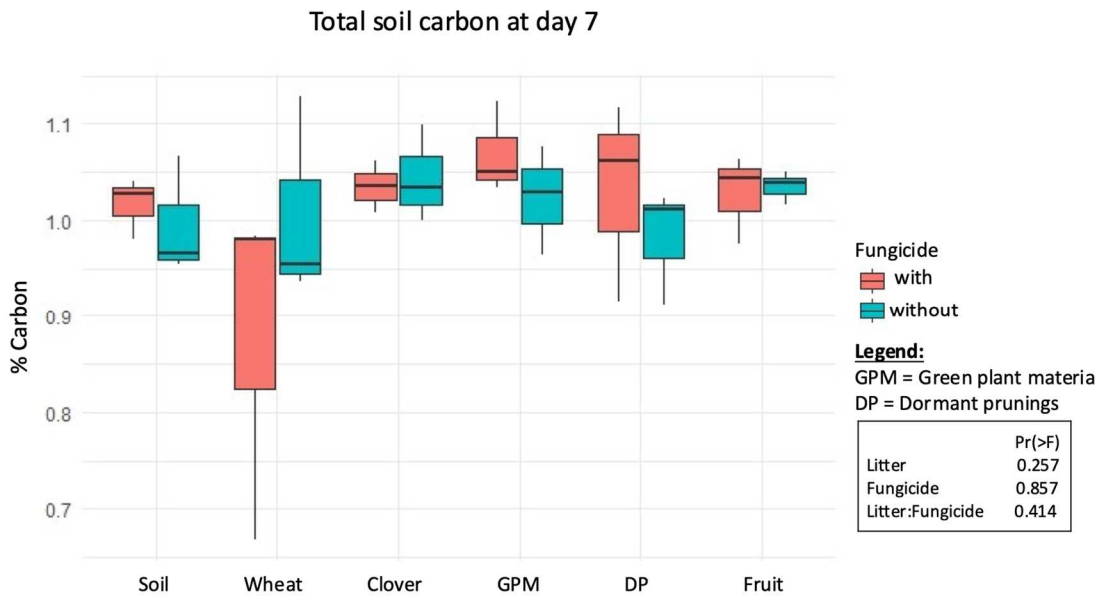

Fig. 1. Total soil carbon at day 7 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

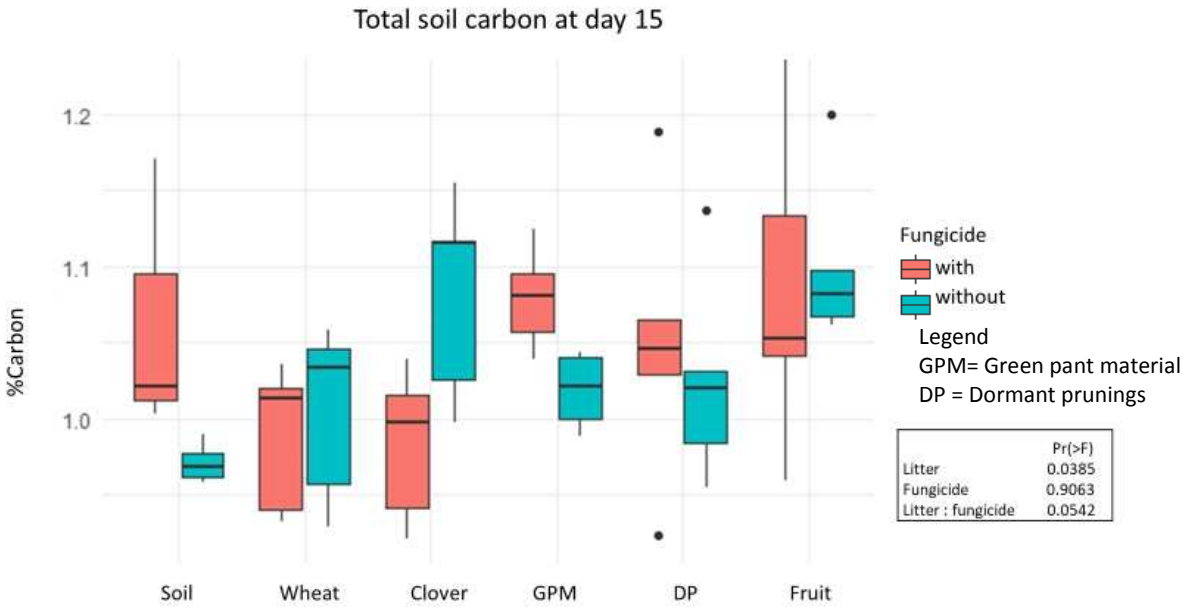

Fig. 2. Total soil carbon at day 15 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

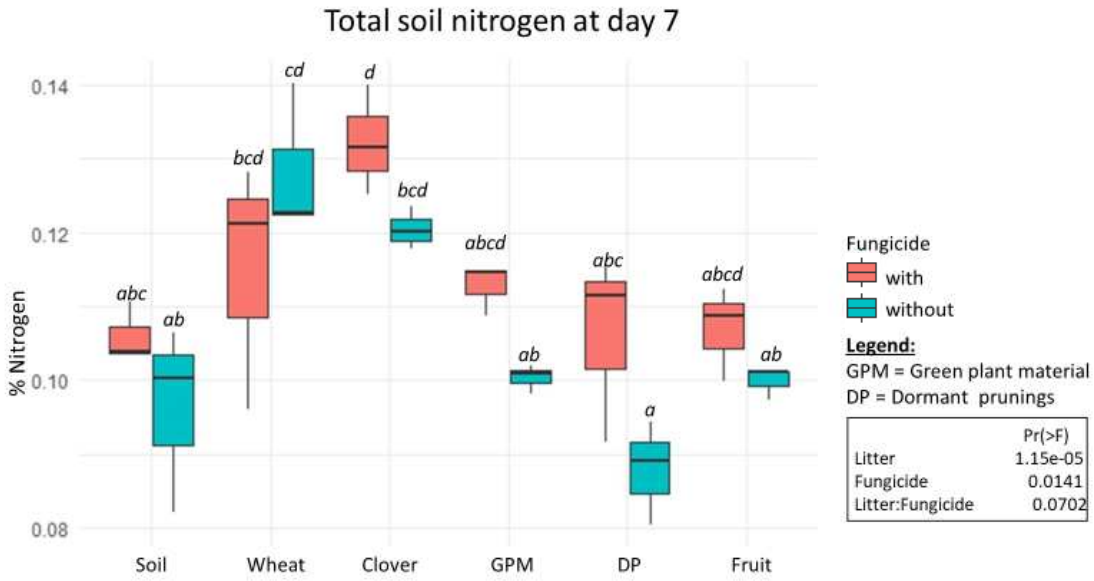

Fig. 3. Total soil nitrogen at day 7 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test on $P < 0.05$. Error bars indicate the standard error of averaged samples.

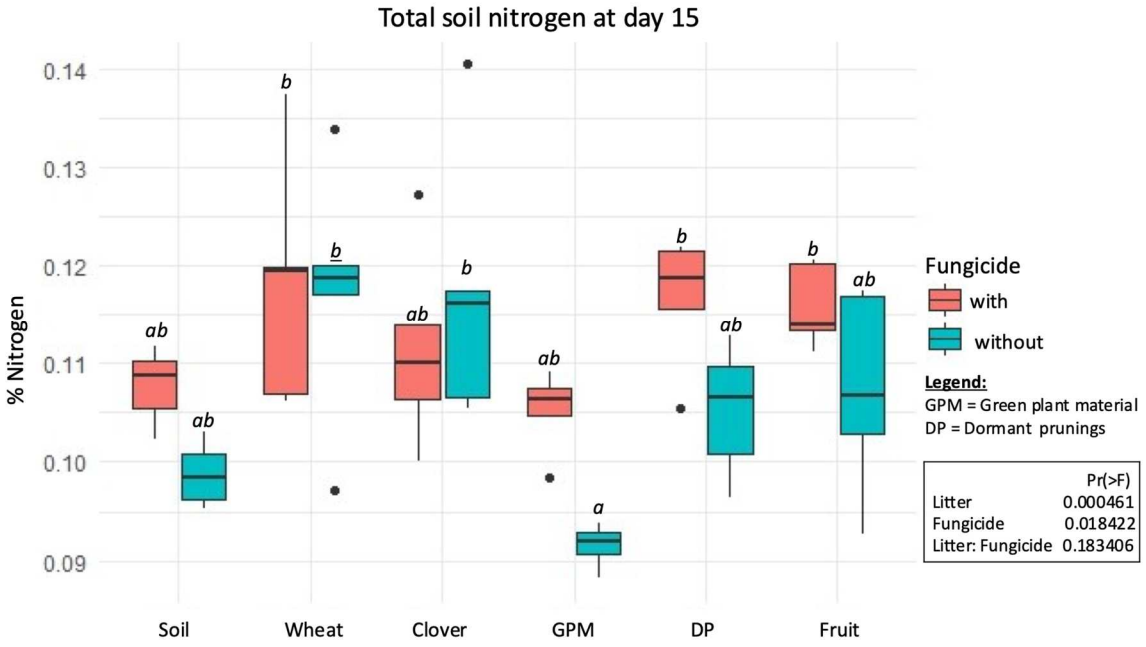

Fig. 4. Total soil nitrogen at day 15 litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

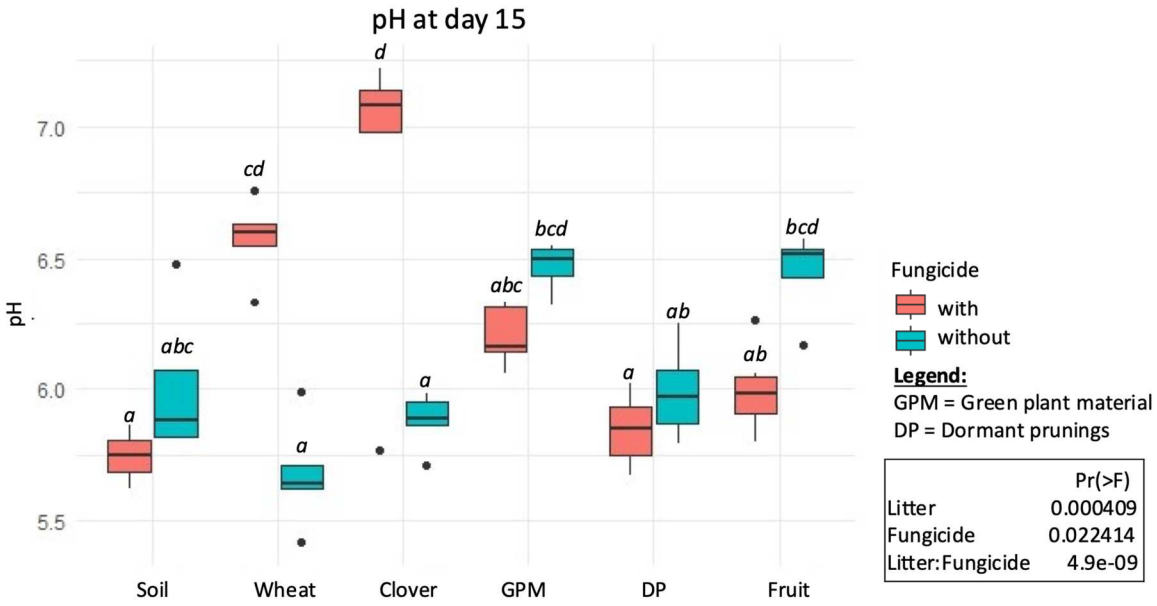

Fig 5. pH of soil at day 15 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

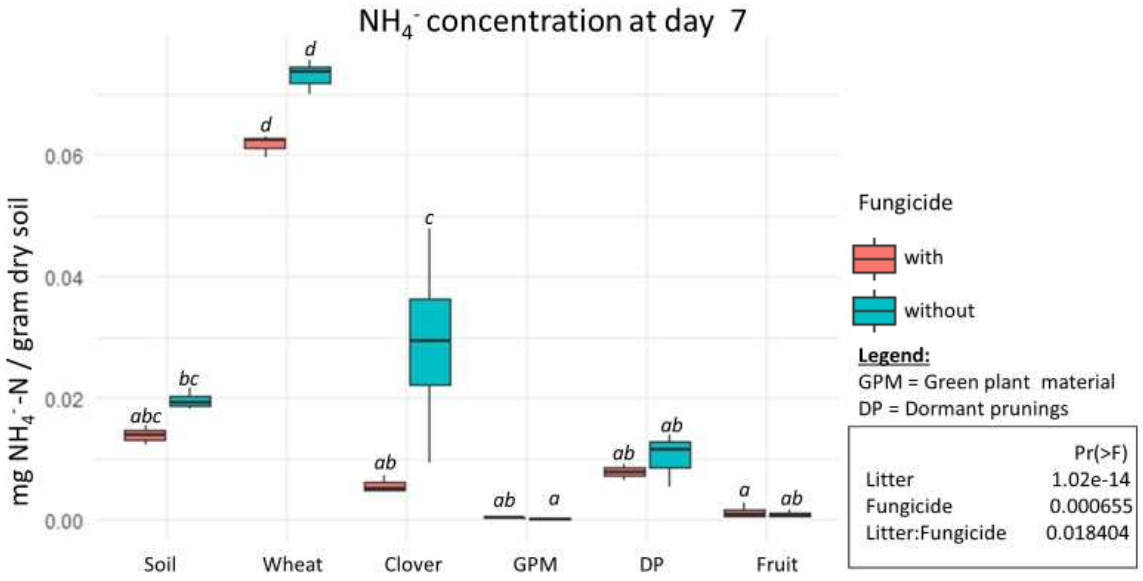

Fig 6. NH_4^+ concentration of soil at day 7 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

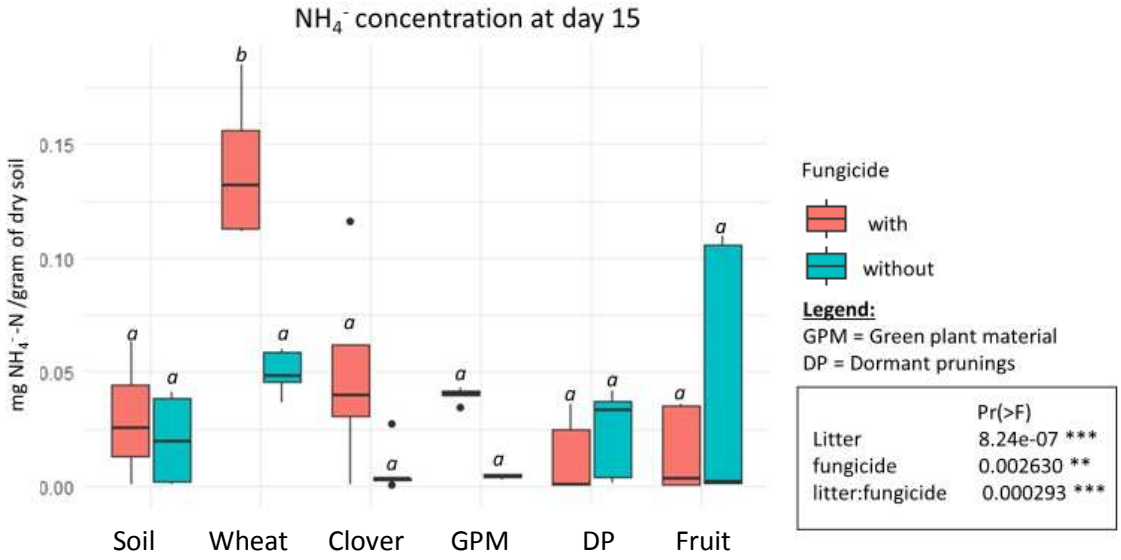

Fig 7. NH_4^+ concentration of soil at day 21 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

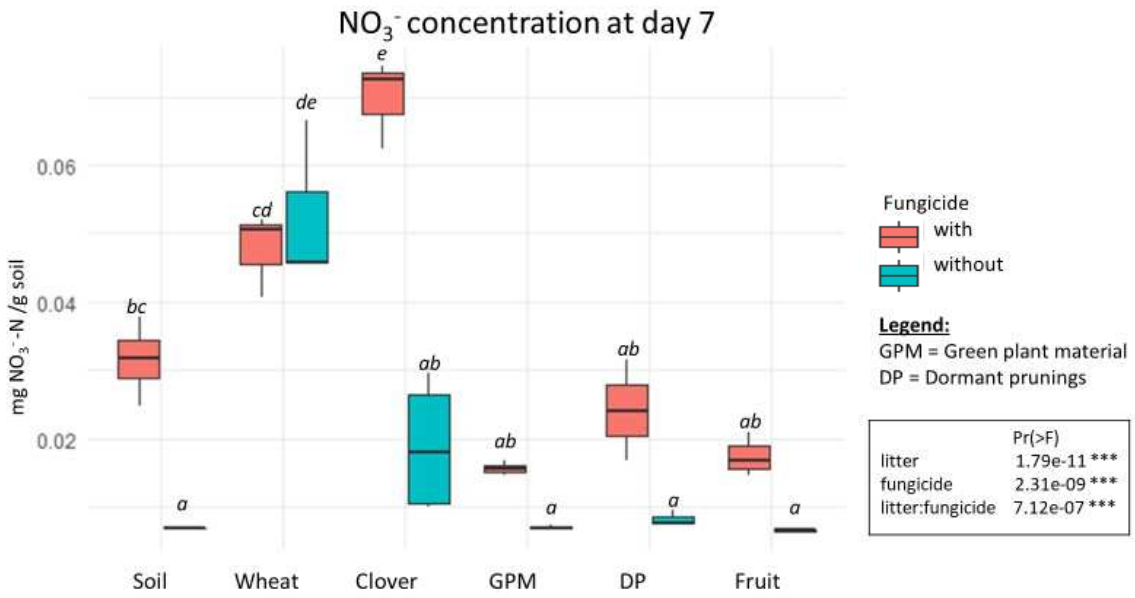

Fig 8. NO_3^- concentration of soil at day 7 based on litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

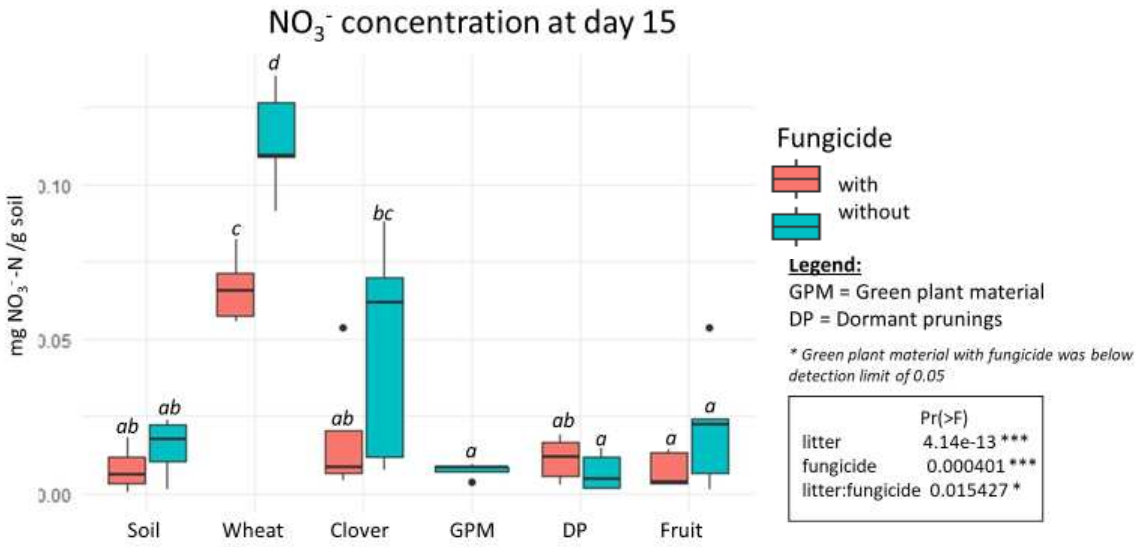

Fig 9. NO_3^- concentration of soil at day 15 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

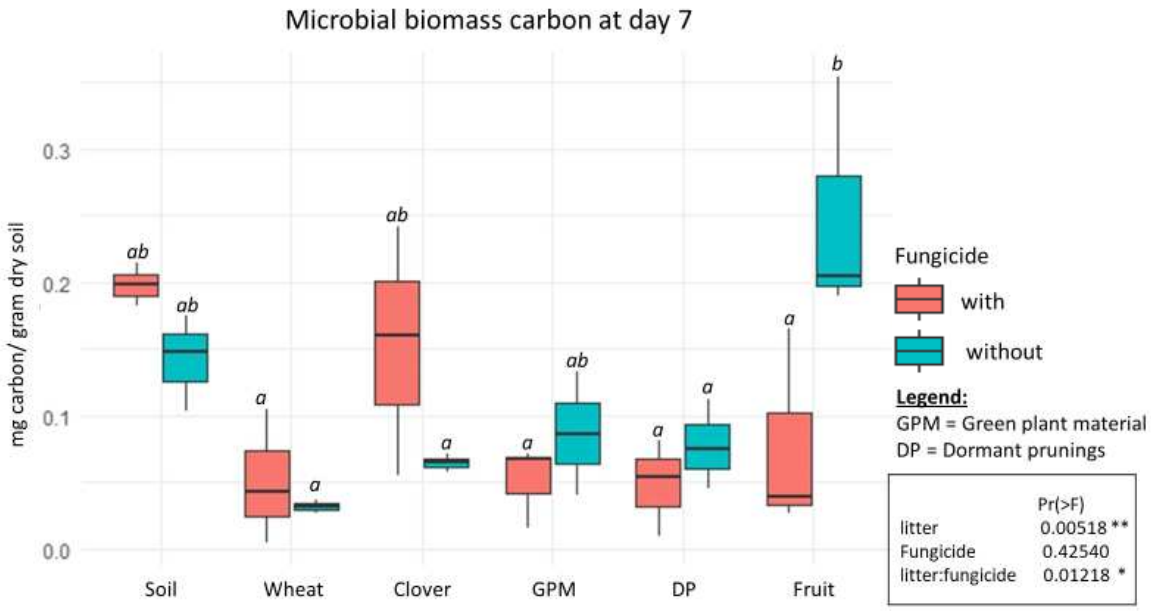

Fig 10. Microbial biomass is present in the soil on day 7 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

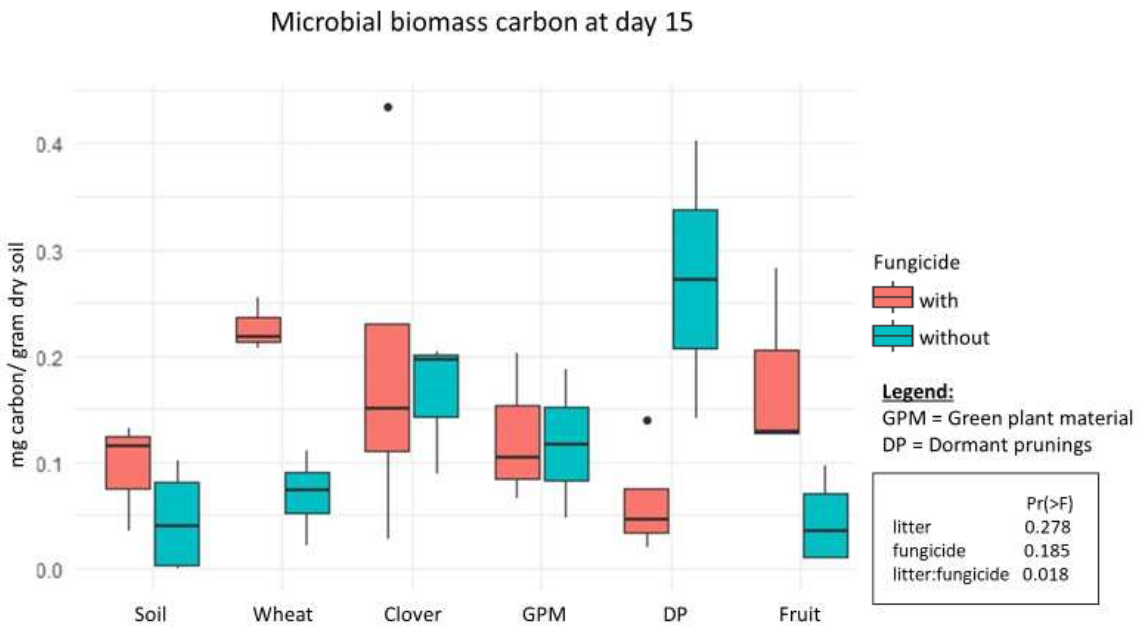

Fig 11. Microbial biomass is present in the soil at day 15 in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples

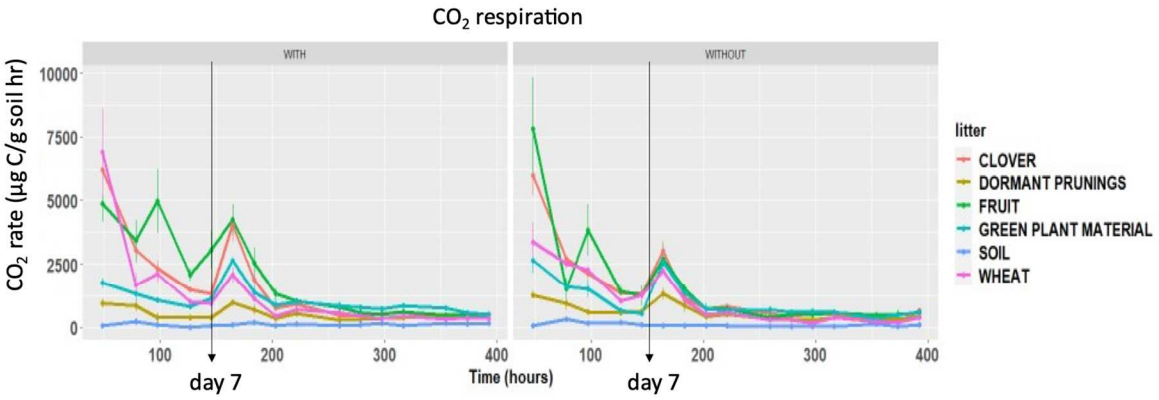

Fig 12. Soil respiration rate over 400 hours in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

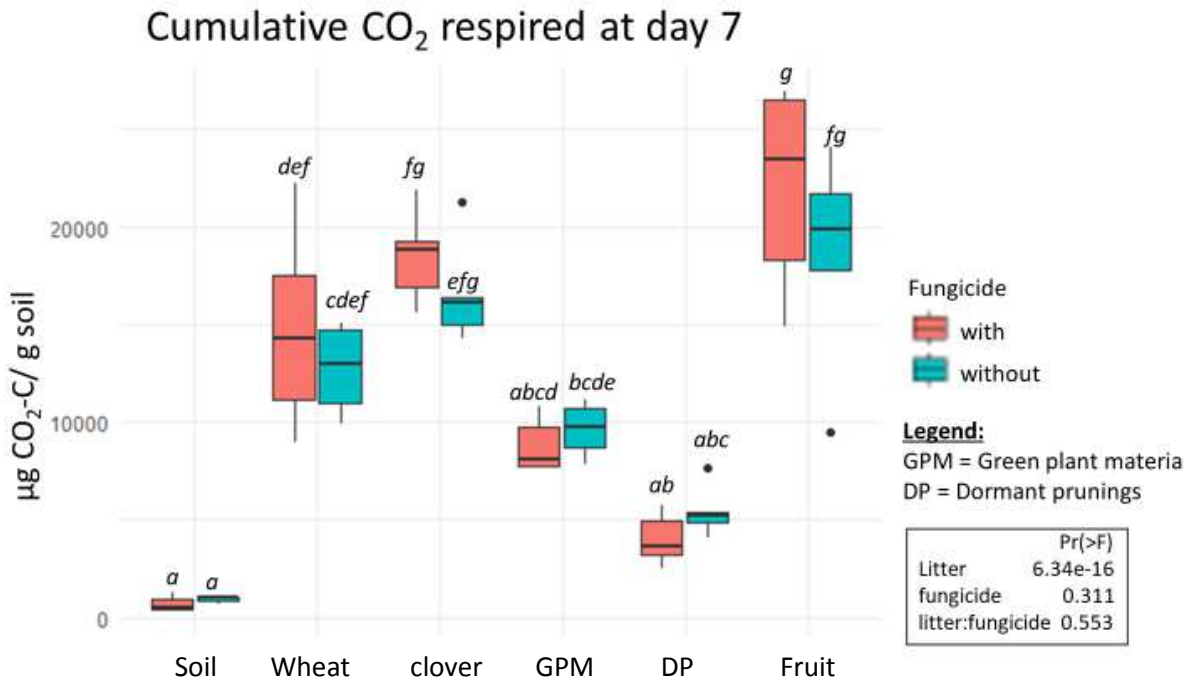

Fig 13. Cumulative soil respiration rate over 165 hours in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

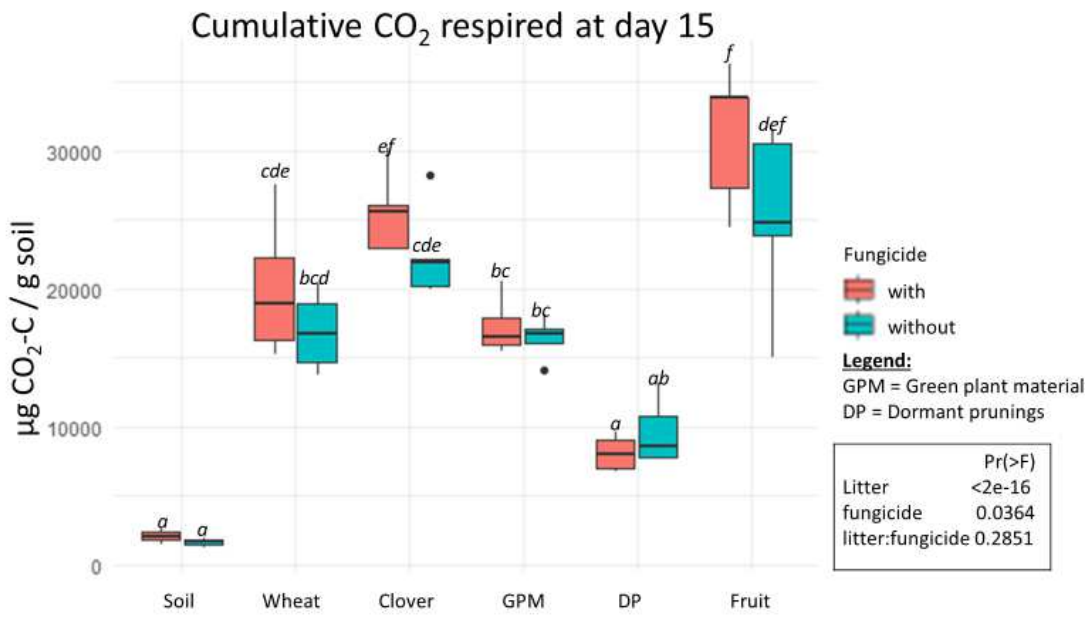

Fig 14. Cumulative soil respiration rate over 400 hours in response to litter addition treatments (wheat, clover, green plant material, dormant prunings, and fruit) with and without mancozeb fungicide treatment. The letters in italics indicate significantly different means according to the Tukey test at $P < 0.05$. Error bars indicate the standard error of averaged samples.

CHAPTER V

Discussion

The study aimed to evaluate how the litter additions from viticulture practices affect soil fertility and health. A specific goal was to evaluate how the litter additions from the viticulture practices influence the inorganic and organic nitrogen in the soil by stimulating microbial activity as tracked by soil respiration and biomass carbon.

In the total nitrogen analysis, on both day 7 and day 15, the fungicide treatments had higher nitrogen present as a result of the nitrogen found in the fungicide being added to the soil.

In the ammonium at day 7, the fungicide may have killed the microbes converting the ammonium into other forms of nitrogen, thus lowering the results when compared to the treatments that did not receive fungicide. On day 15, the fungicide had worn off, and the microbes could recover and convert more nitrogen into ammonium, especially in the high-quality litter, wheat, treatments. These findings are similar findings by Chen, Edwards & Subler (2001) in which they showed adding fungicide to soil influences the microbial activity of the soil and nitrogen dynamics of the soil.

In the nitrate analysis, the fungicide treatments may have caused the nitrogen to be readily nitrified, thus having a higher nitrate concentration than ammonium concentration. Additionally, in the nitrate analysis from day 7 to day 15, the increase may be due to decomposing litter releasing nitrogen into the soil.

Microbial biomass carbon at day 7 was lower with the addition of the fungicide, showing the fungicide suppressed microbial growth. In comparison, the higher microbial biomass carbon at day 7 on the fruit may have resulted from the fruit having a native population of microbes living on the fruit compared to the other litter additions. On day 15, the microbial populations across the treatments were not significantly different; this may be due to the decomposition of the litter and fungicide effect wearing off, allowing the microbial activity to return to the regular rate. These findings correspond with the results from Wainwright and Pugh (1973) which showed at low concentrations fungicide additions stimulated nitrification and ammonification. The microbial population living on the fruit may have also caused an initial higher CO_2 respiration rate when compared to the fruit which received the fungicide additions. Although, low-quality litter fruit treatment has a lower inorganic nitrogen concentration, this treatment has a higher CO_2 indicating the nitrogen is immobilized by the microbes.

The cumulative CO_2 across the treatments matched the carbon to nitrogen ratio except for the fruit, which may have resulted from the native population of microbes respiring and fermenting the decaying fruit. The cumulative CO_2 was correlated with the carbon to nitrogen ratio of the litter, similar to findings by Sall et al. (2003).

A limitation of the study was the over CO_2 accumulation in the mason jars during the incubation period, this resulted in an oversaturation of the infrared gas analyzer and inaccurate readings for some over the treatment replicates.

The differences in biological measurements, especially CO_2, were most apparent in the first seven days, meaning the microbes responded more strongly to reside additions within the first few days after the additions. While this study only added a small amount of litter residue

once, in reality, litter inputs occur continuously throughout the growing season; additionally, the amount of litter added may be significantly higher than the amount added during the study. Also, the introduction of plant litter into the soil may overlap, whereas in this study, the plant litter was isolated. However, this study highlights the impacts each single litter additions have on the nitrogen cycle and the microbial community. The effects may be short-lived and repeated additions at critical growing points may optimize plant growth; however, further research needs to be conducted. Furthermore, in vineyard systems, the temperature and moisture fluctuate through the day and year. Although the study controlled the temperature and moisture conditions for optimal microbial growth, the study demonstrates, under optimal microbial growing conditions, the influence of litter additions on the nitrogen cycle.

CHAPTER VI

Global Impact Statement

Soil fertility is a primary concern in agricultural systems of both developing and developed countries. A common way to increase soil fertility is using synthetic fertilizers. However, in areas with limited access to synthetic fertilizers, targeted plant residue management can provide alternative inputs of nutrients that can promote soil fertility and health (Bai et al., 2022). Since residue management differs across ecosystems and various economic contexts there may be limitations to how residue management can replace fertilizer inputs. However, this study aids in demonstrating the potential of using residue additions as fertilizer sources for areas that may not have access to synthetic fertilizers. However, future studies addressing how residue management improves soil fertility and the economic tradeoff using residues are needed to show which regions benefit economically from using residues vs. synthetic organic fertilizers.

Additionally, other studies that more closely monitor and quantify residue inputs and subsequent changes in nitrogen and nutrient cycling over time are needed since the nitrogen availability, and nitrogen demand of the plants drastically change over time and across agricultural systems. Additionally, this study solely focused on nitrogen as the critical nutrient source; however, other macro and micro-nutrients found in plant litter may be released, thus improving soil fertility. For this reason, producers may be better off focusing on a particular time point in the season or a specific type of residue instead of managing and retaining all possible residues. However, there may be other benefits to retaining and managing residues. For instance, retaining and managing residues in vineyards may increase microbial community diversity and

potentially increase resistance to pathogens. Additionally, the fruit quality and quantity should be addressed as changes in soil fertility, soil microbial diversity, and increasing pathogen resistance may alter the fruit quality and quantity.

CHAPTER VII

Conclusion

It is important to understand the influence of viticulture canopy management tactics on soil nitrogen mineralization and immobilization dynamics. This study helps shed light on how different qualities of litter affect the microbial cycling of carbon and nitrogen. This information will help aid in future decision-making of the application and timing of fertilizers, ensuring the fertilization rates and timing correspond to the microbial activity increasing plant uptake of nutrients. In vineyard systems, there are several time points in which litter additions and fungicide applications coincide with critical plant growing points. By understanding how litter additions and fungicide application influence microbial activity and the nitrate and ammonium concentrations in the soil, vineyard managers can adjust their fertilization programs while optimizing plant growth. It is well-recognized that microbial decomposition influences nitrogen mineralization and plant-available nutrients in the soil. For future studies, more attention should be focused on how these practices influence the other macro and micronutrients in the soil and the grape berry composition. Other areas for future research include exploring plant litter additions with various plant types of varying quality across contrasting soil types. In addition, future research should conduct cost to benefit analysis of using plant litter additions as a substitute for synthetic fertilizers.

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