AN ABSTRACT OF THE DISSERTATION OF

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Title: Patterns of Carbon Storage Within a Mature Northern Highbush Blueberry Production System

Abstract Approved:

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The objectives of this study were to: 1) determine how organic matter (incorporated vs. surface mulch) and nitrogen fertilization rate impact northern highbush blueberry (*Vaccinium corymbosum L.)* plant biomass, carbon accumulation, plant losses and allocation, and mycorrhizal infection in mature plants, and 2) determine the magnitude of carbon fluxes (carbon net primary production (NPP), soil respiration, and fruit and pruning exports) and stocks within a blueberry production system, and how these are affected by typical management practices. Treatments were in effect for nine years since planting establishment; here we report on data collected in 2011 and 2012. Many of these treatments seem to have short- and long-term effects on blueberry plants. Long-term effects included the impact of pre-planting incorporation of sawdust, which as a main effect, had an overall positive effect on yield, and soil fertility, with all soil nutrients being above recommended sufficiency levels for blueberry production. Soil pH was increased by incorporation, and was affected by an incorporation by mulch interaction where incorporated bare plots had the highest pH, and the largest average plant dry weight and carbon (C) mass (3.5 and 1.7 kg/plant, respectively) despite the pH being above the recommended level for blueberry production. Incorporated plots in general, had a higher total field C stock averaging 97.6 t·ha^{-1} for mulched plots and 93.7 t·ha^{-1} for bare plots. Mulching as a C stock contributed 12.3 t·ha^{-1}, 13% of the total C stock. Mulching as main treatment effect was not found to be beneficial in terms of increasing plant and soil C stocks. Although mulching did increase soil organic C in 2012, this did not seem to affect total soil C stocks, perhaps because soil respiration was also increased by the mulch. Nitrogen fertilizer rate did not affect plant biomass or C stock, nor did it affect soil C stocks and nutrients. Net primary productivity averaged 588 g·m^{-2}.year and was not affected by the treatments, although incorporated plots had about 25% more NPP than non-incorporated plots. Our results have illustrated that with a goal of optimizing plant growth, yield, and C stocks, blueberry production systems that include pre-plant incorporation of organic matter without addition of surface mulch and moderate rates of nitrogen fertilizer are best. In addition, a between-row perennial grass cover crop is recommended to increase field C stocks and to limit soil erosion. The information gathered in this study can be used to estimate the contribution of C storage in temperate perennial crops to global C stocks. Recommended management practices could lead to a policy system where farmers receive incentives for sustainable low C agriculture.

Patterns of Carbon Storage Within a Mature Northern Highbush Blueberry Production System

by

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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

Patterns of Carbon Storage Within a Mature Northern Highbush Blueberry Production System

Chapter 1: Introduction

Food systems contribute 19–29% of global anthropogenic greenhouse gas (GHG) emissions, the majority of which are a direct consequence of agricultural production practices such as land cover change, the use of fossil fuels, crop burning, and tillage (Fraser et al., 2008; Girgenti, et al., 2013; Norse, 2012; Vermeulen et al. 2012). Carbon (C) sequestration, abiotic and biotic, can offset emissions in the atmosphere providing effective mitigation to climate change (Heath et al., 1996; Lal 2008; Lal et al. 2003; Marland et al., 2001, 2003). Research on terrestrial C sequestration has been primarily focused on forest and grassland ecosystems (Keightley, 2011; Testi et al 2008). When the focus is agriculture, most studies look at the components of "low C agriculture" which include soil C sequestration, the reduction of GHG emissions and annual crop C storage (Jia et al. 2012; Lal, et al., 1998; Morgan et al. 2010; Norse, 2012; Paustian et al. 1995, 1997; Reicosky et al. 1997).

Conventional annual cropping systems have been widely studied in terms of C storage in plants and soils (Witt et al., 2000; Zan et al, 2001). In contrast we know very little about the degree to which perennial woody cropping systems contribute to GHG emissions and how much C is stored in the fields. Woody perennial cropping systems have the potential of storing C because of their low soil disturbance and their ability to store C within the woody biomass of plants, in resistant organic polymeric carbon compounds such lignin (Horwath, 2007; Kroodsma and Field, 2006). Knowing more about these systems would allow us to improve the accuracy of estimates of the agricultural contribution to GHG emissions, help us develop management recommendations specific to perennial systems, and enables us to evaluate the degree to which perennial production should be incentivized or rewarded as a form of C storage.

Northern highbush blueberry (*Vaccinium corymbosum L.)* production systems are a good example of a perennial crop with potential for C storage in both plant and soil components. Blueberries are an important crop, with global blueberry production reaching 376,400 tonnes in 2010 from a planting area of 77, 288 hectares, with an average area growth increase of 7,058 ha/year since 2005. Of total world blueberry area, 58% is located in North America, where 44,633 hectares (245,650 tonnes) are dedicated to highbush blueberry production (65% of global production), (Bañados et al., 2012; Brazelton, 2011). A number of factors suggest that blueberry systems could provide a net C store: 1) blueberry plants live for many years only reaching maturity eight years after planting, and mature fields can be over 35 years old; 2) blueberry production systems commonly incorporate perennial cover crops between rows; 3) there is typically no tillage after fields are established; and 4) there is crop residue from annual prunings left within fields and from the annual senescence of leaves (Scagel and Yang 2005; Strik, 2006; White, 2006; Yang 2002b). Some aspects of blueberry production contribute to biomass and C losses from the system and from the plant, including the fruit removed at harvest, prunings and senescing leaves.

In blueberry production systems, organic matter is often incorporated into the soil prior to planting to increase soil organic matter content and maintain the appropriate pH level, around 4.5 to 5.5, for optimal blueberry growth (Hayes and Swift, 1986; Strik et al. 1993). However, the overall efficacy of organic matter incorporation and its effects on plant growth and allocation are not well understood. The use of surface mulch also appears to have complex and context dependent effects on blueberry growth. Surface mulches, like sawdust, have been shown to increase plant growth and yield in highbush and rabbiteye blueberry relative to bare soil (Chandler and Mason 1942; Clark, 1991; Haynes and Swift, 1986; Kozinski, 2006; Lareau, 1989; Magee and Spiers, 1995; Moore, 1979; White, 2006; Yadong et al., 2006).

Organic matter inputs combined with nitrogen (N) fertilization can potentially interact to drive both the patterns of N available to the plant and rates of organic matter decomposition (Khan et al. 2007; Paustian et al 1992). Recommended rates of N fertilization for blueberry vary by production region and site, but plants respond best to moderate application rates ranging from 25−100 $\text{kg}\cdot\text{ha}^{-1}$ of N per year for optimum growth and production (Bañados et al., 2012; Hart et al., 2006; Pritts et al., 1992). The addition of organic matter and N fertilization could also influence the abundance of Ericoid mycorrhizal fungi (EMF) fungi and this could be one mechanism driving blueberry plant response to both management practices. Ericoid mycorrhizal fungi are prevalent in commercial blueberry fields with an average of 22% of total blueberry root length colonized by the fungi, ranging from 0.5 to 44% infection (Scagel and Yang, 2005).

The objectives of this study were to: 1) determine how organic matter (incorporated vs. surface mulch) and N fertilization rate impact plant biomass, C accumulation, plant losses and allocation, and mycorrhizal infection in mature blueberry plants; and 2) determine the magnitude of C fluxes (C NPP, soil respiration, fruit and pruning exports) and stocks within a typical blueberry production system, and how these are affected by management practices.

Chapter 2: Effect of organic matter (incorporated vs. surface mulch) and nitrogen fertilization rate on plant biomass and allocation, carbon mass and mycorrhizal infection in mature blueberry plants

Abstract

The effects of pre-plant incorporation with sawdust, sawdust mulch, and N fertilization rate on plant biomass, C storage, and fruit production in mature 'Elliott' northern highbush blueberry plants was studied. Treatments were in effect for nine years since planting establishment; here we report on data collected in 2011 and 2012 (eighth and ninth year, respectively). Weeds were managed in all treatment plots. Many of these treatments seem to have both long-term and short-term effects on mature blueberry plants. Long-term effects include the pre-planting incorporation of sawdust, which as a main effect, had an overall positive effect on yield and on soil nutrition, with all soil nutrients being above recommended sufficiency levels for blueberry production. Incorporation and an incorporation by mulch interaction increased soil pH to above the recommended level for blueberry production. However, the largest plants with highest plant biomass and C storage were also found in incorporated, bare soils. Organic matter addition was beneficial for plant biomass and C production; however, too much organic matter addition, with both pre-planting incorporation and sawdust mulch, led to the second lowest values for dry weight and C mass. The results of this study suggest that specifically adding fresh sawdust mulch in established plantings has short-term effects on plant biomass and C allocation. The lack of a significant response to N fertilizer rate over this long-term study appears to be related to the ability of blueberries to obtain sufficient nutrients from various sources (e.g. fertilization, mineralization of organic materials, the

soil, and through association with mycorrhizae). The lack of response to N fertilizer may have been related to the amount of available N in the soil pool, which was not significantly affected by fertilization but was decreased by the addition of sawdust mulch. This might explain why the detrimental effect of mulching on nutrient absorbing fibrous root biomass, and mycorrhizal infection did not impact total plant growth, biomass or yield. Because soil nutrient pools were found to be above the recommended level for blueberry production, the plants were getting enough nutrients.

Introduction

Northern highbush blueberry (*Vaccinium corymbosum L.)* is an understory, deciduous, woody perennial shrub that is native to eastern North America. Commercial cultivation of this species began in the early 1900s, and it is now grown worldwide (Strik, 2006; U.S. Highbush Blueberry Council, un-published). The most common native habitats of northern highbush blueberry include marshes, oak and pinewoods, rocky hillsides, swamps, and moist to wet peats (Cain and Eck 1966; Nesom and Davis, 2002). Blueberry plants grow naturally in soils with a high organic matter content, low pH, good porosity, and low fertility (Hanson et al., 2002;Hayes and Swift, 1986; Yadong et al., 2006). In soils adapted to blueberries, most of the plant available inorganic nitrogen (N) is in the ammonium form (NH_4^+) because low soil pH inhibits microbial nitrification (Dancer et al., 1973; Hanson et al., 2002). Blueberry plants predominantly take up ammonium-N rather than nitrate-N (Goulart et al., 1995, 1997, 1998; Hanson 2006; Haynes and Swift, 1986; Merhaut and Darnell 1996; Peterson et al., 1988; Spiers 1986).

Commercial blueberry production systems often include cultural practices that enhance the suitability of the soil to improve blueberry plant growth, including incorporation of organic matter prior to planting, raised beds to improve soil drainage, and fertilization with ammonium-N at optimal rates (Hanson et al., 2002; Hayes and Swift, 1986; Spiers, 1986).

In blueberry production systems, organic matter is often incorporated into the soil prior to planting to increase soil organic matter (OM) content and maintain the appropriate pH level, around 4.5 to 5.5, for optimal blueberry growth (Hayes and Swift, 1986; Strik et al. 1993). However, the overall efficacy of organic matter incorporation and its effects on plant growth and allocation are not well understood. The reported effects of organic soil amendment on blueberry production have been inconsistent, with some studies reporting positive effects on plant growth and yield (Haynes and Swift, 1986; Moore, 1979; Spiers, 1982), while others reporting negative effects (Townsend, 1973a; White, 2006). Different types of organic soil amendments and their effects on blueberry production have also been extensively studied, including: peatmoss (Haynes and Swift, 1986; Lareau, 1989; Spiers 1982; Townsend, 1973a), bark (Bollen and Gleanie, 1961; Haynes and Swift, 1986; Odneal and Kaps, 1990; Spiers 1982) and sawdust (Lareau, 1989; Spiers, 1982; Townsend; 1973; White, 2006). The specific effects of amendment with sawdust compared with other types of amendments have also been inconsistent. Spiers (1982) found that peatmoss increased plant growth of young rabbiteye blueberry (*V. virgatum* Aiton syn. *V. ashei* Reade) compared to sawdust amendment. Increasing rates of sawdust amendment decreased growth and yield (Moore, 1979), whereas the opposite was found when increasing rate of amendment with peatmoss (Spiers, 1982). In contrast, other studies have found that sawdust amendment increased plant growth and yield compared to amendment with peatmoss (Lareau, 1989; Townsend; 1973a).

In blueberry, surface mulches can be used to improve the water-holding capacity of the soil or reduce irrigation requirement (Sinkevičienė, et al., 2009; Spiers, 1986), insulate roots from high temperatures and help control weeds (Atucha et al., 2011; Korcak, 1988; Krewer et al., 2009; Moore, 1990; Savage and Darrow, 1942; Shutak and Christopher, 1952; Sinkevičienė, et al., 2009; White, 2006). However, similar to organic matter incorporation, the use of surface mulch appears to have complex and context dependent effects on blueberry growth. Surface mulches, such as sawdust, have been shown to increase plant growth and yield in highbush and rabbiteye blueberry relative to bare soil (Chandler and Mason 1942; Clark, 1991; Haynes and Swift, 1986; Kozinski, 2006; Lareau, 1989; Magee and Spiers, 1995; Moore, 1979; White, 2006; Yadong et al., 2006). In rabbiteye blueberry growing in fine, sandy soil, mulching the soil surface increased plant height, and shoot and root weight (Patten et al., 1988). In some studies, however, weeds were not controlled in un-mulched, control plots, when comparing to those mulched with sawdust, thus confounding the effects of mulch on plant growth with the negative effects of the weeds (e.g. Krewer et al., 2009; Sciarappa et al., 2008). The effect of surface mulches on root weight and distribution have been varied, with an overall conclusion that mulch can be used to improve root distribution (Goulart et al., 1997; Korcak, 1988; Patten et al., 1988; Spiers, 1986, 2000). These studies, however, were done in establishing fields and the assessment of treatment effects on root distribution and biomass was not provided for different types of roots (e.g. fibrous and storage).

Organic matter inputs combined with N fertilization can potentially interact to drive both the patterns of N available to the plant and rates of organic matter decomposition (Khan et al. 2007; Paustian et al 1992). Recommended rates of N fertilization for blueberry vary by production region and site, but plants respond best to moderate application rates ranging from 25−100 kg·ha^{-1} of N per year for optimum growth and production (Bañados et al., 2012; Hart et al., 2006; Pritts et al., 1992). Higher N fertilization rates are often observed in the industry, depending on pre-plant amendments, mulch used, cultivar, and grower preference (Strik, personal observation; Yang, 2002b). The effects of N fertilizer rate on blueberry production have been varied, with some studies reporting positive effects of increased fertilizer N on plant growth and yield (Hanson and Retamales, 1992), while others either reported no effect (Lareau, 1989; White, 2006), or a positive effect only at low levels (Bañados et al., 2012; Clark et al. 1998; Cummings 1978; Spiers, 1982). Mulched sites have a higher soil C to N ratio (C/N) due to the increased organic matter contribution of carbon. This may require growers to have to apply higher rates of N fertilizer (Hart et al., 2006; Kozinski, 2006) as sawdust mulch has been shown to immobilize some fertilizer N (White, 2006).

The addition of organic matter and N fertilization could also influence the abundance of ericoid mycorrhizal fungi (EMF) fungi and this could be one mechanism driving blueberry plant response to both management practices. Ericoid mycorrhizal fungi are prevalent in commercial blueberry fields with an average of 22% of total blueberry root length colonized by the fungi, ranging from 0.5 to 44% infection (Scagel and Yang, 2005). EMFs are symbionts that infect roots, and utilize the host plant's photosynthates as their C and carbohydrate source, while the fungus increases the ability of the plant to forage nutrients (Cheng et al., 2012; Goulart et al., 1997). Mycorrhizal plants are better able to utilize non-ammonium inorganic N, because the mycorrhiza is able to assimilate peptides, amino acids, and proteins from the soil and transfer the N to the plant (Goulart et al., 1997; Yang et al., 2002a). The addition of organic matter and the use of N fertilizer in the blueberry production system have been shown to have varying impacts on mycorrhizal infection, in many cases with interactions between treatments, mainly because of differences in soil pH, cultivar, plant age, stage of growth, organic matter source, etc. (Boyer et al., 1982; Goulart et al., 1993; Jacobs et al., 1982; Scagel and Yang, 2005; Stevens et al., 1996; Yadong et al., 2006; Yang et al., 2002). Most studies have focused on the establishment years or on relatively young plants. Because commercial blueberry plantings can be productive for over 40 years, the long term impact of common production systems on mycorrhizal infection is of interest.

The objective of this study was to determine how organic matter (incorporated vs. surface mulch) and N fertilization rate, impact plant biomass, C accumulation, plant losses and allocation, and mycorrhizal infection in mature blueberry plants.

Materials and Methods

Experimental site. This study was done in an 'Elliott' northern highbush blueberry (*Vaccinium corymbosum* L.) field established in Oct. 2003 at the North Willamette Research and Extension Center (NWREC; 45°16' 47.55” N and 122°45' 21.90” W), Aurora, OR. Standard sized (3.8 L; 2-year-old) container-grown plants were obtained from a commercial nursery just prior to planting. The data reported here were collected in 2011 and 2012, when the planting was eight- and nine-years-old and was considered mature. Soil at the site was a Willamette silt loam (fine-silty mixed superactive mesic Pachic Ultic Argixeroll), with a pH of 5.4, and 4-6% organic matter content prior to planting.

Experimental design. Twelve treatments were arranged in a split plot design with pre-plant incorporation (with or without incorporation of Douglas-fir sawdust [*Pseudotsuga menziesii* Mirbel]) as main plots, and a combination of three nitrogen (N) fertilization rates (low, medium, and high) and two surface mulch treatments (bare soil and sawdust mulch) as the subplot treatments replicated four times. The four main plots consisted of two rows of plants each (one with pre-plant incorporation of sawdust and one without), and each subplot consisted of 20 plants. Subplots were separated by a 3m long unplanted section. Rows were spaced 3.0m apart, and plants were established 0.76m apart (4,300 plants/ha). A guard row was also planted on each side of the field. Plants were established on raised beds that were approx. 0.3m high.

<u>Incorporation (I).</u> Just prior to planting (Oct. 2003) raised beds were constructed with or without incorporation of sawdust. Sawdust (60% 2 mm or finer; C:N 790:1) was

incorporated by applying in a strip 0.1 m deep and 0.4 m wide (141 $m^3 \cdot ha^{-1}$) centered down the length of each incorporated row (White, 2006). Nitrogen fertilizer in the form of ammonium sulfate (16–16–16) was added to each incorporated row at a rate of 45 $kg \cdot ha^{-1}$ of N to help facilitate decomposition of sawdust, a standard commercial practice (Eleveld et al., 2005; Julian et al., 2011; Strik et al., 1993). The sawdust and fertilizer were incorporated into the existing soil using a rototiller. Raised beds were constructed on incorporated and non-incorporated rows using a bed shaper. Rows without incorporated sawdust received the same rate of phosphorus (P) and potassium (K) as incorporated rows.

Surface mulch (M). In treatments with a surface mulch, sawdust was applied to the top and sides of the raised beds to a depth of 5-8 cm (averaging 155 $m^3 \cdot ha^{-1}$) immediately after planting using a sawdust spreader, per standard commercial practice. New sawdust was added every few years, as required to maintain a mulch depth of approx. 7 to 8 cm (Strik et al., 1993). The most recent application of fresh sawdust mulch, prior to this study, occurred in Sept. 2010.

Nitrogen fertilization rate (Nrate) and other nutrient fertilization. Plants were fertilized with a low, medium, or high rate of N with the rates per hectare increasing as the planting aged (Table 1), per standard commercial practice (Hart et al., 2006). In 2011 and 2012, the total N fertilizer was divided into thirds and applied in mid-April (bloom), mid-May, and mid-June (green fruit) using urea (46–0–0) as a broadcast band applied to the in-row area, on top of the sawdust mulch, if present. Phosphorus and K were applied annually, in spring, at a rate of 33 and 70 kg·ha^{-1}, respectively, (0–22–25; P as triple superphosphate and K as potassium sulfate).

Irrigation. The planting was drip irrigated with a single line per row containing 3.8 L·h^{-1} emitters placed 0.15 m on either side of the base of the plant. The emitters were installed with spaghetti tubing attached, the end of which was inserted into the soil to a depth of about 6 cm. Inserting the tubing into the soil slightly alleviated the problem of water run-off from the raised beds. The planting was irrigated based on weather and related crop evapotranspiration and soil moisture (see below) from 10 June to 27 Sept. 2011 and 14 May to 12 Oct. 2012.

Rows with pre-plant incorporation of sawdust were irrigated for 15 min., six times/d for a total of 1.5 h·d^{-1} between 05:00 and 17:00, seven days/wk (79.8 L·plant^{-1}·wk^{-1}). Rows with no incorporation of sawdust prior to planting, were irrigated for 13 min., six times/day, for a total of 1.3 h·d^{-1} between 5:30 and 17:30, seven days/wk (69.2 L·plant^{-1}·wk^{-1}). The difference in irrigation was found necessary in the establishment years (White, 2006) and subsequently in order to maintain similar average soil moisture between the main plot incorporation treatments. Soil moisture was monitored using time domain reflectometry (TDR), from Trase System, Soil Moisture Equipment Corp., Santa Barbara, California (model 6060e06). 20 cm buriable uncoated wavelength probes were placed on 8 plots in order to get an average soil moisture content of between 24-30%.

Data collection. Biomass and plant carbon. Plants were pruned in Jan. 2011, Jan 2012 and Dec. 2012, per standard commercial practice (Strik et al., 1993). The wood removed (prunings) was divided into 1-year-old wood (NW; “new wood”; last season’s growth) and older wood (> 1-year-old; OW; “old wood”), was dried to a constant weight at 60 °C and dry weight (DW) per plot and per plant obtained.

Immediately after pruning, one plant per plot was randomly selected for destructive harvest. Plants were excavated using a tractor, taking care to remove as much of the root system as possible; any root material that remained in the ground once the plant was removed was collected. Plant rooting depth was estimated and recorded. Soil was removed from plant roots using high pressure water. Plants were then separated into small, fine, fibrous roots (≤ 1 mm diam.; “FR”), large, storage roots (> 1 mm diam.; “SR”), crown, 1–year-old wood (NW), and old wood (OW). Plant parts were oven dried to a constant DW at 60 ºC and sub-samples removed and sent to Brookside Laboratories, Inc. (New Knoxville, OH) for analysis of carbon concentration.

Leaves were collected from a randomly selected plant/plot, as they senesced in autumn 2011 and 2012, by placing a net around each selected plant when leaves first started changing color. Once all leaves had senesced, they were collected, dried at 60 ºC until a constant DW, and a sub-sample taken and sent to Brookside Laboratories for analysis of C concentration.

Blueberry fruit were harvested using an over-the-row, rotary machine harvester (Littau Harvesters Inc., Stayton, OR) on 31 Aug., 7 Sept., and 19 Sept. 2011 and 15 Aug., 22 Aug., and 5 Sept. 2012. On each harvest date, data were collected on total yield/plot.

On the second harvest of each year, 25 fruit were randomly sub-sampled for analysis of C concentration (Brookside Laboratories). In both years of the study, *Blueberry Shock Virus* affected some plants or sections of plants in some plots. Since plants affected by this pollen-borne virus do not produce fruit in the "shock" year, the proportion of the plot producing fruit was estimated to calculate yield per plant.

Mycorrhizal staining and counting. To assess treatment effects on the level of mycorrhizal infection, root samples were collected in October of each year. In each plot, four soil cores of approx. 3 cm diam. were taken to a depth of 0.30 m (with the mulch in the sawdust treatment removed), one in each corner about 20-25 cm from the plant crown. Each core was divided into the top 0.15 m and bottom 0.15 m of soil. Soil cores were washed with water and the roots present separated and stained with Trypan blue as described by Phillips and Hayman (1970). The percentage of mycorrhizal infection was quantified using the grid-line technique (Giovannetti and Mosse, 1980).

Soil analysis. Soil samples were taken from each plot in July of 2011 and 2012. Soil cores were sampled to a 0.3 m depth approx. 0.5 m from the crown of three randomly chosen plants in each plot. Cores were mixed and a subsample of the mixture was prepared for analysis by air-drying and screening to pass a 2-mm mesh screen. Extractable soil S, Ca, Mg, K, Na, B, Fe, Mn, Cu, Zn, Al, and P were determined via ICP after extraction via the Mehlich 3 method (Mehlich, 1984) at Brookside Laboratories. Soil NO_3-N and NH_4-N were determined via automated colorimetric methods after extraction with 1 M KCl (Dahnke, 1990) and soil pH via the 1:1 soil:water method (McLean, 1982).

Calculations. <u>Biomass and growth allocation.</u> The total biomass of dormant plants was calculated by adding the DW of each plant part (crown, FR, SR, NW, OW). Biomass allocation was calculated by dividing the DW of each plant part by total plant DW (and multiplying by 100 to get percent allocation).

Total annual plant biomass growth, was calculated in 2012 by adding the growth of the crown and fibrous and storage roots (the increase in DW from 2011 to 2012) and the growth of old wood (calculated by subtracting the DW of OW in 2011 from the dormant plant and OW prunings removed in 2012) to the DW produced in 2012 (DW of NW, fruit, and leaves). The allocation of annual growth in 2012 was calculated by dividing the growth (DW) of each plant part by total growth (and multiplying by 100 to get percent allocation).

<u>Carbon concentration, mass and allocation.</u> Total mass of C per plant part was calculated by multiplying the DW of each plant part by the corresponding concentration of C. Allocation of plant C was calculated as described previously for DW.

Statistical analyses. Analysis of all treatment effects on plant biomass and allocation, C concentration, content, and allocation, and mycorrhizal infection was done using analysis of variance for a complete factorial split-plot design (incorporation as main plot and the combination of mulch and Nrate as the split plots) using the PROC MIXED procedure in SAS (software package version 9.3; SAS Institute, Cary, NC). Results were analysed separately for the two years because there were significant year effects. Treatment means were compared using a Fisher's protected least significant difference

(LSD). The PROC CORR procedure in SAS was used to determine the Pearson's correlation coefficient between DW of 1-year-old wood in winter 2011 with the DW of fruit produced in 2012. All analyses included checking data for homogeneity of variance; no transformation of data was required. P-values ≤0.05 were considered significant, while p-values between 0.05 and 0.1 were considered trends or tendencies.

Results

Dormant plant biomass and C allocation. In 2011, total dormant plant biomass and C mass were not significantly affected by the main effect of incorporation with sawdust or the presence of sawdust mulch (Tables 2-2 and 2-3). There was, however, a significant incorporation by mulch (I×M) interaction effect on total plant biomass, as well as the biomass of storage tissues such as old-wood and storage roots, which accounted for over 50% of the total dormant plant dry weight (Table 2-2). Plants grown in plots with incorporated sawdust but no mulch had the greatest DW and C content of storage roots and old wood (Tables 2-2 and 2-3) and tended (*P* = 0.0830) to have the greatest crown DW and C mass. In plots with no sawdust incorporated the presence of a mulch increased total plant DW and C mass. There was a significant I×M interaction on the allocation of biomass to storage roots with the highest allocation in bare, un-mulched soil when sawdust was incorporated and the lowest in bare soil when no sawdust was incorporated (Table 2-7). Plants grown with no added organic matter had the lowest total biomass, ranging from 0.4 to 0.9 kg·plant^{-1} less DW than those grown with sawdust either as a pre-plant amendment or as a mulch (Table 2-2). These plants also had the lowest C mass, ranging from 0.2 to 9.5 kg·plant^{-1} less C than those grown with some form of organic matter addition (Table 2-3).

In 2011, root depth tended to increase with incorporation (*P* = 0.0731), with plants growing in non-amended plots having an average root depth of 0.46 m, while those in incorporated plots having a root depth of 0.47 m; this trend was not seen in 2012. In both years plants growing in un-mulched plots had the greatest root depth when fertilized with the low rate of N (0.47m) and the least with the medium rate of N (0.45m), whereas plants in mulched plots had the greatest root depth when fertilized with the medium rate of N (0.51 m), and the least depth with the high rate of N (0.44 m) (trend; *P* = 0.0733, *P* = 0.0982 for M×Nrate in 2011 and 2012, respectively).

Plants grown with a sawdust mulch produced less fibrous root DW and C mass than those grown in bare soil plots (Tables 2-2 and 2-3). Mulch also significantly affected the biomass and C allocation to storage roots (Table 2-7 and 2-8), with the highest DW and C in bare soil and the lowest in mulched plots. There was also a significant interaction effect (I×M) on the allocation of biomass to fibrous roots (Table 2-7). Incorporation of sawdust as a main effect only affected the C allocation to fibrous roots, with the highest C allocation in non-incorporated plots and the lowest in incorporated soils (Table 2-8). This pattern was also seen with the DW of new-wood, which tended (*P* = 0.0663) to be greater (14%) in incorporated plots than un-amended plots, but there was no such trend for C mass.

Nrate did not affect the DW or C mass of any plant part (Tables 2-2 and 2-3). The allocation of biomass to fibrous roots, however, was significantly increased by Nrate (Table 2-7). The allocation of C to fibrous roots was also increased by Nrate (Table 8). There was also a Nrate by mulch interaction (Nrate×M) on the DW and C content of old wood (Tables 2 and 3; Figs. 2-1 and 2-2), with greater values in bare soil plots than in mulched plots when plants were fertilized with the low or high rate of N, but the opposite was found when plants were fertilized with the medium rate of N (Figs. 2-1 and 2-2). This same interaction pattern tended to be observed for total plant DW and C mass (P = 0.0732; P = 0.0643, respectively).

In 2012, incorporation had no effect on total dormant plant DW or C mass (Tables 2 and 3). There was a significant I×M interaction only for crown DW (Table 2) and a trend (P = 0.0604) for crown C mass with plants grown in incorporated plots having the greatest DW or C content when grown with no mulch and the opposite being found when plants were grown in un-incorporated soil.

Similarly to what was found in 2011, mulching reduced the DW and C content of fibrous roots relative to bare soil (Tables 2 and 3). Biomass allocation to fibrous roots was also significantly reduced in mulched plants compared to bare soil (Table 7); allocation of C to fibrous roots followed a similar pattern (Table 8). Unlike in 2011, higher rates of Nrate increased the DW and C content of fibrous roots. Allocation of biomass to fibrous roots was also affected by N rate (Table 7). There was an interaction between incorporation and Nrate (I×Nrate) on the DW of fibrous roots, with incorporated plots having fibrous root biomass averaging 23.7, 19.6, and 37.3g and non-incorporated plots averaging 14.6, 27.5, and 25.0 g for the low, medium, and high rate of N, respectively (Table 1). The interaction effect (I×Nrate) was marginally non-significant for C mass (P = 0.0536). Nrate did not affect DW or C mass of any other plant part or the total plant.

Carbon concentration. In 2011, mulch significantly increased the concentration of carbon (%C) of fibrous roots and crown, with plants grown in bare soil having a lower %C than those grown in mulched plots (Table 9). Increased N decreased the %C of new wood (Table 9). In 2012, mulch continued to significantly increase the %C of fibrous roots but did not affect %C of the crown, and increased N continued to decrease the %C of fibrous roots. There was a significant interaction between N and sawdust incorporation on the %C of storage roots, with higher N increasing %C of storage roots in incorporated plots and decreasing %C of storage roots in un-incorporated plots (Table 9).

Allocation of annual growth. There were no treatment effects on the allocation of annual growth or DW; thus all treatments were averaged. Almost 50% of the annual growth was allocated to fruit and increases in growth of old wood (Fig. 3).

Gains and losses. In 2011, there were no treatment effects on total plant loss of DW or C (Table 4). The DW of prunings (OW and NW) for plants grown in incorporated plots was greater when no mulch was present, but the opposite was found for plants grown in un-incorporated soil. A similar response was found for the C mass lost in OW prunings (Table 4).

In 2012, pre-plant incorporation of sawdust significantly increased fruit DW and C loss at harvest, leaf C content, OW prunings, and total plant DW and C loss (Table 4). Total plant loss of biomass (DW) and C was 17.4 and 17.7% greater, respectively, in incorporated plots than un-amended plots. The DW of one-year-old prunings (NW) in the winter preceding fruit harvest was significantly correlated to fruit DW in 2011 (r = 0.56; $P < 0.0001$) and in 2012 (r = 0.48; $P < 0.006$). There was a significant three-way interaction among the treatments on their effect on C lost through senesced leaves (I × M ×Nrate; Table 4). Losses in C with leaves at senescence were greatest in incorporated plots with bare soil and the lowest rate of N applied and in un-amended soil with no mulch at the highest rate of N applied (Fig. 4). There were no treatment effects on gains in biomass and C mass in 2012 for the dormant plant or individual plant parts (Table 4).

Soil nutrients. Incorporation of sawdust prior to planting in Oct. 2003 increased soil organic matter, Ca, Mg, Mn and Cu concentration in July 2011 (Table 5), and tended to increase soil pH, K, and Fe ($P = 0.0797$, $P = 0.0608$, $P = 0.0718$, respectively). Mulch decreased soil organic matter, ammonium-N, P, and S concentration (Table 5) and tended to decrease the concentration of nitrate-N and K and increase soil Mn ($P = 0.0624$, $P = 0.0864$, $P = 0.0608$, $P = 0.0803$, respectively). There was a significant interaction between incorporation and mulch for soil, pH, S and Mg, with the highest values found in incorporated, bare soil plots and the lowest in non-incorporated bare soils (Table 5).

In 2012, incorporation with sawdust significantly increased organic matter and tended to increase soil Fe and Mn concentration ($P = 0.0535$ and $P = 0.0875$, respectively). Mulch significantly decreased soil nitrate-N, P, K and Zn concentration and significantly increased organic matter, and tended to increase Mn concentration ($P = 0.0728$). There was a significant incorporation by mulch interaction on soil organic matter (Table 5).

Mycorrhizal infection. In both years, mycorrhizal infection was affected by mulch, Nrate, and the interaction of mulch and N rate (Table 6). In 2011, there was a trend for roots found at a depth of 15 to 30 cm to have a lower mycorrhizal infection when sawdust was incorporated prior to planting, whereas incorporation reduced infection at both depths in 2012. The presence of sawdust mulch significantly decreased mycorrhizal infection at both rooting depths. Higher rates of N fertilization reduced mycorrhizal infection (Table 6) with plants fertilized with the low

Nrate having an average value of 12% more infection than those fertilized with the high N rate. Mycorrhizal infection was reduced by higher rates of N fertilization in bare soil plots, but there was little effect of N rate in mulched plots (Fig. 5 and 6). There was no significant correlation between mycorrhizal infection and total plant dry weight or fruit biomass in either year.

Discussion

The effects of pre-plant incorporation with sawdust, sawdust mulch, and N fertilization rate on plant biomass, and fruit production in the mature years of our study differed from what was found during establishment (White, 2006). Many of these treatments seem to have both short-term and long-term effects on mature blueberry plants. This is interesting partly because we don’t really know much about these dynamics in mature fields, instead much of the literature has come from younger establishing plantings. In the establishment years of this trial, incorporation of sawdust prior to planting reduced plant dry weight and early fruit production compared to un-amended soil, perhaps a result of increased soil porosity and more rapid flow through of irrigation water and fertilizer N reducing plant water and N availability (White, 2006). Although the impact of pre-plant amendments on blueberry plant growth and early fruit production have been mixed (Haynes and Swift, 1986; Moore, 1979; Spiers, 1982; Townsend, 1973a; Yang et al., 2002a), likely a result of differences in soil and amendment type among studies, there was no information until now on the long-term impacts of pre-plant amendment.

In the last year of our study, nine years after planting establishment, incorporation had a positive effect on the plant, increasing yield relative to un-amended soil, as found by Spiers in (1982). Even though yield was increased, dormant plant biomass was not affected. However, incorporation increased the weight of one-year-old wood in 2011 and there was a correlation between weight of one-year-old wood and fruit biomass produced the following year, as has been found by others in blueberry (Larco et al., 2013). Considering that the effect of pre-plant incorporation with sawdust in this planting changed from a negative effect on early fruit production in the third growing season (2006) to a positive effect on yield in the ninth growing season (2012), shows how pre-plant decisions can have long-term effects on blueberry plant growth and production.

An overall positive effect of incorporation on soil nutrition was found, although all plots were found to have soil nutrients being above recommended sufficiency levels for blueberry regardless of treatment (Hart et al., 2006). Soil pH, however, was elevated beyond the recommended range of 4.5 to 5.5 for blueberry, and was likely increased with pre-plant incorporation as a result of higher soil calcium. We found that these mature blueberry plants had more biomass when growing in plots with the highest soil pH, a contrast to what has been documented for establishing blueberry plants (Cummings et al., 1981), indicting that a plant's preferences may be dynamic throughout its life cycle. It is important to note that the significant increase in organic matter, pH, and K, Ca, Mg, Fe, Mn and Cu concentration was found in 2011; in 2012 only Fe and Mn concentration tended to be higher in the incorporated treatment. With only two years of soil data it is not clear if the impact of incorporation was decreasing over time or if there was too much variability among treatments to find significance.

Organic matter addition in blueberry production was confirmed by this study, as well as by others, to be beneficial to the plant; with plants in un-amended bare plots having the lowest biomass and C storage (Cummings et al., 1981; Goulart et al., 1995; Haynes and Swift, 1986; Moore, 1979; Spiers, 1982, 2000; Yadong et al., 2006). However, too much organic matter addition, with both pre-planting incorporation and sawdust mulch, led to the second lowest values for dry weight and C mass. The results of this study also suggest that, specifically adding fresh sawdust mulch in established plantings has short-term effects on plant biomass and C allocation. In the year following the addition of fresh sawdust (2011), there was a significant mulch by incorporation interaction whereas these treatments had no effect the following year, two years after replenishing the mulch. Data on this short-term effect of mulch maintenance is lacking as most studies aren’t long enough or are conducted during establishment when the first mulch application occurs. The (IxM) interaction found in our study for mature plants, contrasts what has been found in other studies for establishing plants. Goulart et al. (1995) found that establishing ‘Bluecrop’ blueberry plants were shorter and had less canopy volume and foliage density in plots that were un-amended with no surface mulch and the greatest growth occurred in incorporated, mulched plots. In contrast, during the establishment years of the field used in this study the greatest growth and biomass occurred in un-amended soils with a surface mulch and the least growth occureed in amended soils with no mulch (White 2006). Seven years later, plants grown in amended soils with no mulch had the greatest biomass. Our results indicate that plants growing in amended soil had a greater growth rate during plant maturation to compensate for the reduced growth observed during the establishment years. Even though these blueberry plants were considered “mature”, dormant plant biomass increased 40% from 2011 to 2012. However, the annual growth observed was not significantly affected by amending with sawdust prior to planting and nine years of mulching vs. non-mulching and varying rates of N fertilization.

It is clear that the relationship between blueberry plant growth and organic matter, when applied as a pre-plant amendment or as mulch is complicated. In our study, weeds were managed in all treatments and the entire planting was well irrigated. Mulch has been shown to benefit blueberry plant growth when weeds are not managed in the comparator bare soil plots (e.g. Krewer et al., 2009;Sciarappa et al., 2008; Sinkevičienė, et al., 2009**)** or when plantings are not irrigated (Spiers, 1986). In our study, there was no main effect of sawdust mulch on above-ground plant biomass, C storage, growth, or fruit production even though mulch affected soil nutrient concentration and organic matter. In soils, mulch was found to decrease P and K relative to un-mulched plots, however, this was not the case during establishment years for soil P, which was increased with mulch (Sinkevičienė, et al., 2009; White, 2006). Despite the decrease in soil P and K, their concentrations were well above recommended sufficiency for blueberry (Hart et al., 2006).

Root depth was not affected by treatment, and did not differ during the two years of the study. Other studies performed on root size and distribution have either not differentiated between different root types and sizes (fibrous vs. storage) (Bryla and Strik, 2007; Patten et al., 1988; Spiers, 1986, 2000) or have focused in detail on the different orders of root sizes, separating what was considered fibrous roots in this study into 5 to 7 different orders according to diameter (Valenzuela-Estrada et al., 2008). These differences in methodology make it hard to compare this study’s results with different studies, especially those that do not differentiate between fibrous and storage roots. Storage roots in our mature plants reached a maximum depth of 0.46 m similar to what was reported in five-year-old ‘Elliott’ (Bryla and Strik, 2007). Gough (1980) found roots of mature blueberry plants at depths up to 0.8 m with most at 0 to 0.6 m and at a distance of up to 1.8 m from the plant crown. Rooting depth in blueberry may be influenced by soil type, depth of the water table, species, cultivar, and cultural conditions such as the addition of organic matter (Austin, 1982; Spiers, 1986). In all of our treatments, fibrous roots were concentrated in the uppermost 0.15 m of soil, with mulched plots having significantly reduced fibrous root biomass potentially due to increased temperatures and differences in soil moisture. Bryla and Strik (2007) reported that root diameter increased with rooting depth in ‘Elliott’ and Valenzuela-Estrada et al. (2008) found that ‘Bluecrop’ root length declined in an exponential fashion with an increase in root order. In young rabbiteye blueberry, 40-50% of roots were concentrated in the core closest to the crown, with a linear decrease in root density with depth, particularly in the first 0.10 m distance from the crown (Patten et al., 1988).

In this long-term trial, the lack of significant response to N fertilization rate in blueberry plants appears to be related to the plant’s ability to obtain sufficient nutrients from various sources (e.g. fertilization, mineralization of organic materials, the soil, and through association with mycorrhizae). Although young blueberry plants have been shown to be quite responsive to addition of organic matter (Cummings et al., 1981; Haynes and Swift, 1986; White, 2006) and varying rates of N fertilizer (Bañados et al 2012; Cummings et al., 1981; White, 2006), mature plants have been shown in this study and by others (Bañados, 2006) to be less responsive to changes in N fertilization. The documented lack of response to changes in N fertilization for large blueberry plants is often attributed to the large “buffering” capacity of the crown, roots, and old wood which serve as storage organs for N and other nutrients (Bañados, 2006). This might explain why the detrimental effect of mulching found on fibrous root biomass, biomass allocation, C storage, C allocation, and mycorrhizal infection on fibrous roots did not impact total plant growth and biomass. The lack of response to N fertilizer may have been related to the amount of available N in the soil pool, which was not significantly affected by fertilization, but was decreased by the addition of sawdust mulch. In the soil, the presence of a mulch decreased ammonium-N and nitrate-N; the response in ammonium-N was first seen a year after addition of fresh sawdust to the mulch layer, whereas the nitrate-N response was seen a year later. The decrease in available N was expected due to the increase in C:N ratio caused by the addition of fresh sawdust to the mulch and the associated immobilization of some fertilizer N (White, 2006). Blueberry plants have been shown to compensate for low N fertilization by using soil available N to achieve similar growth rates and production (Bañados et al., 2012). Because soil nutrient pools were found to be above the recommended level for blueberry production, it safe to assume that the plants were getting enough nutrients even with the reduced biomass of their nutrient absorptive fibrous roots and the reduced infection of their nutrient-foraging symbiotic fungi. Increased rates of N fertilizer also decreased mycorrhizal infection, likely because the fungi played more of a saprophytic role than a symbiotic one as the plant had enough N available and therefore did not need the fungal association as much.

Mycorrhizal infection was also lowered by organic matter addition as incorporation or as mulch. This negative response of incorporation has been reported in the establishment years of blueberry in other studies (Goulart et al, 1998; White, 2006; Yang et al., 2002a; Yadong et al., 2006). Yang et al. (2002a) also found that pre-plant amendment with sawdust (C/N ratio of 59:1) reduced infection more than forest litter and no amendment. In our study, the C/N ratio of the sawdust used to amend the soil was 790:1. The mycorrhizal C drain caused by the symbiosis between the host plant and the fungus, may be offset in part by the gain of C and N in organic sources such as amino acids that ericoid mycorrhizae are able to transfer to the host plant (Stribley and Read, 1974a and 1974b). High levels of mycorrhizal infection in plots with no added organic matter pre-plant or as a mulch was also found by Goulart et al. (1995, 1997). Despite the lack of a correlation between total plant DW and mycorrhizal infection in mature plants in our study, plant DW is positively correlated with mycorrhizal infection in establishing years (Yang et al., 2002a) or has an inconsistent relationship (White, 2006) in establishing plants. The increase in plant biomass in mycorrhizal plants during establishment may be due to a combination of needed increased mineral nutrient acquisition and low C drain in the host blueberry plant, or a lower C cost per unit of nutrient return, favoring immature plants but not necessary in mature plants which can get their nutrient needs from the established soil pool. It is important to note that differences in mycorrhizal infection could be attributed to a dilution effect from increased root volume and growth, and not a real decrease in actual number of infection sites, resulting in a reduced mycorrhizal concentration (Goulart et al., 1993, 1995).

Some of the variation in dormant plant biomass from year to year was likely due to changes in pruning severity. In our study, dormant plant biomass was measured after plant pruning. However, our calculations of the gain in biomass and C in 2012 indicate that these mature plants were still growing and, therefore, currently increasing the C pool of the plant. Previous studies of C dynamics in blueberry and other perennial fruit crops have focused on how the plant partitions carbohydrates between soluble sugars and starches (Darnell, 1991), how C reserves are used to supply sinks like fruits and new leaves and how these change with removal of plant parts (Gauci et al., 2009, Candolfi-Vasconcelos, et al., 1994), measurement of plant photosynthetic and respiration rates (Lakso et al., 1999), or how the plants translocate photosynthates by using methods such ^{14}C labeling (Darnell, 1991; Privé et al., 1994). There is no information, to date, on total plant C storage or partitioning in blueberry. The results from this study indicate that blueberry plant C storage is affected by the long-term and short-term effects of managements, although it seems that the decision to incorporate organic matter pre-planting, is the first determinant of plant biomass and consequently C storage. It is important to note that the differences in %C in plant parts in both years were all less than 2%; and although some were statistically significant, most may not be biologically significant. Therefore, it appears that the management and treatment effects on C storage come from the response in plant biomass and not from the response of C concentrations in the plant.

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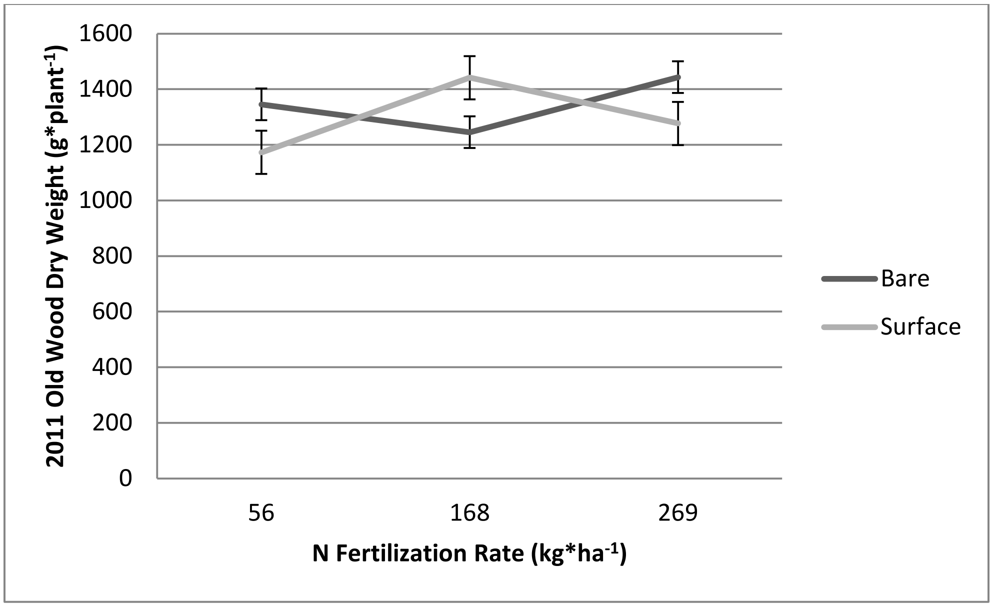

Figure 2-1. Effect of N Fertilizer rate and mulching on old wood dry weight in 2011 (Mean ±SE) Legend: Mulch treatment

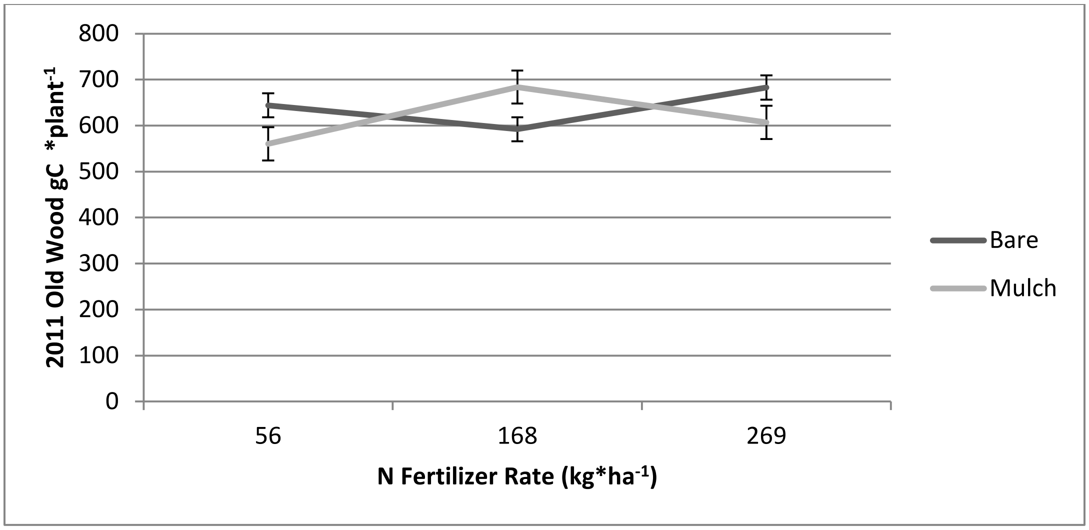

Figure 2-2 Effect of N Fertilizer rate and mulching on old wood g C per plant in 2011 (Mean ±SE) Legend: Mulch treatment

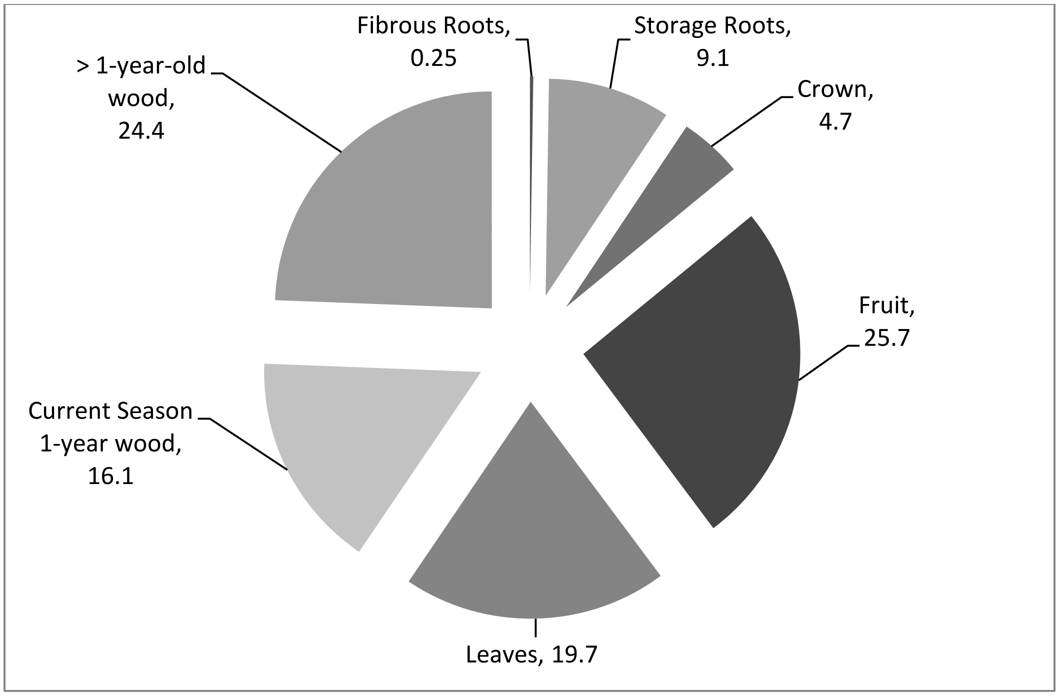

Figure 2-3 Plant annual biomass growth allocation 2012 (all treatments were averaged due to lack of significance, n=48, values in percentages)

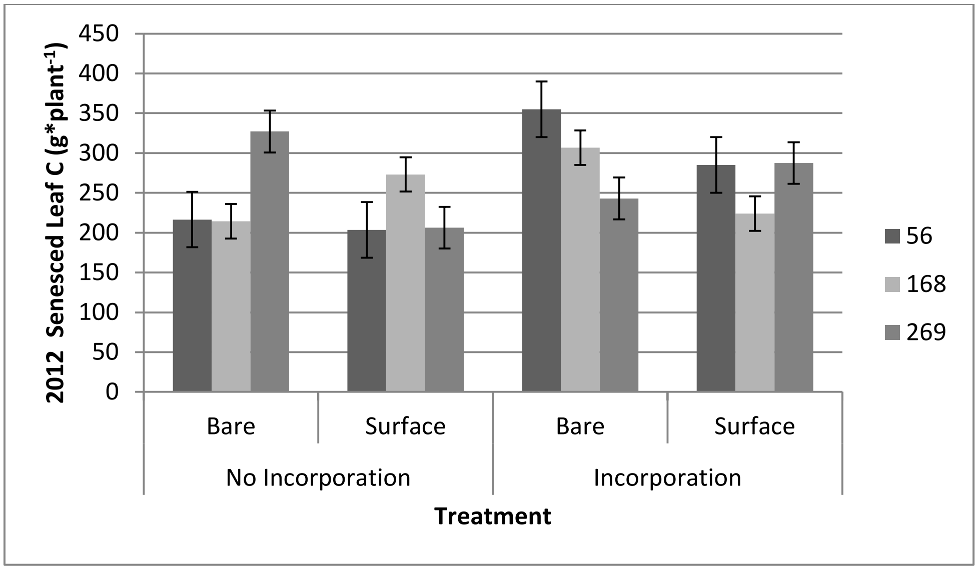

Figure 2-4. Three-way interaction effect of pre-plant (2003) incorporation of sawdust, sawdust mulch and nitrogen fertilizer rate (throughout planting life) on senesced leaves of 'Elliott' blueberries in 2012. Nitrogen fertilizer rate in (kg N*ha^{-1}) (Mean ± SE)

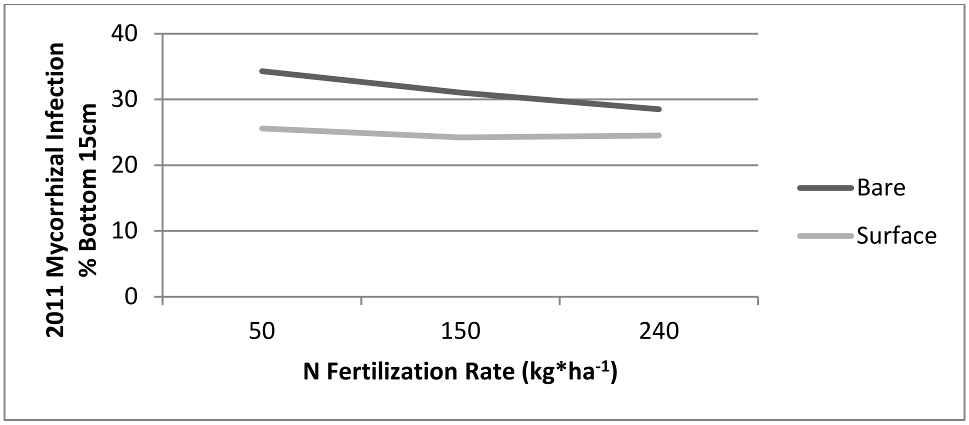

Figure 2-5. Effect of the interaction between sawdust mulch and nitrogen fertilizer rate (throughout planting life) on the percent mycorrhizal infection of roots found from 0.15 to 0.30 m depth, 'Elliott', 2011. Legend: Mulch treatment

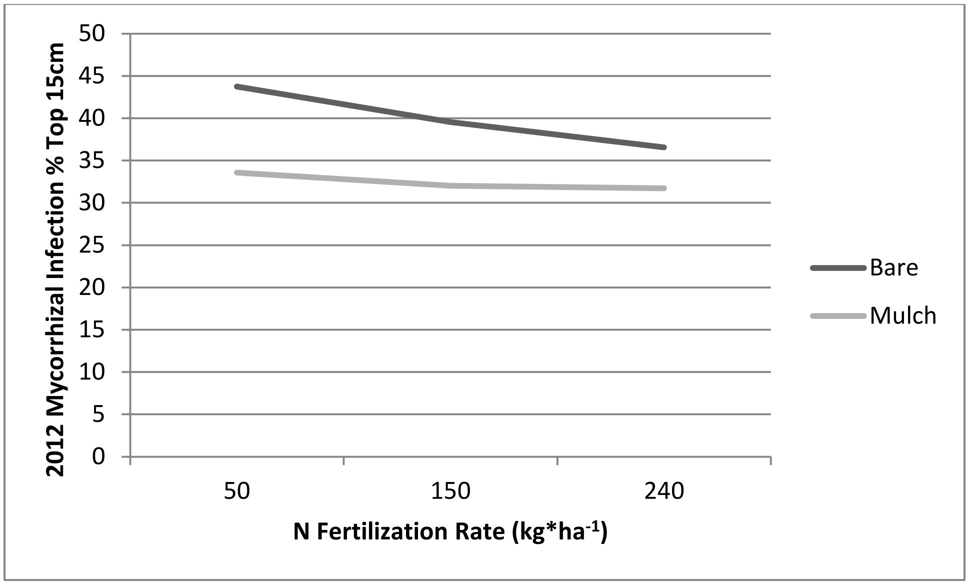

Figure 2-6. Effect of the interaction between sawdust mulch and nitrogen fertilizer rate (throughout planting life) on the percent mycorrhizal infection of roots found from 0 to 0.15 m depth, ‘Elliott’, 2011. Legend: Mulch treatment

Table 2-1. Nitrogen (N) fertilizer rate treatments applied to ‘Elliott’ blueberry from the first (2004) through ninth (2012) growing seasons at the North Willamette Research and Extension Center, OSU.

	N Fertilizer Rate (kg N*ha^{-1})							
N treatment	2004	2005	2006	2007	2008	2009	2010-2012	Total
Low	22	22	31	31	39	39	56	352
Medium	67	67	395	395	112	112	168	1052
High	112	112	153	153	185	185	269	1707

Table 2-2. Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch and nitrogen fertilizer rate (throughout planting life) on the dry weight of dormant plants of ‘Elliott’ blueberry in 2011 and 2012.

	Dormant plant dry weight[x]										
Treatment	Fibrous roots (g)		Storage roots (kg)		Crown (g)		Old wood (kg)		1-year-old wood(g)	Total (kg)	
2011											
No Incorporation	Bare	Mulch	Bare	Mulch	668.3		Bare	Mulch	198.0	Bare	Mulch
	35.2	19.7	1.0	1.2			1.1	1.3		2.6	3.2
Incorporation	Bare	Mulch	Bare	Mulch	695.4		Bare	Mulch	231.1	Bare	Mulch
	34.3	20.8	1.3	1.1			1.5	1.2		3.5	3.0
Significance											
Incorporation (I)	ns[z]		ns		ns		ns		0.0663	ns	
Mulch(M)	0.0004[y]		ns		ns		ns		ns	ns	
Nrate[x]	ns		ns		ns		ns		ns	ns	
IxM	ns		0.0008		ns		0.0398		ns	0.0101	
IxNrate	ns		ns		ns		ns		ns	ns	
MxNrate	ns		ns		ns		0.0289[u]		ns	ns	
IxMxNrate	ns		ns		ns		ns		ns	ns	
2012											
No Incorporation	Bare	Mulch	1.1		Bare	Mulch	1.7		390.8	4.0	
	29.1	22.2			626.6	788.3					
Incorporation	Bare	Mulch	1.2		Bare	Mulch	2.0		357.0	4.5	
	27.2	15.7			855.0	674.4					
Nrate	L	19.2a[w]									
	M	23.6ab									
	H	31.1b									
Significance											
Incorporation (I)	ns		ns		ns		ns		ns	ns	
Mulch(M)	0.0040		ns		ns		ns		ns	ns	
Nrate	0.0110		ns		ns		ns		ns	ns	
IxM	ns		ns		0.0446		ns		ns	ns	
IxNrate	0.0245		ns		ns		ns		ns	ns	
MxNrate	ns		ns		ns		ns		ns	ns	
IxMxNrate	ns		ns		ns		ns		ns	ns	

[z] ns=Non significant

[y] $P \leq 0.05$

[x] “Nitrogen fertilizer rate in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

[w] Mean separation (in columns and rows) by LSD test at $P \leq 0.05$ (lowercase)

[v] Plants were destructively harvested in January 2011 and Dec 2012.

[u] interaction seen in Figure 1

Table 2-3. Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch and nitrogen fertilizer rate (throughout planting life) on the carbon mass of dormant plants of ‘Elliott’ blueberry in 2011 and 2012.

	Dormant Plant Carbon[v]									
Treatment	Fibrous roots(g)		Storage roots(g)		Crown(g)	Old wood (g)		1-year-wood (g)	Total (kg)	
2011										
No Incorporation	Bare	Mulch	Bare	Mulch	295.6	Bare	Mulch	97.2	Bare	Mulch
	17.2	9.7	327.4	434.6		538.4	627.5		1.2	1.5
Incorporation	Bare	Mulch	Bare	Mulch	309.1	Bare	Mulch	112.6	Bare	Mulch
	16.8	10.3	499.3	408.0		696.0	588.1		1.7	1.4
Significance										
Incorporation(I)	ns[z]		ns		ns	ns		ns	ns	
Mulch(M)	0.0005[y]		ns		ns	ns		ns	ns	
Nrate[x]	ns		ns		ns	ns		ns	ns	
IxM	ns		0.0006		ns	0.0264		ns	0.0075	
IxNrate	ns		ns		ns	ns		ns	ns	
MxNrate	ns		ns		ns	0.0290[u]		ns	ns	
IxMxNrate	ns		ns		ns	ns		ns	ns	
2012										
No Incorporation	11.1		535.6		566.4	780.1		189.9	1.8	
Incorporation	9.6		586.3		566.5	971.6		174.0	1.7	
Bare	12.2									
Mulch	8.6									
Nrate	L	8.4a[w]								
	M	9.9ab								
	H	12.8b								
Significance										
Incorporation(I)	ns		ns		ns	ns		ns	ns	
Mulch(M)	0.0120		ns		ns	ns		ns	ns	
Nrate	0.0381		ns		ns	ns		ns	ns	
IxM	ns		ns		ns	ns		ns	ns	
IxNrate	ns		ns		ns	ns		ns	ns	
MxNrate	ns		ns		ns	ns		ns	ns	
IxMxNrate	ns		ns		ns	ns		ns	ns	

[z] ns=Non significant

[y] $P \leq 0.05$

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

[w] Mean separation (in columns and rows) by LSD test at $P \leq 0.05$ (lowercase)

[v] Plants were destructively harvested in January 2011 and Dec 2012.

[u] interaction seen in Figure 1.

Table 2-4. Effects of pre-planting (2003) incorporation of sawdust and sawdust mulch (throughout planting life) on annual fruit, leaf and pruning biomass and C mass losses, and on dormant plant annual gain in biomass and C mass, of ‘Elliott’ blueberry in 2011 and 2012.

	Biomass (g)				Plant C (g)			
	No-		Incorporation		No-		Incorporation	
Plant Parts	Bare	Mulch	Bare	Mulch	Bare	Mulch	Bare	Mulch
2011								
Losses								
Fruit	830.8				340.4			
Significance	ns				ns*			
Leaves	447.3				234.8			
Significance	ns				ns			
OG Pruning	294.9	370.8	473.6	343.1	140.0	177.4	225.8	162.3
Significance	0.0034^{I*M}				0.0174^{I*M}			
NG Pruning	125.5		157.6		66.1			
Significance	0.0344^{I}				ns			
Total	1809.6				830.3			
Significance	ns				ns			
2012								
Losses								
Fruit	609.7		672.4		246.8		272.1	
Significance	0.0153^{I}				0.0027^{I}			
Leaves	512.6				252.70	227.61	301.6	281.8
Significance	ns				$0.0315^{I*M*Nrate}$			
Old Wood	110.60	124.59	237.0	172.9	52.5	59.6	112.9	81.7
Significance	0.0297^{I} , 0.0476^{I*M}				0.0297^{I} , 0.0378^{I*M}			
1-year wood	61.7				19.7		40.4	
Significance	ns				0.0529^{I}			
TOTAL	1253.3		1517.2		570.6		693.7	
Significance	0.0002^{I}				0.0004^{I}			
2012								
Gains								
Fibrous Roots	5.8				1.3			
Significance	ns				ns			
Storage Roots	269.1				126.1			
Significance	ns				ns			
Crown	146.7				80.0			
Significance	ns				ns			
1-year wood	356.0				275.8			
Significance	ns				ns			
Old Wood	702.0				259.7			
Significance	ns				ns			
TOTAL	1541.3				779.1			
Significance	ns				ns			

[z] ns=Non significant

[y] P-value if $P \leq 0.05$

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

[w] Plants were destructively harvested in January 2011 and Dec 2012.

Table 2-5 Cont. Effects of pre-plant (2003) incorporation of sawdust and sawdust mulch (throughout planting life) on soil properties and nutrients 2011 and 2012. (continues on the next page)

	pH				Soil Nutrients							
Treatment					Nitrate (ppm)		Ammonium (ppm)		P (mg/kg)		K (mg/kg)	
2011												
No Incorporation	Bare	Surface	3.2		0.68		Bare	Surface	Bare	Mulch	Bare	Mulch
	5.82	6.11					2.4	1.3	185.9	161.33	177.8	159.6
Incorporation	Bare	Surface	4.3		0.8		Bare	Surface	Bare	Mulch	Bare	Mulch
	6.24	6.19					3.6	1.6	184.33	160.09	204.4	185.5
Significance												
Incorporation(I)	0.0797		0.0065		ns		ns		ns		0.0608	
Mulch(M)	ns		ns		0.0864		0.0013		0.0245		0.0038	
Nrate	ns		ns		ns		ns		ns		ns	
IxM	0.0239		ns		ns		ns		ns		ns	
IxNrate	ns		ns		ns		ns		ns		ns	
MxNrate	ns		ns		ns		ns		ns		ns	
IxMxNrate	ns		ns		ns		ns		ns		ns	
2012												
No Incorporation	5.8		Bare	Mulch	Bare	Mulch	Bare	Mulch	Bare	Mulch	Bare	Mulch
			3.0	3.2	2.5	1.18	7.2	5.5	170.2	161.0	200.7	168.8
Incorporation	6.0		Bare	Mulch	Bare	Mulch	Bare	Mulch	Bare	Mulch	Bare	Mulch
			3.74	4.6	2.96	0.63	8.0	7.6	195.89	152.5	215.9	173.4
Significance												
Incorporation(I)	ns		0.0109		ns		ns		ns		ns	
Mulch(M)	ns		0.0037		0.0146		ns		0.048		0.011	
Nrate	ns				ns		ns		ns		ns	
IxM	ns		0.0311		ns		ns		ns		ns	
IxNrate	ns				ns		ns		ns		ns	
MxNrate	ns				ns		ns		ns		ns	
IxMxNrate	ns				ns		ns		ns		ns	

[z] ns=Non significant

[z] ns=Non significant

[y] P-value if P ≤ 0.05

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

Table 2-5 Cont. Effects of pre-plant (2003) incorporation of sawdust and sawdust mulch (throughout planting life) on soil properties and nutrients 2011 and 2012.

			Soil Nutrients						
Treatment	S (ppm)		Ca (mg/kg)	Mg (mg/kg)		Mn (mg/kg)	Cu (mg/kg)	Zn (mg/kg)	
2011									
No Incorporation	Bare	Mulch	1208.4	Bare	Mulch	37.8	0.94	0.78	
	20.6	15.7		248.6	329.2				
Incorporation	Bare	Mulch	1428.1	Bare	Mulch	55.0	1.09	0.87	
	16.3	15.3		370.0	344.8				
Significance									
Incorporation(I)	ns		0.0197	0.0272		0.0092	0.019	ns	
Mulch(M)	0.0029		ns	ns		ns	ns	ns	
Nrate	ns		ns	ns		ns	ns	ns	
IxM	0.0233		ns	0.003		ns	ns	ns	
IxNrate	ns		ns	ns		ns	ns	ns	
MxNrate	ns		ns	ns		ns	ns	ns	
IxMxNrate	ns		ns	ns		ns	ns	ns	
2012									
No Incorporation	13.4		1194.67	290.4		39.6	1.14	Bare	Mulch
								1.44	1.12
Incorporation	11.2		1358.4	310		59.7	1.25	Bare	Mulch
								1.46	1.13
Significance									
Incorporation(I)	ns		ns	ns		ns	ns	ns	
Mulch(M)	ns		ns	ns		ns	ns	0.0156	
Nrate	ns		ns	ns		ns	ns	ns	
IxM	ns		ns	ns		ns	ns	ns	
IxNrate	ns		ns	ns		ns	ns	ns	
MxNrate	ns		ns	ns		ns	ns	ns	
IxMxNrate	ns		ns	ns		ns	ns	ns	

[z] ns=Non significant

[y] P-value if P ≤ 0.05

[x] “Nitrogen fertilizer rates in 2011 were 215 (“Low”), 645 (“Medium”), and 1033 (“High”) (Table 1)

Table 2-6. The main effects of pre-planting (2003) incorporation of sawdust, sawdust mulch (throughout planting life) and nitrogen fertilization on ‘Elliott’ blueberry roots mycorrhizal infection % found in the top (0 to 0.15 m) and bottom (0.15 to 0.30 m) of the rooting profile, in 2011 and 2012.

	2011		2012	
Mycorrhizal infection	Top	Bottom	Top	Bottom
Incorporation				
Not incorporated	36.64	29.10	37.60	29.66
Incorporated	33.86	26.38	34.38	27.10
Mulch				
Bare	38.58	30.72	39.56	31.37
Surface	31.92	24.76	32.43	25.39
Nitrogen Rate				
Low	37.96a[w]	29.92a	38.66a	30.52a
Medium	34.22b	26.79b	35.19b	27.50b
High	33.57b	26.50b	34.13b	27.12b
Significance (P-value or NS)				
Incorporation(I)	ns[z]	ns	0.018	0.0493
Mulch(M)	<.0001[y]	<.0001	<.0001	<.0001
Nrate[x]	0.0002	0.0014	<.0001	0.001
IxM	ns	ns	ns	ns
IxNrate	ns	ns	ns	ns
MxNrate	ns	0.0288	0.0015	0.0679
IxMxNrate	ns	ns	ns	ns

[z] ns=Non significant

[y] P-value if P ≤ 0.05

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

[w] Mean separation (in columns and rows) by LSD test at P ≤ 0.05 (lowercase)

[v] Soil core samples taken in October of 2011 and 2012

Table 2-7 Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch and nitrogen fertilizer rate (throughout planting life) on dormant plant biomass allocation, of ‘Elliott’ blueberry in 2011 and 2012.

	Dormant Plant % Allocation						
Treatment	Fibrous Root		Storage Root		Crown	Old Wood	1-year Wood
2011							
	Bare	Mulch	Bare	Mulch	22.5	54.5	10.2
	1.3	0.6	23	25.1			
No Incorporation							
	L	0.7a^{w}					
	M	0.9ab					
	H	1.0b					
	Bare	Mulch	Bare	Mulch	23	59.7	12.7
	0.9	0.6	25.7	24.8			
Incorporation							
	L	0.5a					
	M	0.7ab					
	H	0.8b					
Significance							
Incorporation(I)	ns^{z}		ns		ns	ns	ns
Mulch(M)	<.0001^{y}		ns		ns	ns	ns
Nrate^{x}	0.0452		ns		ns	ns	ns
IxM	0.0232		0.0477		ns	ns	ns
IxNrate	ns		ns		ns	ns	ns
MxNrate	ns		ns		ns	ns	ns
IxMxNrate	ns		ns		ns	ns	ns
2012							
	Bare	Mulch	31.3		21.3	53.7	11.8
	0.9	0.5					
No Incorporation	L	0.6					
	M	0.5					
	H	1.0					
	Bare	Mulch	33.9		21.0	58.2	11.8
	0.5	0.4					
Incorporation	L	0.3					
	M	0.6					
	H	0.5					
Significance							
Incorporation(I)	ns		ns		ns	ns	ns
Mulch(M)	0.0176		ns		ns	ns	ns
Nrate	0.0484		ns		ns	ns	ns
IxM	ns		ns		ns	ns	ns
IxNrate	ns		ns		ns	ns	ns
MxNrate	ns		ns		ns	ns	ns
IxMxNrate	ns		ns		ns	ns	ns

[z] ns=Non significant

[y] P-value if P ≤ 0.05

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269(“High”) (Table 1)

[w] Mean separation (in columns and rows) by LSD test at P ≤ 0.05 (lowercase)

Table 2-8 Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch and nitrogen fertilizer rate (throughout planting life) on dormant plant carbon allocation, of ‘Elliott’ blueberry in 2011 and 2012.

Treatment	Dormant Plant % Allocation						
	Fibrous Root		Storage Root		Crown	Old Wood	1-year Wood
2011							
	Bare	Mulch	Bare	Mulch	21	43	6.9
	1.3	0.7	24.6	29.4			
No Incorporation	L	0.9a^{x}					
	M	1.1ab					
	H	1.2b					
	Bare	Mulch	Bare	Mulch	20	42.6	7.2
	1	0.7	30.1	27			
Incorporation	L	0.7a^{w}					
	M	0.9ab					
	H	1b					
Significance							
Incorporation(I)	ns^{z}		ns		ns	ns	ns
Mulch(M)	<.0001^{y}		ns		ns	ns	ns
Nrate^{x}	0.0500		ns		ns	ns	ns
IxM	0.0237		0.0227		ns	ns	ns
IxNrate	ns		ns		ns	ns	ns
MxNrate	ns		ns		ns	ns	ns
IxMxNrate	ns		ns		ns	ns	ns
2012							
No Incorporation	Bare 0.7	Mulch 0.4	26.0		25.7	38.5	9.2
Incorporation	Bare 0.5	Mulch 0.3	26.2		22.9	42.8	7.7
Significance							
Incorporation(I)	ns		ns		ns	ns	ns
Mulch(M)	0.0458		ns		ns	ns	ns
Nrate	ns		ns		ns	ns	ns
IxM	ns		ns		ns	ns	ns
IxNrate	ns		ns		ns	ns	ns
MxNrate	ns		ns		ns	ns	ns
IxMxNrate	ns		ns		ns	ns	ns

[z] ns=Non significant

[y] P-value if $P \leq 0.05$

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

[w] Mean separation (in columns and rows) by LSD test at $P \leq 0.05$ (lowercase)

Table 2-9 Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch and nitrogen fertilizer rate (throughout planting life) on plant carbon concentration, of ‘Elliott’ blueberry in 2011 and 2012.

	Carbon concentration (%)											
Treatment	Fruit	Leaf		Fibrous Root		Storage Root		Crown		Old Wood	1-year Wood	
2011												
No Incorporation	41.02	Bare	Mulch	Bare	Mulch	47.1		Bare	Mulch	47.67	L	47.9a[w]
		51.8	52.9	45.2	49.5			40.6	44.5		M	47.4ab
				47.1	47.7			42.4	43.0		H	47.7b
	40.96	Bare	Mulch	Bare	Mulch	47.5		Bare	Mulch	47.5	L	47.8a
Incorporation		52.1	52.8	48.9	46.0			44.3	41.6		M	47.5ab
											H	47.1b
Significance												
Incorporation(I)	ns	ns[z]		ns		ns		ns		ns	ns	
Mulch(M)	ns	0.0066[y]		0.0017		ns		0.0254		ns	ns	
Nrate[x]	ns	ns		ns		ns		ns		ns	0.0073	
IxM	ns	ns		ns		ns		ns		ns	ns	
IxNrate	ns	ns		ns		ns		ns		ns	ns	
MxNrate	ns	ns		ns		ns		ns		ns	ns	
IxMxNrate	ns	ns		ns		ns		ns		ns	ns	
2012												
	C%	C%		C%		C%		C%		C%	C%	
		51.2		Bare	Mulch	L	47.3	47.3		46.6	48.6	
	40.47			38.2	42.5	M	46.8					
No Incorporation						H	46.7					
				L	43.7a							
				M	42.4ab							
				H	39.7b							
	40.49	51.1		Bare	Mulch	L	46.9	47.0		47.7	48.7	
				42.0	41.9	M	47.1					
Incorporation						H	47.1					
				L	44.7a							
				M	42.4ab							
				H	43.6b							
Significance												
Incorporation(I)	ns	ns		ns		ns		ns		ns	ns	
Mulch(M)	ns	ns		0.0118		ns		ns		ns	ns	
Nrate	ns	ns		0.0388		ns		ns		ns	ns	
IxM	ns	ns		ns		ns		ns		ns	ns	
IxNrate	ns	ns		ns		0.006		ns		ns	ns	
MxNrate	ns	ns		ns		ns		ns		ns	ns	
IxMxNrate	ns	ns		ns		ns		ns		ns	ns	

[z] ns=Non significant

[y] P-value if P ≤ 0.05

[x] “Nitrogen fertilizer rates in 2011 were 56 (“Low”), 168 (“Medium”), and 269 (“High”) (Table 1)

[w] Mean separation (in columns and rows) by LSD test at P ≤ 0.05 (lowercase)

Chapter 3: Carbon stocks and fluxes in a northern highbush blueberry field and their response to organic matter addition and nitrogen fertilization

Abstract

Woody perennial cropping systems potentially have carbon (C) stocks and dynamics that are more similar to less managed natural systems than to annual cropping systems. However, few studies have described C budgets for perennial systems or evaluated the effects of common management practices on C dynamics. This study reports total above and below ground C stocks and fluxes in the form of Net Primary Production (NPP), soil respiration, and management related imports and exports for a mature nine year old ‘Elliott’ northern highbush blueberry field in the Willamette Valley of Oregon. It also tested the individual and combined effects of organic matter incorporation at time of planting, periodic surface mulching, and nitrogen (N) fertilization on C allocation patterns and fluxes. Patterns of C allocation were assessed directly through destructive biomass sampling and allocated among the main standing stocks in the system: above and below ground blueberry plant parts, a perennial grass inter-row cover crop, soil below blueberry plants, and soil below the cover crop. The change in plant C allocation between 2011 and 2012 was used to estimate annual NPP. The blueberries in this study fixed an average of 588.8 g C m^{-2} y^{-1}. This is broadly comparable to values reported for naturalized woody shrubs in this ecoregion and for cultivated orchards in other temperate zones. Sixty percent of this NPP was allocated to woody plant parts that have long residency times in the system, and only 19% of NPP left the system in the form of harvested fruit. Seventy-six to 87% of the C in the system was found in the soil stock, with the largest soil C stocks found under the perennial inter-row cover cop. Inputs of organic matter in the form of pre-planting incorporation of sawdust and the

periodic application of surface mulch influenced the patterns of C fluxes and stocks in the system, although the effects varied over the two years that data were collected and treatments interacted with each other. Even nine years after its application, sawdust incorporation significantly increased fruit C stocks. Periodic mulching had more transient effects, primarily contributing to increased rates of soil respiration. The increased flux from the soil likely counterbalanced the organic matter inputs resulting in no observed significant effect of organic matter additions on total soil C stocks. Incorporation and periodic mulching did have an interactive effect on the total standing stock of C in the system with the largest stock of 9.7 $t*ha^{-1}$ C observed in incorporated un-mulched treatments. Nitrogen fertilization at the rates applied in this study did not influence C allocation or fluxes in the system. These results suggest that blueberry cultivation has a generally low direct C footprint. Management practices could reduce this footprint even further. Systems that incorporate organic matter during pre-planting can increase fruit yield while at the same time increasing the long-term C storage within fields. Initial sawdust incorporation can also reduce the need for periodic mulching that is both costly and associated with a large greenhouse gas (GHG) footprint and flux of C out of the system. The use of cover crops is also another strategy that significantly increases the soil C pool as well as providing a number of direct agronomic benefits such as in reduced soil erosion.

Introduction

Food systems contribute 19–29% of global anthropogenic greenhouse gas (GHG) emissions, the majority of which are a direct consequence of agricultural production practices such as land cover change and tillage (Vermeulen et al. 2012). Yet terrestrial ecosystems, including many agro-ecosystems, have the potential to store large amounts of atmospheric C, and enhancing this capacity can be an important tool for mitigating climate change (Heath et al., 1996; Lal 2008; Lal et al. 2003; Marland et al., 2001, 2003). Much of our understanding of terrestrial carbon dynamics comes from studies of relatively unmanaged systems such as forests and grasslands, while comparatively less is understood about intensively managed systems (Testi et al 2008; Keightley, 2011).

Most studies of carbon dynamics in managed farm land have been done in annual cropping systems (Paustian et al. 1995, 1997; Reicosky et al. 1997; Lal, et al., 1998; Morgan et al. 2010; Jia et al. 2012). This is understandable given the dramatic effect on global terrestrial C fluxes caused by the conversion of perennial dominated natural systems into annual agricultural production (Murty et al 2002). This conversion has caused a total of 50 to 100 billion tons of C to be depleted from the global soil organic C (SOC) pool (Jarecki and Lal, 2003; Alvarez 2005). Unlike forest and grassland ecosystems where SOC pools are in equilibrium, agro-ecosystems have SOC pools with higher C loses than gains (Jarecki and Lal, 2003). Agricultural ecosystems can lose up to 1500 g C m^{-2} in their SOC pools upon conversion from natural lands (Mann, 1986; Jarecki and Lal, 2003). Efforts to mitigate the GHG impact of annual crop production have focused on improving C storage in the soil by altering management practices (Lal et al. 1998, 2008; Jarecki and Lal, 2003). These strategies include: 1) minimizing soil disturbance with practices such as conservation tillage, crop rotation, and cover crops; 2) maximizing crop residues retained in soils by keeping prunings and other plant materials in the field; and 3) maximizing water and nutrient use efficiencies of crop production systems (Halvorson et al., 2002; Jarecki and Lal, 2003; Paustian, 2000).

In contrast to annual agro-ecosystems, woody agro-ecosystems potentially have standing C pools and fluxes that are more similar to natural forest and shrubland systems. Perennial cropping systems typically have considerably lower levels of soil disturbance compared to annual systems (Paustian et al. 2000). In addition, woody crops typically partition considerably less of their assimilated C to harvested plant parts (Cannell, 1980). As a consequence woody cropping systems have the potential to store considerable amounts of C over periods of years to decades in the woody biomass of the crop itself, as well as potentially having long term SOC stores that are more typical of non-cultivated forest and shrubland (Horwath, 2007; Kroodsma and Field, 2006). Recently, more interest has been shown in researching the C stored in the biomass of perennial agricultural crops such as orchards and vineyards (Keightley, 2011; Sheaffer, C. et al. 2005; Volk et al. 2004; Wu et al. 2012). Wu et al., (2012) found that net C sink in an apple orchard (*Malus domestica* Borkh.) ranged from 14 to 32 Tg*ha^{-1} C with a total C storage in biomass ranging from 230 to 475 Tg*ha^{-1} C between 1990 and 2010. This estimate was equal to 4.5% of China’s total net C sink in terrestrial ecosystems, demonstrating the potential that one perennial crop has on the C sink of a country and globally. However, overall we have a poor understanding of C allocation patterns and fluxes in woody cropping systems (Zanotelli et al. 2013). Woody cropping systems vary considerably in a number of important ways related to C allocation and dynamics (Montanaro et al. 2012). Our lack of understanding of this variation hampers our ability to more accurately describe the overall contribution of agriculture to GHG emissions and to develop management recommendations and appropriate incentives to enhance the C storage capacity of perennial agriculture.

Northern highbush blueberry (*Vaccinium corymbosum L.)* production systems are a good example of a perennial crop with potential for carbon storage in both plant and soil components. Blueberries are an important crop, with global blueberry production reaching 376,400 tons in 2010 from a planting area of 77,288 ha, with an average area growth increase of 7,058 ha*year^{-1} since 2005. Of total world blueberry area, 58% is located in North America, where 44,633 hectares (245,650 tons) are dedicated to highbush blueberry production (65% of global production) (Bañados et al., 2012; Brazelton, 2011). A number of factors suggest that blueberry systems could provide a net C store: 1) blueberry plants live for many years only reaching maturity eight years after planting, and mature fields can be over 35 years old; 2) blueberry production systems commonly incorporate perennial cover crops between rows; 3) there is typically no tillage after fields are established; 4) there is crop residue from annual prunings that are commonly chopped and left within fields as well as from the annual senescence of leaves (Scagel and Yang 2005; Strik, 2006; White, 2006; Yang 2002b). The magnitude of C losses associated with the crop and prunnings relative to carbon stored in other pools is not known for blueberry systems.

A number of other blueberry management practices could also influence C storage fluxes, but it is far from clear what these effects are. Nitrogen fertilization could potentially reduce the level of SOC in the system. Some studies have identified a long-term negative impact of nitrogen fertilization on SOC in annual systems, but in general, the effect of N on soil C dynamics are poorly understood because of the general lack of long-term data (Khan et al. 2007). Data from perennial systems are particularly scarce, but in at least some perennial systems nitrogen fertilization has a net positive long term effect on SOC (Fornara and Tilman 2012). Fertilization could also increase the storage of C in plant tissues by increasing plant growth. However, the effects of N fertilization on blueberry production are varied, with some studies reporting positive effects of increased fertilizer N on plant growth and yield (Hanson and Retamales, 1992), while others either reported no effect (Lareau, 1989, Nemeth et al., 2013), or a positive effect only at low levels (Bañados et al., 2012; Clark et al. 1998; Cummings 1978; Spiers, 1982).

Organic matter is often imported to blueberry production systems through its use as a soil amendment prior to planting or as a surface mulch. The goal of pre-plant amendment with organic matter is to increase soil organic matter content and maintain the appropriate soil pH level, around 4.5 to 5.5, for optimal blueberry growth (Hayes and Swift, 1986; Strik et al. 1993). The long-term fate of this added organic matter as well as its impact on the biological processes that drive C fluxes is not well understood. Incorporation of organic matter at the time of planting could enhance C storage in both above and below ground plant parts through its positive influence on soil fertility (Bhogal et al. 2009). However, studies evaluating the impact of pre-plant incorporation on blueberry growth have produced inconsistent results, with some studies reporting positive effects on plant growth and yield (Haynes and Swift, 1986; Moore, 1979; Spiers, 1982), while others reporting negative effects (Townsend, 1973; White, 2006). Surface mulches, such as sawdust, have been shown to increase plant growth, biomass and yield in highbush and rabbiteye blueberry relative to bare soil, consequently increasing the C mass stored in plant tissues (Chandler and Mason 1942; Haynes and Swift, 1986; Kozinski, 2006; Lareau, 1989; Magee and Spiers, 1995; Moore, 1979; White, 2006; Yadong et al., 2006). Amending the soil with organic matter also potentially contributes to SOC, but the bulk of these additions likely do not enter the long term SOC pool and instead leave agricultural system as a result of soil respiration (Wang et al. 2003; Bajoriene et al., 2013).

In this study I describe the magnitude of C fluxes (C NPP, and fruit and pruning exports) and pools within a typical blueberry production system. I also use a replicated long-term field experiment to test the individual and combined effects of incorporating organic matter at planting, surface mulching, and N fertilization on the C pools and fluxes within a mature blueberry production system.

Materials and Methods

Experimental site. This study was done in an 'Elliott' northern highbush blueberry (*Vaccinium corymbosum* L.) field established in Oct. 2003 at the North Willamette Research and Extension Center (NWREC; 45°16' 47.55” N and 122°45' 21.90” W), Aurora, OR. Standard sized (3.8 L; 2-year-old) container-grown plants were obtained from a commercial nursery. At the start of this study in 2011 the plantings were eight years old an age which is considered to be mature. Soil at the site was a Willamette silt loam (fine-silty mixed superactive mesic Pachic Ultic Argixeroll), with a pH of 5.4, and 4-6% organic matter content prior to planting.

Experimental design. Twelve treatments were arranged in a split plot design with pre-plant incorporation (with or without incorporation of Douglas-fir sawdust [*Pseudotsuga menziesii* Mirbel]*)* as main plots, and a combination of three nitrogen (N) fertilization rates (low, medium, and high) and two surface mulch treatments (bare soil and sawdust mulch) as the subplot treatments replicated four times. All treatments began during plant establishment in 2003-4, and continued for 9 years throughout this study. The four main plots consisted of two rows of plants each (one with pre-plant incorporation of sawdust and one without), and each subplot consisted of 20 plants. Subplots were separated by 3-m wide inter-rows planted in a perennial ryegrass (*Lolium perenne* L.) cover crop. Rows were spaced 3.1m apart, and plants were established 0.76m apart (4,300 plants/ha). A guard row was also planted on each side of the field. Plants were established on raised beds that were approx. 0.3m high (Figure 3-1).

Organic matter Incorporation (I). Just prior to planting (Oct. 2003) raised beds were constructed with or without incorporation of sawdust. Sawdust (60% 2 mm finer; C:N 790:1) was incorporated by applying in a strip 0.1 m deep and 0.4 m wide (141 $m^3 \cdot ha^{-1}$) centered down the length of each incorporated row (White, 2006). Nitrogen fertilizer (16–16–16) in the form of ammonium sulfate was added to each incorporated row at a rate of 45 $kg \cdot ha^{-1}$ of N to help facilitate decomposition of sawdust, a standard commercial practice (Eleveld et al., 2005; Julian et al., 2011; Strik et al., 1993). The sawdust and fertilizer were incorporated into the existing soil using a rototiller. Raised beds were constructed on incorporated and non-incorporated rows using a bed shaper. Rows without incorporated sawdust received the same rate of phosphorus (P) and potassium (K) as incorporated rows.

Surface Mulch (S). In treatments with a surface mulch, sawdust was applied to the top and sides of the raised beds to a depth of 5-8 cm (averaging 155 $m^3 \cdot ha^{-1}$) immediately after planting using a sawdust spreader, per standard commercial practice. New sawdust was added every few years, as required to maintain a mulch depth of approximately 7 to 8 cm (Strik et al., 1993). The most recent application of fresh sawdust mulch, prior to this study, occurred in Sept. 2010.

Fertilization. Plants were fertilized with a low, medium, or high rate of N (Nrate) with the rates per hectare increasing as the planting aged per standard commercial practice (Table 3-1); Hart et al., 2006). Every year since planting the total N fertilizer applied was divided into thirds and applied in mid-April (bloom), mid-May, and mid-June (green fruit). In 2011 and 2012, Urea (46–0–0) was used as a broadcast band applied to the in-row area, on top of the soil or sawdust mulch, if present. Phosphorus and K were applied annually, in spring, at a rate of 33 and 70 $kg \cdot ha^{-1}$, respectively, (0–22–25; P as triple superphosphate and K as potassium sulfate).

Irrigation and soil moisture. The planting was drip irrigated with a single line per row containing 3.8 $L \cdot h^{-1}$ emitters placed 0.15 m on either side of the base of the plant. The emitters were installed with spaghetti tubing attached, the end of which was inserted into the soil to a depth of about 6 cm. Inserting the tubing into the soil slightly, alleviated the problem of water run-off from the raised beds. The planting was irrigated based on weather and related crop evapotranspiration and soil moisture (see below) from 10 June to 27 Sept. 2011 and 14 May to 12 Oct. 2012.

Rows with pre-plant incorporation of sawdust were irrigated daily for 15 min six times per day for a total of 1.5 $h \cdot d^{-1}$ between 5:00 and 17:00 (79.8 $L \cdot plant^{-1} \cdot wk^{-1}$). Rows with no incorporation of sawdust prior to planting were irrigated daily for 13 min, six times per day, for a total of 1.3 $h \cdot d^{-1}$ between 5:30 and 17:30 (69.2 $L \cdot plant^{-1} \cdot wk^{-1}$). The difference in irrigation was required in order to maintain similar average soil moisture between the main plot incorporation treatments both at establishment and subsequently (White, 2006).

Data collected

Blueberry Plant Carbon Pool C storage within blueberry plant tissues was estimated with dry weight biomass and carbon concentrations for the following plant parts: fruit, senescing leaves, prunings of woody material, and the dormant plant including below ground structures. Blueberry fruit yield was obtained by harvesting the fruit using an over-the-row, rotary machine harvester (Littau Harvesters Inc., Stayton, OR) on 31 Aug., 7 Sept., and 19 Sept. 2011 and 15 Aug., 22 Aug., and 5 Sept. 2012. On each harvest date, fruit biomass data were collected on a total yield $plot^{-1}$ basis. Leaves were collected from a randomly selected plants within plots as they senesced in autumn 2011 and 2012 by placing a net around each selected plant when leaves first started changing color and collecting them once all leaves had senesced. Plants were pruned in Jan. 2011 and Dec. 2012, per standard commercial practice (Strik et al., 1993). More information can be found in Nemeth et al. (2013). Immediately after pruning, one dormant plant per plot was randomly selected for destructive harvest in each year. Plants were then divided into fibrous roots (< 1mm), storage roots (>1mm), crown, 1-year-old wood (last season's growth) and old wood growth.

All plant parts were dried to a constant weight at 60°C and the biomass of each plant part was estimated. Sub-samples from each plant part were removed and sent to Brookside Laboratories, Inc. (New Knoxville, OH) for analysis of C concentration (Kirsten, 1976). The tissue C concentrations (found in Table 2-9 Nemeth et al., 2013) were used to convert the biomass estimates to total C allocation within each plant part. The individual values for the dormant plant components, leaves, and fruit were summed to estimate total C per plant and total C per plant was multiplied by the blueberry density of the field (4600 plants per ha) to estimate total standing plant C in the experimental field.

Cover Crop Plant Carbon Stock

In November 2011 and 2012, above and below ground biomass of the ryegrass cover crop was harvested from within three randomly chosen 10 x10 cm quadrats. Quadrats were dug with a shovel to a depth of 10cm. Roots were washed and above and below ground samples were dried to a constant weight at 60°C, weighed and sent to Brookside Laboratories for analysis of C concentration (Kirsten, 1976). The tissue C concentrations (averaged at 32%) were used to convert the biomass estimates to an estimate of C stored within the cover crop on a per area basis.

Soil Carbon Stock

Soil C was estimated from samples taken from each plot in July of 2011 and 2012. Soil was sampled using four 30 cm deep, 5cm wide, cores spaced 50 cm from the crown of three randomly chosen plants in each plot. Cores were mixed and a sub-sample was sent to Brookside Laboratories, Inc. for analysis of organic, inorganic and total C concentration, organic matter, and bulk density (Nelson and Sommers 1996). Soil tons of C per hectare was calculated and averaged for each treatment using the "Equivalent Soil Mass Calculation" (Ellert et al., 2002). In October 2011 and 2012, soil samples were collected from underneath the cover crop grass (0 to 10 cm depth). The soil C stored from a 20-30 cm depth was estimated by taking a core from an uncultivated area adjacent to the study site. The values for both these depth were added to find the total soil C under the cover crop.

Mulch Carbon Stock

Surface mulch was randomly collected in July 2011 and 2012 from four locations within each mulch treatment plot in one randomly chosen replicate, making sure that all mulch layers from surface to soil were included. Each of the four plot sub-samples weighed approximately 40 g; these were mixed and a sub-sample was sent to Brookside Laboratories for analysis of carbon concentration and percent moisture (Kirsten, 1976). Mulch dry volume and C mass (tons/hectare) were calculated using the equations below:

$Equation\ 2.\ Mulch\ dry\ volume$

$$= \frac{Mulch\ wet\ volume\ \ m^3}{ha} - \left(\frac{\%\ Moisture\ *\ Mulch\ wet\ volume\ \ m^3}{100\ ha}\right)$$

$$Equation\ 3. \frac{Tons\ C\ Mulch}{hectare} = \frac{(Mulch\ dry\ volume\ \frac{m^3}{ha} * \frac{Mulch\ density\ kg}{m^3}) * \%\ C}{100}$$

Where the mulch density of 133 kg*m^{-3} was obtained from "Forest Products Measurements and Conversion Factors" (Briggs, 1994). Mulch wet volume was 1155 m^3*ha^{-1}.

Total standing stock of carbon

Since nitrogen fertilization rate was not found to significantly affect soil or plant C stocks estimates or the total standing carbon stock were pooled across nitrogen fertilization treatments. .Total standing stock of carbon was calculated for all the combinations of organic matter addition, (Incorporation x Mulch). Total field standing carbon stock was calculated asas.

Equation 4.

$$Total\ Field\ Carbon\ =$$

$$Total\ Plant\ Pool\ (Woody\ +\ Roots\ -\ Fruit\ \pm\ Leaves\ \pm\ Pruning) +\ Cover\ Crop\ Pool\ +$$

$$Soil\ \ Pool\ (SIC\ \ + SOC + Cover\ Crop)\ +\ Mulch\ pool$$

For the soil and cover crop pools, it is important to note that cover crop area amounted to about 70% of the field while blueberry soil area accounted for the remaining 30% (Figure 1); this was taken into consideration when calculating total field C.

Harvested fruits are a consistent yearly loss to the system and therefore were excluded from the standing stock total. For this study, pruning crop residues and senescent leaves were assumed to remain in the field and therefore were included in the standing stock estimate.

Carbon Net Primary Productivity

Carbon net primary productivity (NPP) for the field was calculated as:

$$Equation\ 6. Carbon\ NPP\ (g\ m^{-2}year^{-1}$$

$$= (2012\ Crown, fibrous\ roots\ and\ storage\ roots - 2011\ Crown, fibrous\ roots\ and\ storage\ ro$$

$$+ \left((2012\ Old\ wood\ +\ Old\ Wood\ pruning) - 2011\ Old\ wood\right)$$

$$+\ 2012\ 1yearold\ wood\ +\ 2012\ fruit\ +\ 2012\ leaf$$

C Losses due to fluxes such as herbivory and root exudates were not measured in in this study.

Statistical analyses

Analysis of all treatment effects on response variables (blueberry plant C stocks parts and total, and soil organic, inorganic and total C stock) were done as a complete factorial analysis of variance for a split plot design (incorporation as the main effect and mulch as the split plot effect) using the PROC MIXED procedure in SAS (SAS Institute, Cary, NC). The cover crop was not part of the experimental design so treatment effects on the C stocks within the cover crop and the cover crop soils were not tested. However, both of those pools were included in the calculation of total field C mass. There was a significant year effect on blueberry carbon allocation patterns so both years were analyzed separately in subsequent analyses. All analyses included checking data for homogeneity of variance; no transformation of data was required.

Results

Plant carbon stock. Organic matter incorporation at the time of planting and periodic mulching after establishment influenced patterns of standing C stocks within blueberry plants, but these effects varied considerably by plant part and by year and there were often interactions between treatments (Table 3-2). Blueberry plants that received organic matter incorporation tended to have increased total standing stocks of C in both 2011 and 2012 relative to plants that received no organic matter incorporation. However, this effect was not significant in either year (Table 3-2). Similarly, mulching had no significant main effect on total carbon or patterns of allocation within the plants in either year (Table 3-2). However, total plant C was influenced by a significant incorporation x mulch interaction in 2011 although not 2012. In 2011 the highest C mass was found in incorporated plots with bare soil (Table 3-2). In non-incorporated soils, plots with mulch had the highest plant C mass. There was 11.3% more C stored in plants grown on incorporated, non-mulched plots than incorporated mulched plots. Plots with no organic matter addition had the lowest plant carbon mass, with an average of 19.5% less C (kg m^{-2}) than incorporated plots with no mulch (Table 3-2).

Organic matter incorporation significantly increased carbon stocks in both fruit and prunings. There was an average of 0.01 kg m^{-2} more C stored in the fruit of plants grown with organic matter incorporation in both years. The effect of incorporation on carbon partitioned in prunings depended on mulching, but this interaction was apparent only in 2011 (Table 2). Nitrogen fertilization rate had no significant main effects on patterns of carbon partitioning, although it did have an interacting role with mulching and organic matter incorporation on the amount of carbon partitioned in leaves.

In 2011, 76% of total plant C mass was allocated to the dormant plant C pool, 10% to fruit, 10% to senescent leaves and 4% to prunings. In 2012, 66% of total plant C mass was allocated to the dormant plant, 12% to the fruit, 11% to senescent leaves and 11% to prunings.

Soil carbon stock. None of the treatments significantly affected total soil C under blueberry rows in either 2011 or 2012. Although not significant, organic matter incorporated plots did have an average of 1.1 kg m^{-2} more total soil C than non-incorporated plots (Table 3-3). This non-significant difference was driven largely by differences in SOC. In 2012, mulching and nitrogen fertilization had significant effects on SOC. Incorporated mulched plots had an average of 0.6 kg*m^{-2} more SOC than bare plots, however, in non-incorporated plots the difference between mulched and bare plots was only 0.01 kg*m^{-2} SOC per hectare (Table 3). Low rates of fertilizer N produced the highest SOC mass, whereas high rates lead to the lowest SOC mass in both incorporated and non-incorporated plots. Soil organic matter content in 2012 was significantly increased by incorporation and mulch (Table 3-3), with incorporated, mulched plots having the highest organic matter concentration. Similar patterns were observed in 2011 but the effects where not significant. Incorporation also tended to increase the SOC% (P = 0.0765), with an average of 2.1% SOC in incorporated plots, versus non-incorporated soils averaged 1.6%. Cover crop soil C amounted to 7.8 kg*m^{-2} tons of C in the first 30 cm depth and accounted for 75% of the total soil C stock, and 57% to 65% of the total field C stock without and with mulch, respectively.

Total field carbon. When the results of this study are scaled to an entire field and combined in terms of an integrated production system that includes an inter-row cover crop, a mature field with organic matter incorporation at time of planting would have 4-5% more total standing stock of C than a field without incorporation (Table 3-4). The allocation of C to different pools in the field was very similar across years and across incorporation treatments, changing mainly with the addition of mulch to the field (Figure 3-2).

Net Primary Productivity. Net primary productivity (NPP) of the blueberry plant averaged 589 g C m $^{-2}$ per year and was not affected by organic matter addition. Over 50% of C NPP allocation went to the dormant plant, about 19% of C NPP was allocated to fruit and to leaves, respectively (Figure 3-3). In terms of biomass, NPP averaged 1309 g*m^{-2} per year. Incorporated plots had an average of 105 g*m-^{2} more C NPP than non-incorporated plots, with an average C NPP of 496 g*m^{-2} in amended plots, although this was not statistically significant.

Discussion

This study reports the first quantitate estimates of C fluxes and allocation patterns within a mature blueberry production system, and it tested how typical management practices chronically applied over nine years influenced these patterns in a mature field.

The mature blueberries in this system had an average annual C NPP of 589 g C m^{-2} per year. This is very similar to the mean (±SE) annual NPP of 535 ±19 reported for naturalized *Rubus armeniacus* at a similar elevation in the same Willamette Valley Ecoregion (Law and Waring 1994). It is modestly below annual NPP values reported for hardwood deciduous trees, such as sweetgum *(Liquidambar styraciflua),* with C NPP averaging between 814 and 974 g*m^{-2} per year (Norby et al., 2002). In terms of biomass NPP, blueberries fall in the range of annual biomass NPP of 1160 - 1540 g*m^{-2} per year reported for a *Pseudotsuga menziesii* dominated forest at similar elevations within the ecoregion (Law and Waring 1994; Runyon et al. 1994).

The blueberries in this study also had NPP values that were modestly lower than those reported for woody tree crops such has apples that have reported C NPP values ranging from 786 - 960 g*m^{-2} per year (Panzacchi et al. 2012; Zantonelli et al., 2013). However, the annual NPP of the blueberries in this study was higher than reported values for annual cropping systems such as grain-corn, soybeans and small grain-cereals that ranged between 350 and 450 g*m^{-2} per year (Bolinder et al., 2007). A more pronounced difference between the blueberries in this study and other cultivated crops was in the allocation patterns of NPP. Annual crops generally have high harvest indices and 20-45% of the total annual crop NPP is removed from these systems in the form of harvested plant parts (Bolinder et al., 2007). The total C NPP reported for blueberries in this study was lower than the C NPP of 960 g*m^{-2} per year reported by Zantonelli et al. (2013) for apple orchards (*Malus domestica* Borkh.). However, in the apple system 49% of the annual NPP was taken away in the form of the harvested fruit (Zantonelli et al. 2013). In contrast to the allocation patterns reported for apples, much more of the blueberry NPP was stored in plant parts that have relatively long residence times. Sixty percent of NPP was allocated to woody tissues, with annual pruning potentially removing an additional 2% of NPP, although in many blueberry production systems these prunings are left within fields where some of the C likely enters the soil organic matter pool. Only about 19% of blueberry NPP is lost from the system through the harvested fruit. When these allocation differences are taken into account blueberry systems potentially store more C than orchards even though they have a lower overall NPP. Accounting for C loss from fruit, the apple orchard reported in Zantonelli et al 2013 retained a total of 441 $g*m^{-2}$ of C a year of NPP compared to 477 $g*m^{-2}$ of C a year retained in the blueberry field in this study.

The addition of organic matter into the system in the form of pre-planting incorporation of sawdust and the periodic application of surface mulch influenced the patterns of C fluxes and stocks in the system, although the effects were variable from year to year and the two management strategies interacted with each other. Overall, organic matter incorporation at time of planting had surprisingly long-term effects. Incorporating organic matter at the time of the 2003 planting significantly influenced the proportion of C stored in fruit nine years later. Incorporation as a main effect significantly influenced fruit C stocks although not total plant C stock. Since the fruit is lost each year this had a negative effect on the net C balance of the system. Of course, from a broader management perspective increased fruit yield and quality is the main goal of production. Indeed, since fruit made up a relatively small percentage of NPP, incorporation tended to have an overall positive effect on total field C stocks, with incorporated plots having an average of 3.9 t C ha^{-1} more than non-incorporated plots. Incorporation seemed to influence both plant and soil C stocks, either as an interaction with mulch or as a main effect influencing fruit and soil organic matter. Soil microbial activity tends to increase with organic matter incorporation potentially increasing the available C stocks in the soil by increased decomposition of the sawdust.

In contrast to pre-planting incorporation, the effects of periodic surface mulching appear to have little influence on patterns of C allocation or NPP and have little residence time in the system. Mulching represents a significant pathway for C flux through the system. In mulched fields the mulch made up about 13% of the total field C stock, more than the blueberry plants themselves (12%). Much of this organic matter appears to decompose relatively quickly releasing CO_2 into the atmosphere as microbial respiration that was reflected in a significantly higher recorded soil respiration value in the mulched vs. non-mulched plots, which was done once a year as an observational tool using Draeger tube methodology (Buyanovsky and Wagner, 1983). The mulching maintenance done in 2010 potentially caused a spike in soil respiration that was observed in 2011. In addition to acting as a direct source of GHG emissions, the harvest and transportation of the mulch also potentially involve considerable embedded GHG emissions. Reducing the frequency and amount of mulch applied may be a potential strategy for improving the overall carbon balance of blueberry production.

The results of this study suggest that mulching and pre-planting sawdust incorporation can interact to have some positive effects on plant growth. These interactions were most pronounced in 2011 the year after surface mulch was applied, with incorporated, bare plots having the highest plant C, while no addition of organic matter resulted in the lowest C stock, and too much organic matter (incorporation and mulch) resulted in lower C mass than in non-incorporated, mulched plots. The lack of this response in 2012, suggests that the residual effects of mulch on blueberry plants do not last longer than a year. Dormant plant C storage was also influenced by this incorporation by mulching interaction, which makes sense considering that about 65% of total plant C is contributed by the dormant plant.

There were no long-term direct effects of N fertilization on patterns of plant biomass and C allocation. This lack of response in blueberry plants appears to be related to the plant's ability to obtain sufficient nutrients from various sources (e.g. fertilization, mineralization of organic materials, the soil, and through association with mycorrhizae) and to the blueberries "buffering" capacity of the crown, roots, and old wood which serve as storage organs for N and other nutrients (Bañados, 2006). The lack of response to N fertilizer may have been related to the amount of available N in the soil pool which was not significantly affected by fertilization, but was decreased by the addition of sawdust mulch. Mulching also decreased other soil nutrients, although these were all found to be above recommended sufficiency levels (Nemeth et al., 2013).

As is typical for terrestrial systems, soil represented the largest pool of C in the blueberry field. The percentage of the soil C stock out of the total field C stock, varied between 76% and 87% depending if mulch was added to the system or not, respectively. It is important to note that the increase in soil C stock from 2011 and 2012 could be due to variability in the system and not necessarily due to an annual increase in soil C stocks. The cover crop soils contained significantly more C than blueberry soils and this pool represented 75% of the total standing soil stock of C in the system. Cover crop soil C stocks were the largest in the field representing between 57-65% of total field C stocks. The cover crop itself was a far smaller pool of C, representing between 1% and 2% of the total field C stock despite occupying 70% of the total field area. Although the cover crop in this study was periodically mowed, as is typical in most production systems the residues were left on site and the C and nutrients from these remain in the field and are largely retained within the system. Consequently, cover crops are a significant positive contribution to the overall C balance of the system. Including cover crops like *L. perenne*, can also provide other more direct agronomic benefits such as reducing soil erosion and improving overall soil fertility (Jarecki and Lal, 2003; Paustian, 2000).

Overall, the blueberry production system in this study had NPP and patterns of standing C stocks that are broadly similar to less managed perennial systems in western Oregon. Also, in contrast to other woody crops such as apples much more of the carbon that is fixed in the system is retained in relatively long-term stocks. Common management practices modify this overall picture somewhat. Organic matter incorporation at the time of planting has a positive influence on both crop yields that can offset the need for periodic mulching. The use of cover crops is essential for maintenance of soils with high OM, SOC, and total C storage. Encouraging both practices could reduce the C footprint of blueberry production and would also have additional direct benefits to farmers such as reduced costs associated with periodic mulching. However, a more comprehensive economic analysis is required. For instance, reduced mulching may involve increased costs for weed control and increase in irrigation needs? (Atucha et al., 2011; Korcak, 1988; Krewer et al., 2009; Moore, 1990; Savage and Darrow, 1942; Shutak and Christopher, 1952; White, 2006).

This study indicates that management practices can have long-term effects on system dynamics. There is a general need for longer studies that quantify C fluxes and allocation patterns in agricultural systems. This is particularly the case for woody perennial systems. Wu et al., (2012) found that orchards have a great potential for C sequestration, but that this storage is dynamic and changes throughout the life cycle of the apple trees. Carbon mass peaked in 18 year-old trees and then declined with age. This C life cycle, would not have been detected with a shorter term study. Once long-term studies are present and certain management practices are shown to increase C sequestration in crop fields, agricultural policy can embark into the direction of rewarding farmers for C storage (Merwin, 2010).

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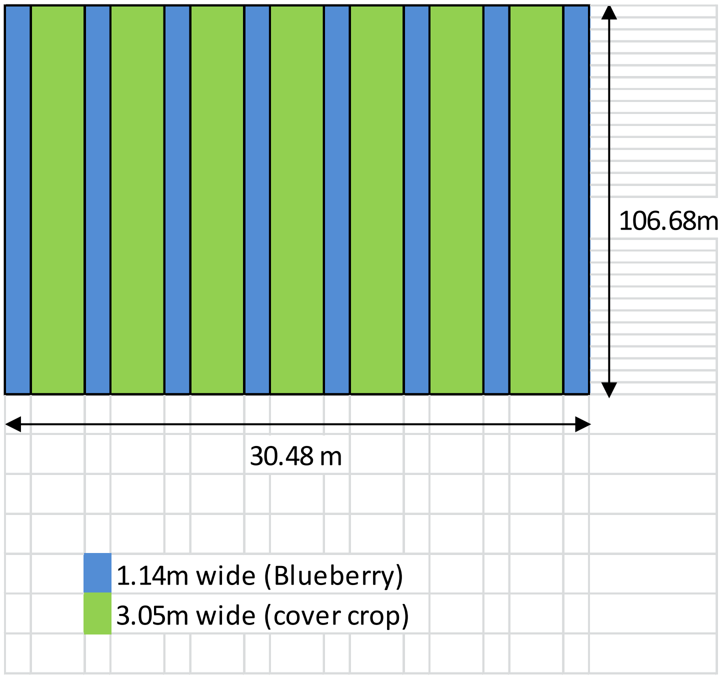
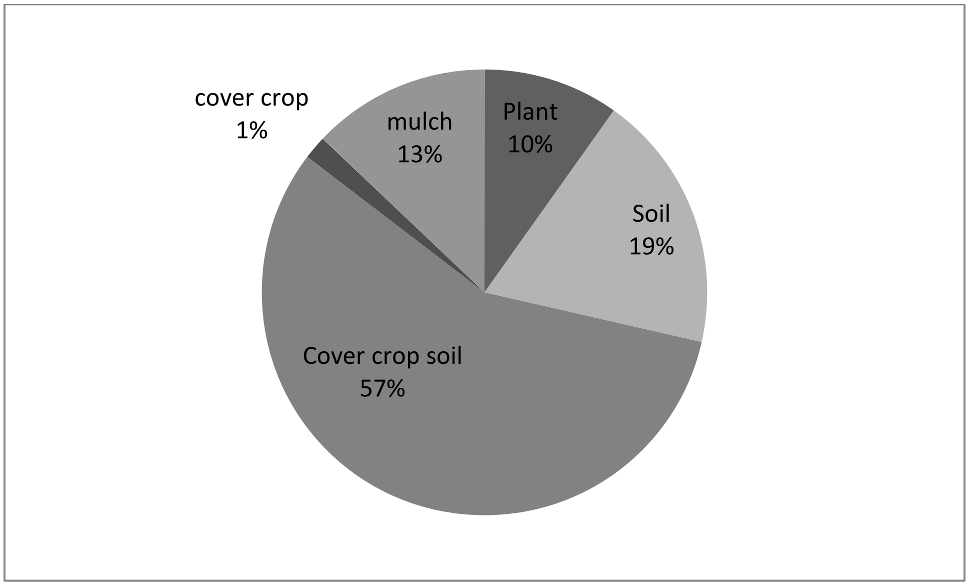


Figure 3-1. Experimental 'Elliott' blueberry field map (North Willamette Research and Extension Center, Aurora, OR) showing blueberry (*Vaccinium corymbosum* L.) and inter-row cover crop (*Lollium perenne* L.) surface area.

Mulched



Unmulched/Bare

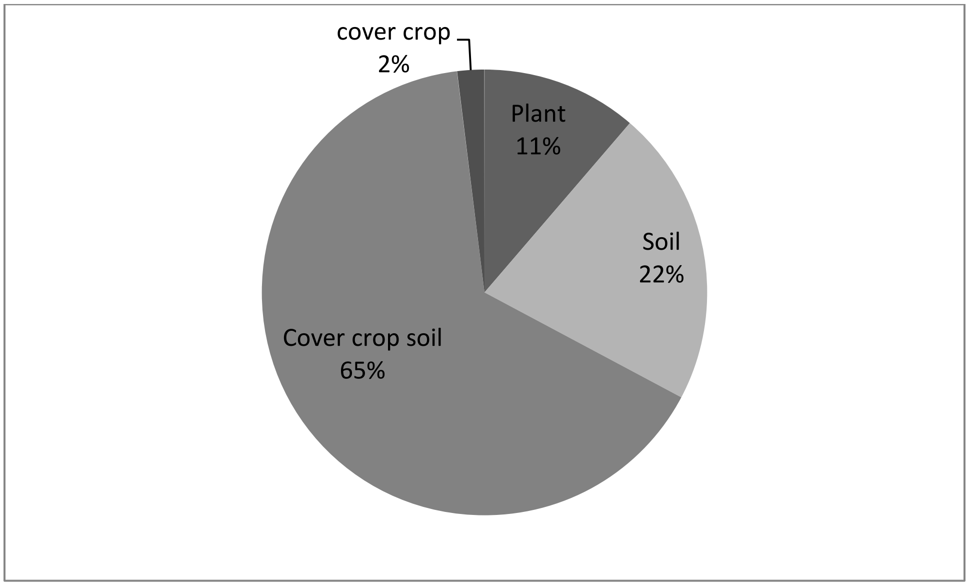


Figure 3-2. Effect of mulching on carbon stock allocations in an ‘Elliott’ blueberry field averaged by other treatments (incorporation and nitrogen fertilizer rate) and by year.

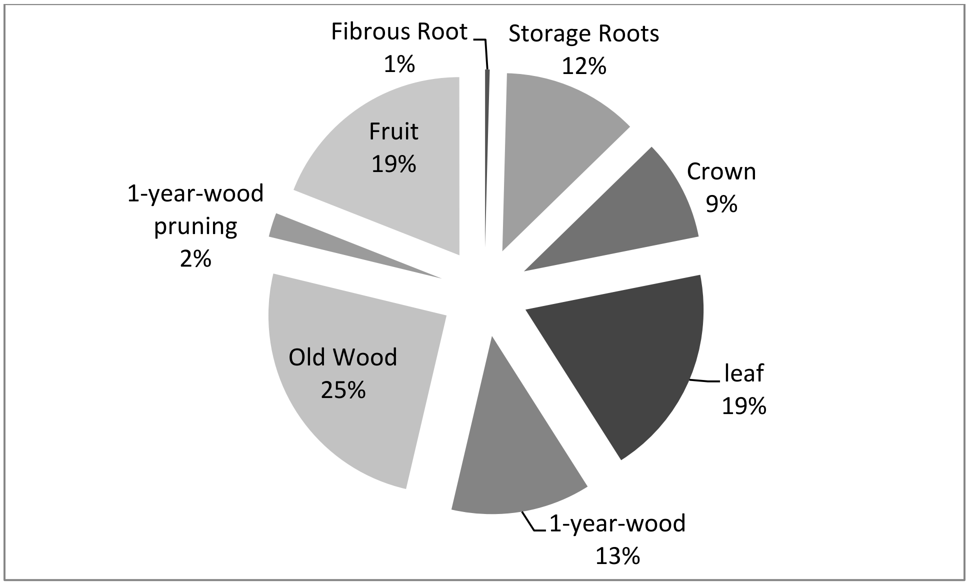


Figure 3-3. 2012 Carbon net primary production allocation (NPP) (%) to different plant parts of a mature 'Elliott' blueberry plant.

Table 3-1. Nitrogen (N) fertilizer rate treatments applied to 'Elliott' blueberry from the first (2004) through ninth (2012) growing seasons at the North Willamette Research and Extension Center, OSU.

	N Fertilizer Rate (kg N/ha)							
N treatment	2004	2005	2006	2007	2008	2009	2010-2012	Total
Low	22	22	31	31	39	39	56	352
Medium	67	67	395	395	112	112	168	1052
High	112	112	153	153	185	185	269	1707

Table 3-2. Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch (throughout planting life) and N fertilizer rate on the carbon mass of 'Elliott' blueberry plant parts in 2011 and 2012.

	Plant C Mass (Kg/m^2)									
Treatment	Dormant Plant		Fruit	Pruning		Leaf			Total[w]	
2011										
	Bare	Mulch		Bare	Mulch		B	S	Bare	Mulch
No Incorporation	0.53	0.64	0.11	0.08	0.10	L	0.08	0.08	0.88	0.97
						M	0.11	0.12		
						H	0.10	0.09		
	Bare	Mulch		Bare	Mulch		B	S	Bare	Mulch
Incorporation	0.71	0.60	0.12	0.13	0.10	L	0.12	0.11	1.09	1.02
						M	0.12	0.10		
						H	0.11	0.10		
Significance										
Incorporation(I)	Ns[z]		0.0055	0.018		0.0025			ns	
Mulch(M)	ns		ns	ns		ns			ns	
Nrate[x]	ns		ns	ns		ns			ns	
IxM	0.0075[y]		ns	0.0265		ns			0.0297	
IxNrate	ns		ns	ns		0.0306			ns	
MxNrate	ns		ns	ns		ns			ns	
IxMxNrate	ns		ns	ns		0.023			ns	
2012										
	0.81		0.11	0.03			B	S	1.07	
No Incorporation						L	0.09	0.09		
						M	0.09	0.12		
						H	0.14	0.09		
	0.91		0.12	0.06			B	S	1.18	
Incorporation						L	0.15	0.12		
						M	0.13	0.10		
						H	0.11	0.12		
Significance										
Incorporation(I)	ns		0.006	0.0319		ns			ns	
Mulch(M)	ns		ns	ns		ns			ns	
Nrate	ns		ns	ns		ns			ns	
IxM	ns		ns	ns		ns			ns	
IxNrate	ns		ns	ns		ns			ns	
MxNrate	ns		ns	ns		ns			ns	
IxMxNrate	ns		ns	ns		0.0314			ns	

[z] ns=Non significant

[y] $P \leq 0.05$

[x] "Nitrogen fertilizer rates in 2011 were 56 ("Low"), 168 ("Medium"), and 269 ("High") (Table 1)

[w] Total = dormant plant + prunnings + leaves

Table 3-3. Effects of pre-planting (2003) incorporation of sawdust, sawdust mulch (throughout planting life) and N fertilizer rate on the soil carbon mass of 'Elliott' blueberry field in 2011 and 2012.

	Organic Matter		Soil C (kg/m^2)			
Treatment	(%)		SOC		SIC	total C[v]
2011						
					L 1.67	
No Incorporation	3.2		3.9		M 0.50	4.89
					H 0.74	
					L 0.56	
Incorporation	4.3		5.1		M 0.50	5.98
					H 1.64	
Significance						
Incorporation(I)	**0.0065**[y]		ns		ns	ns
Mulch(M)	Ns[z]		ns		ns	ns
Nrate[x]	ns		ns		ns	ns
IxM	ns		ns		ns	ns
IxNrate	ns		ns		0.0253	ns
MxNrate	ns		ns		ns	ns
IxMxNrate	ns		ns		ns	ns
2012						
	Bare	Mulch	Bare	Mulch		
	3.0	3.2	4.5	4.6		
No Incorporation					1.52	6.10
			L 5.08a[w]			
			M 4.12b			
			H 4.45ab			
	Bare	Mulch	Bare	Mulch		
	3.74	4.6	5.6	6.2		
Incorporation					1.04	6.98
			L 6.4a			
			M 5.3b			
			H 6.1ab			
Significance						
Incorporation(I)	**0.0109**		ns		ns	ns
Mulch(M)	**0.0037**		0.0306		ns	ns
Nrate			0.006		ns	ns
IxM	**0.0311**		ns		ns	ns
IxNrate			ns		ns	ns
MxNrate			ns		ns	ns
IxMxNrate			ns		ns	ns

[z] ns=Non significant

[y] P ≤ 0.05

[x] "Nitrogen fertilizer rates in 2011 were 59 ("Low"), 169 ("Medium"), and 269("High") (Table 1)

[w] Mean separation (in columns and rows) by LSD test at P ≤ 0.05 (lowercase)

[v] Total = Soil organic carbon (SOC) + Soil inorganic carbon (SIC)

Table 4. Total carbon stocks in a mature 'Elliott' blueberry field in 2011 and 2012, and the effects of organic matter addition (pre-planting (2003) incorporation of sawdust and/or sawdust mulch (throughout planting life)) on total carbon storage of the field.

	Total C storage (t/ha)						
	Carbon Pools					Totals	
Treatment	Plant[1]	Soil [2]	Cover Crop Soil[3]	Cover Crop Plant[4]	Mulch[5]	Total 1[6]	Total 2[7]
2011							
Non-incorporated	8.31	14.55	54.30	1.64	±12.34	91.15	78.81
Incorporated	9.09	17.93	54.30	1.64	±12.34	95.30	82.96
2012	Plant[1]	Soil [2]		Cover Crop	Mulch[3]	Total 1[4]	Total 2[5]
Non-incorporated	9.60	18.31	54.30	1.64	±12.34	96.19	83.85
Incorporated	10.67	20.93	54.30	1.64	±12.34	99.88	87.54

[1] Total plant carbon pool (dormant plant + prunnings + senesced leaves)
[2] Total soil carbon pool (organic + inorganic) 30% of field soil
[3] Cover crop soil- 70% of field soil
[4] Cover crop plant -70% of field area
[5] Total mulch carbon pool (0 if no mulch treatment)
[6] Total field carbon stock with mulch pool
[7] Total field carbon stock without mulch pool

Chapter 4: Conclusions

In this research I evaluated how management practices in northern highbush blueberry production systems affect the C dynamics of blueberry plants and of the field as a whole. The first objective of this study was to find how organic matter addition (incorporation vs. mulch) and N fertilization rate affect plant biomass, growth allocation, C storage and mycorrhizal infection in roots. The second objective focused on how these management practices affect the total C stocks of the field, including the blueberry plants, the soil beneath these plants, the cover crop plant and soil, and the mulch, if present. Net primary productivity was also investigated as C flux and as biomass growth including all plant parts; the ones that remain in the field and on the plant and the ones that are removed, such as harvested fruit. The first objective was highly related to the second, because blueberry plants contributed to almost 12% of total C stocks in the field and were found to be the most responsive stock to the treatments studied.

Addition of organic matter was beneficial to the plant and to the soil. Plants in non-amended, bare (no organic matter added) plots were smaller and stored the least amount of C. Dormant plant dry weight and consequently dormant plant C mass were significantly affected by an incorporation by mulch interaction, where the largest plants were found in incorporated plots with bare soil. The effect of this interaction was also seen in soil pH and in soil ammonium-N and nitrate-N, where the highest values were found in incorporated mulched plots. These responses are believed to be linked to the short-term effect of the mulch maintenance done in 2010, the year before the study began. The year following the addition of fresh sawdust to the mulch layer, total plant responses and soil ammonium-N responses occurred; two years after the maintenance the interaction was no longer seen in plants and was only affecting nitrate-N in soils. It is important to note that too much organic matter, with both incorporation and mulch, resulted in lower plant dry weight and C mass than in non-incorporated, mulched plots, however, soil organic matter concentration was highest in these plots.

Incorporation as a main effect increased fruit biomass and C stocks. Fruit NPP contributed to about 20% of the total field's C flux. In soils, incorporation was found to increase the concentration of many soil nutrients, where as mulching as a main effect was found to decrease the concentration of phosphorus and potassium. Despite these responses, all nutrients in the soil were above sufficiency levels for blueberry. Mulching as a main effect also reduced fibrous root biomass, C mass and the percent of mycorrhizal infection found in these roots. Soil nutrients, soil pH and organic matter were unresponsive to nitrogen fertilizer rate. In addition, N fertilizer rate did not affect plant or soil total C stock, although in 2012 soil organic carbon was found to be reduced with higher rates of N fertilizer applied. Since soil nutrient concentrations were above recommended levels, it seems that despite the reduction in absorptive fibrous root biomass and the percentage of mycorrhizal infection, this did not affect the blueberry plant's ability to forage for nitrogen.

These results indicate that different management practices can have short- and long-term effects in the plant and in the field. Based on these results, I recommend sawdust be incorporated prior to planting, but no sawdust be added as a mulch. This reduction in organic matter use will save the farmer cost of transportation and maintenance of mulching. Also, in terms of reducing the field's greenhouse gas emissions (GHG), not having mulch eliminates the fossil fuel use of transporting it, and reduces soil respiration. The lack of response to N fertilization rate in both plant and soil stocks, also indicates that it is unnecessary to over-fertilize a blueberry field, which can lead to N leaching, and have financial and environmental costs associated with the production and distribution of fertilizers. The use of cover crops is recommended as we found approximately 57-65% of total field C to be stored in this stock. In terms of soils, 75% of total soil C stocks was under the perennial grass between the blueberry rows. Since 70% of the field is allocated to cover crop rows, the use of inter-row crops could also provide additional benefits to the field.

The results of this study and of others that looked at the C stocks and fluxes of perennial cropping systems can be used to quantify the contribution from various agricultural management practices to global carbon storage. Carbon stocks in these systems can mitigate GHG emissions, which in agricultural systems come from a wide range of agricultural practices and sources including initial land use change from forestry to agriculture, use of fossil fuels, crop burning, tillage, soil respiration etc. Currently, C policy lacks focus on C stocks compared to emissions; correcting policy through the research of C stocks in perennial agricultural systems could create incentives for carbon storage ecosystem service provision as well as encourage better farm stewardship by having this information available to farmers that are environmentally conscientious when it comes to their management practices.