

**Root restriction, under-trellis cover cropping, and rootstock modify vine size and berry composition of Cabernet Sauvignon**

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Vineyards in the Mid-Atlantic often have large, vigorous vines that can be costly to manage and produce inadequate fruit for wine production. Dense canopies increase the incidence of fungal disease, require greater allocation of resources to manage, and inhibit fruit development. The primary objective of these studies was to determine effective vine-size modification treatments that would optimize fruit quality, while reducing labor and chemical control. Research factors included root manipulation, under-trellis ground cover, and rootstock. Treatment levels were root bag (RBG) or no root manipulation (NRM); under-trellis cover crop (CC) or herbicide strip (HERB); and one of three rootstocks: 101-14, Riparia Gloire, or 420-A. Effects of these treatments were measured in two experiments: Experiment I compared combinations of all three treatments, while Experiment II explored the individual effects of root restriction using root bags of varying volumes. Root restriction consistently demonstrated the ability to reduce vegetative growth and vine water status. In the first experiment fruit-zone photosynthetic photon flux density (PPFD) was increased by 234% in RBG vines. Timed canopy management tasks indicated that RBG canopies required about half the labor time of NRM canopies. Anthocyanin concentration and total phenolic content were increased by 20% and 19% respectively in RBG fruit. CC increased fruit-zone PPFD by 62%, and increased soluble solids and color compounds. The 420-A rootstock reduced potassium uptake, resulting in lower must potassium concentration. Results demonstrated that these treatments significantly reduce vegetative growth in a humid climate, decrease management labor, and produce higher quality fruit.

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## **List of Abbreviations**

ABA – Abscisic acid  
AREC – Agricultural Research and Extension Center  
Ca – Calcium  
CC – Under-trellis cover crop  
CEFA – Cluster exposure flux availability  
CEL – Cluster exposure layer  
EPQA – Enhanced Point Quadrat Analysis  
GDD – Growing degree days  
HERB – Under-trellis herbicide strip  
ISE – Ion specific electrode  
K – Potassium  
LEFA – Leaf exposure flux availability  
LEL – Leaf exposure layer  
Mg - Magnesium  
N - Nitrogen  
NRM – No root manipulation  
OLN – Occlusion layer number  
OM – Organic matter  
PPFD – Photosynthetic photon flux density  
PQA – Point Quadrat Analysis  
RBG – Root bag  
RM – Root manipulation  
RR – Root restriction  
UTGC – Under trellis ground cover  
YAN – Yeast assimilable nitrogen

## **Introduction**

Abundant rainfall in the southeastern United States and other wet regions generates excessive vegetative growth in vineyards. The humid climate, in conjunction with high rainfall and relatively high organic matter (OM) soils, fosters a high vigor environment for grapevines which can be costly to control and detrimental to fruit and wine quality. Contemporary viticultural practices in Virginia and comparable climates aim to suppress vigor by utilizing appropriate rootstocks, cover crops, and/or alternative vine-size-modification methods without expensing yields and vine health. Root restriction (RR) has recently been well established as a mode of manipulating shoot vigor in many fruit species including: peach (Boland et al., 2000a; Boland et al., 1994), apple (Bar-Yosaf et al., 1988; Myers, 1992; Webster et al., 2000), cherry (Webster et al., 1997), mandarin (Mataa & Tominaga, 1998), and hybrid table grape cultivars (Wang et al., 2001; Yang et al., 2007; Yu et al., 2015; Yu et al., 2012; Zhu et al., 2006). In a preliminary study root restricted grapevines exhibited reduced growth rates, smaller leaf areas, and altered phenological development, relative to unrestricted root system vines (Hatch et al., 2011). The mechanisms for these altered growth habits likely relate to vine nutrient and water deficiencies due to the reduced root volume, but may involve altered hormone distribution between roots and aerial portion of the vine. Studies have demonstrated the limiting effects of root restriction on grapevine N-uptake and metabolism, (Yang, 2007; Zhu, 2006; Xiu-ming, 2015; Yu, 2012) regulation of growth and morphology (Wang, 1998, 2001; Xie, 2011), and improvement of fruit quality and anthocyanin composition (Wang, 2001; Wang, 2012, 2013) in certain hybrid varieties.

Our comprehensive study involved two field experiments that were established to evaluate the use of root restriction to suppress vegetative growth of Cabernet Sauvignon. Experiment I included a root-restriction treatment where vines were planted in 2006 within 0.015 m<sup>3</sup> fabric root bags. The objective was to compare the response of this restriction treatment with non-restricted vines, with the addition of two other variables: rootstocks (three levels) and vineyard ground cover management (two levels). Experiment II, installed in 2009, examined different sizes of root restriction bags (0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, 0.058 m<sup>3</sup>) compared with unrestricted vines. The effects on canopy growth and fruit components were measured to gauge an optimal rooting volume based on the ideal vine size response.

Although both experiments were previously designed and installed, none of the previously collected data has been systematically summarized. For the purposes of this M.Sc. thesis research, both experiments were intensively managed during the 2015 and 2016 growing seasons and summarized. Ultimately, the two seasons' data will be compiled with prior years' data for publication. This research has the potential to provide successful management strategies to growers aiming to optimize fruit and wine quality, while concurrently reducing labor costs. To our knowledge, this is the first long-term study exploring the use of root restriction bags and their effects on vegetative growth and fruit composition of *Vitis vinifera*.

## Literature Review

### Vigor: Sources and Management

Vigor describes the rate of shoot growth and elongation with respect to time over the course of the growing season. The degree of vigor can be affected by species, variety, rootstock, temperature, available water, nutrition, pruning/training method, and age. Higher levels of soil moisture and mineral nutrient availability, mainly nitrogen, can contribute to a significant rise in vigor. Soil moisture is presumably the predominant element driving excessive vigor in grapevines (Keller, 2015). The relationship between temperature and vigor is based on the concept of an increase in growth rate with increasing mean temperature until about 19°C, and decreasing above 30°C. This positive relationship is the primary reason for the calculation of growing degree days (GDD). GDD are expressed as a sum of average daily degrees above a base temperature from April 1 –October 31. However, because this relationship is not perfectly linear variations of this model have emerged. “Biologically effective degree days” (BEDD) describes the temperature summation between 10°C and 19°C due the flattening growth response above 19°C (Gladstones, 1992).

Vigor can be difficult to completely control due to the environmental factors that promote growth. However, there are canopy training and management techniques that can optimize the growing space. For instance, leaving an increased number of buds during dormant pruning can be an effective method of decreasing vigor. This provides more points of growth to distribute the energy stored in perennial tissue. Shoot removal early in the growing season and proper upward training through catch wires can ensure the canopies do not become unmanageable or unsatisfactory late in the season. Hedging and leaf removal mid-season are also effective tools

of promoting healthy, open canopies that will suppress disease pressure and promote fruit exposure.

## **Rootstocks**

Rootstocks (non-*vinifera*) are a vital part of the health of *V. vinifera* cultivars for wine-quality grape production due to their resistance to the soil pest phylloxera. Additionally rootstocks can alter scion growth fruit composition indirectly through differences in water and nutrient uptake. The choice of rootstock is based on its ability to withstand various adverse soil conditions, while concomitantly supporting proper growth and grape composition. The species of rootstock can be chosen in accordance with soils that are acidic, alkaline, waterlogged, or droughty. Rootstock choice can be very important in the context of controlling or enhancing vine vigor. Rootstock genotypes are also responsible for altering nutrient composition, described by the newly phrased “ionome”, in response to the variable N uptake of different rootstocks (Lecourt et al., 2015). Although the direct effects of rootstock on fruit composition are fairly negligible, the indirect effects from excessive vegetative growth can be more pronounced.

## **Cover Crops**

Intra-row and inter-row cover cropping systems have become standard practice in contemporary viticulture, providing a variety of benefits to grapevines and the surrounding ecology of vineyards. In particularly humid grape-growing regions such as the southeast United States, cover crops provide a biologically preferable and environmentally friendly method of

reducing vine vigor through competition for moisture and nutrients. Cover crops have been shown to reduce canopy size and improve canopy architecture, which can subsequently improve fruit exposure, fruit composition, and wine sensory evaluation in vigorous vines (Xi et al., 2011; Xi et al., 2010). In addition, cover crops provide a number of soil benefits including: reducing erosion, improving retention of soil organic matter, improving water infiltration, reducing leaching of nutrients, and improving soil structure and water holding capacity. The presence of cover crops also provides the added benefits of suppressing invasive weed populations and promoting biodiversity in the vineyard (Keller, 2015). Consequently, the competition for water may also correspond with unwanted mineral nutrient depletion from the vines. This competition for nutrients, particularly nitrogen, can result in reduced yeast-assimilable nitrogen (YAN) for must fermentation. Insufficient YAN levels often results in H<sub>2</sub>S production and off-aromas due to the lesser supply of organic amino acid supplied nitrogen. However, current ongoing research may reveal strategies using leguminous cover crops and timely N-additions to counteract the negative nutritional impact of cover cropping (Moss, 2016).

## **Root Restriction**

### *Effects on Growth and Development*

Root restricted plants exhibit consistently reduced shoot growth rates in fruit species: apple (Bar-Yosaf et al., 1988; Myers, 1992; Webster et al., 2000), peach (Boland et al., 2000a; Boland et al., 1994; Myers, 1992), mandarin (Mataa & Tominaga, 1998), cherry (Webster et al., 1997), grape (Wang et al., 1998; Wang et al., 2001; Yang et al., 2007; Yu et al., 2015; Yu et al., 2012; Zhu et al., 2006), mango (Zaharah & Razi, 2009), and watermelon (Liu & Latimer, 1995);

vegetable species: tomato (Hurley & Rowarth, 1999; Peterson et al., 1991; Shi et al., 2007), pepper (NeSmith et al., 1992), carrot (Thomas, 1993), and squash (NeSmith, 1993); and other plants: cotton (Thomas & Strain, 1991), coffee (Ronchi et al., 2006), and salvia (van Iersel, 1997). In addition, root-restricted plants have displayed smaller leaf areas, coinciding with reduced photosynthetic output (Hurley & Rowarth, 1999; Ismail & Davies, 1998; NeSmith, 1993; NeSmith et al., 1992; Ronchi et al., 2006; Shi et al., 2008; Thomas & Strain, 1991; Wang et al., 2001). Conversely, the biomasses of grapevine root systems under volume restriction were increased compared to control, specifically white adventitious and small fibrous roots with increased absorbing abilities (Yang et al., 2007; Yu et al., 2015; Zhu et al., 2006). Anatomically, the thickness of the cortex increased but the pericycle cross-section area was reduced in roots (Wang et al., 1998).

### *Cellular Morphology*

The main veins of grapevine leaves provide rapid-supply transportation of water and nutrients; and the minor veins act as a slow distributor of water and nutrients, while simultaneously serving as a collection network for assimilates (Canny, 1993). Mesophyll source cells in the leaves load photosynthetically produced sucrose into sieve elements (SE) and companion cells (CC), resulting in an osmotic gradient that generates a water potential gradient. This in turn triggers mass flow transport in the phloem by drawing water into SE from the xylem to create turgor pressure. The phloem sap will then move towards the sink to unload the sucrose and other solutes, causing a loss of water from the SE and a lower turgor pressure at the sink end. Therefore the water potential gradient creates movement in and out of the phloem, and turgor



pressure, generated by loading and unloading, drives movement within the phloem (Fisher & Oparka, 1996; Keller, 2015). Though the pathway of phloem loading has been disputed, phloem unloading happens both symplastically and apoplastically (Patrick, 1997). Phloem unloading predominantly happens symplastically via plasmodesmata, but later shifts to the apoplastic route via the cell walls around veraison (Zhang et al., 2006).

Xie et al. (2011) found a decrease in the size of sieve element and companion cells of main and minor veins in root-restricted leaves. The same study reported a significant increase in the number of plasmodesmata between phloem parenchyma (PP) and SE/CC, illustrating a greater potential for symplastic phloem loading. Similar findings show more plasmodesmata between SE/CC and PP at the unloading end of phloem transport in berry flesh tissue of root-restricted vines (Xie, Forney, et al., 2009). Fruit cells under root restriction had a denser cytoplasm with more mitochondria, endoplasmic reticulum, and vesicles, suggesting a relation to sugar accumulation. The reduction in lateral shoot growth during berry maturation may also affect photosynthate accumulation in fruit due to redistribution of soluble solids.

### *Mechanisms of Developmental Effects*

Physically, the confined root volume limits the plant available water and nutrients due to the inability of the root system to expand and find resources. Root length has been shown to correlate closely with leaf area (Petrie et al., 2006). Reduced leaf areas limit the production of plant biomass and growth by inhibiting the plant's ability to capture photosynthetic radiation. Reduction of photosynthetic activity is considered detrimental to nitrogen metabolism due to the reduced organic carbon and energy provided. The effect of restricting root volume on vine water

status is thought to induce many physiological changes, such as increased abscisic acid (ABA), that contribute to suppression of above-ground growth (Hurley & Rowarth, 1999; Ismail & Davies, 1998). The main role of ABA is in the regulation of plant water balance and osmotic stress tolerance. These functions are carried out through regulation of stomatal conductance and production of cell proteins for dehydration tolerance. Additionally, increased ABA can prevent auxin from loosening cell walls during growth. This increased ABA production by the roots seemingly simulates partial rootzone drying (PRD), wherein drying roots cause the hormonal response leading to stomatal closure and reduced shoot growth (Dry & Loveys, 1998). Typically this response coincides with lower amounts of cytokinins released into the xylem, inhibiting lateral shoot growth (Stoll et al., 2000).

Grapevines and other fruit-bearing higher plants depend on adequate nitrogen (N) as the most important mineral nutrient for growth and development of shoots, roots, and reproductive structures. The essential role of N is in the synthesis of amino acids, which can be further assembled into proteins required for vine health and function. Nitrogen assimilation into amino acids, nucleic acids, chlorophyll, and phytohormones occurs in the roots and leaves of grapevines.  $\text{NO}_3^-$  is the major inorganic form of N absorbed by the roots of higher plants, although plants are capable of taking up  $\text{NH}_4^+$  and amino acids in certain soil environments. Studies have shown the existence of three different uptake systems based on the concentration of available nitrate in the soil. Under high external  $\text{NO}_3^-$  supply (mM range), a low-affinity transport system (LATS) is activated. At low soil  $\text{NO}_3^-$  concentrations ( $\mu\text{M}$  range) there are two different active transport systems: a constitutive high-affinity transport system (cHATS) and an induced high-affinity transport system (iHATS) (Crawford & Glass, 1998). The HATS work in conjunction; cHATS has a higher affinity for  $\text{NO}_3^-$ , while iHATS has the greater capacity for

$\text{NO}_3^-$  uptake. Soil  $\text{NO}_3^-$  is actively absorbed into roots by way of an ATP pump that expels  $\text{H}^+$  ions into the soil, and subsequently cotransports back into the vine roots using a proton gradient. Following absorption,  $\text{NO}_3^-$  is reduced to  $\text{NO}_2^-$  through a nitrate reductase (NR)-catalyzed reaction and the resultant  $\text{NO}_2^-$  is reduced to  $\text{NH}_4^+$  using the catalyst nitrite reductase (NiR). Ammonium is then converted to glutamine by the catalyst glutamine synthetase (GS), and later into two molecules of glutamate by glutamate synthase (GOGAT), one of which is recycled back into the GS/GOGAT cycle. These primary amino acids can then be converted to other amino acids required for N storage, or used for vine growth and metabolism. Reserve N is essential for growth the following season, primarily between budbreak and bloom (Keller, 2015).

Recent studies on hybrid species of grapevines attribute the consistent reduction in above-ground vegetative growth under root restriction to the reduction in N uptake and metabolism, and subsequent adverse effects (Yang et al., 2007; Yu et al., 2015; Yu et al., 2012; Zhu et al., 2006). Zhu et al. (2006) found that  $\text{NO}_3^-$  concentrations were reduced in all plant parts, total N concentration in lignified roots and new leaves was decreased, and NR and NiR activities were significantly reduced in mature leaves and lignified roots in response to root restriction. Reduced  $\text{NO}_3^-$  uptake was also shown in response to root restricted peach (Ran et al., 1994). This reduction in NR and NiR activities suggests that root restriction inhibits the efficiency of nitrate reduction. GS activity has also shown substantial reduction in leaves and roots under root-restricted conditions; and nitrogen remobilization from leaves to roots following harvest was also reduced, leading to decreased reserve N (Yu et al., 2012). C:N balance within the vine has been suggested as the controlling factor in root-to-shoot partitioning and overall biomass allocation (Grechi et al., 2007). The reduced nitrate concentration in leaves of root-restricted vines suggests a diminished ability to transport  $\text{NO}_3^-$  from roots to leaves, thus

inhibiting leaf expansion, reducing leaf area, and adversely affecting photosynthetic capacity (Yu et al., 2015). Yang et al. (2007) reported that the uptake rates of HATS and LATS were enhanced by root restriction, while the affinity of the HATS was significantly reduced. Similar results were reported with root restricted maize, demonstrating increased N uptake rates, decreased shoot growth, and compensatory growth of existing roots (Xu et al., 2009). Bar-Tal et al. (1995) found that root-pruning reduced shoot growth and total N uptake, but increased flux N-uptake in the roots of tomato. These results demonstrate that root restriction limits the plant's ability to transport and reduce nitrate, and assimilate ammonium. Down-regulation of gene expression involved with nitrogen metabolism is closely related to the reduced metabolic efficiency in leaves under root-restriction. The auxiliary role of nitrate as a regulator of nitrogen metabolism gene expression suggests that reduced nitrate uptake has secondary effects on the latter metabolic activities (Yu et al., 2015).

However, while root restricted plants consistently show decreased N-uptake and metabolism, it is unclear whether the inhibition of N uptake is the cause or effect of reduced growth. It is more likely that growth controls metabolism, with vigorous plants spending resources abundantly and less-vigorous plant storing resources as reserves (Meyer et al., 2007). Therefore the metabolic responses observed with root-restricted plants may well be a result of reduced total biomass.

### *Fruit and Wine Quality*

Fruit composition can be substantially affected by the canopy microclimate and sun exposure, which is ultimately determined by the dynamics of vineyard layout, trellis design,

canopy height, architecture, density, and management (Reynolds & Vanden Heuvel, 2009). Particularly dense canopies can cause shading of inner leaves, which then senesce and recycle N, K<sup>+</sup>, and other nutrients to the fruit (Smart et al., 1988). The increase in K<sup>+</sup> in particular can raise juice pH due to its ability as a monovalent cation to substitute for protons. Shading around the fruit zone can slow phenological development, berry enlargement and sugar accumulation. Sun-exposed berries also produce less malate than shaded fruit due to the favored tartrate production and malate catabolism associated with higher temperatures (Pereira et al., 2006). Light exposure is necessary for anthocyanin accumulation and flavanol production, in response to visible light and UVB respectively (Keller, 2015). However, only a relatively low photon flux (~100 μmol m<sup>-2</sup> s<sup>-1</sup>) is necessary for anthocyanin development, above which temperature becomes the primary developmental factor. Cortell and Kennedy (2006) found that the increased accumulation of flavonols, anthocyanins, and proanthocyanidins in exposed Pinot Noir also coincided with increased extractability during fermentation. Unlike varieties such as Syrah, Nebbiolo, and Petit Verdot, Cabernet Sauvignon coloration appears to be more negatively affected by fruit shading (Keller, 2015). By contrast, very low vigor can result in diminished anthocyanin accumulation and even degradation due to inadequate leaf area, resulting in high UVB radiation and berry temperatures exceeding 35°C (Mori et al., 2007).

Moisture is the most significant soil-related factor in the grape and eventual wine quality, as opposed to soil type and parent rock. Increasing soil moisture leads to higher vigor, and thus denser canopies and more shaded fruit. These water-stimulated canopies, especially if accompanied by high nitrogen supply, can result in reduced fruit sugar, high acidity, low pH, and poor color development. Late-season water deficits can cause a decrease in berry size, thus creating a more concentrated solute composition and higher skin and seed:juice ratio. The

nutrient status of soils can also be a large component of fruit quality. As stated earlier, high nitrogen supply can cause excessive primary and lateral shoot growth, causing undesirably dense canopies and competition for assimilates (Keller et al., 1998; Keller & Koblet, 1995). High nitrogen uptake sometimes correlates with higher juice pH due to the concurrent high potassium uptake. The  $K^+$  cation's ability to neutralize the negative charges of nitrate and malate is presumably responsible for the correlated  $K^+$  uptake with  $NO_3^-$  uptake (Peuke, 2010). Low to moderate nitrate levels typically maximize phenolic production and have a direct effect on individual pigments, thus maximizing fruit coloration (Keller, 2015). Abundant nitrate supply suppresses specific secondary-metabolite genes, preventing the production of certain enzymes needed for phenolic accumulation.

Additionally, high vigor can indirectly increase the disease pressure of powdery mildew and bunch rots late in the season. Dense canopies limit the flow of air, thus creating high humid pockets in the canopy interior that allow fungal colonies to thrive. Shading effects of dense canopies on grape clusters also increase disease pressure. Temperatures above 35°C and direct UV radiation prevent development and colonization of fungal colonies, particularly in the case of powdery mildew (Keller et al., 2003).

Ideally the crop load of a vine, or yield:pruning weight, should be around 5-10 (Kliewer & Weaver, 1971). Lower ratios typically result from excessive vegetative growth, low yield, and possible indirect effects of vigorous canopies on fruit set and berry enlargement. On the other hand, higher ratios represent over-cropped vines with insufficient leaf area to mature fruit. Rootstocks can alter crop load through their ability to limit scion vigor and to some extent yield components. Fruit composition may also be slightly altered due to the variability in nutrient uptake among different rootstocks (Keller, 2015).

Root restricted table grapevines have demonstrated increased fruit set, color, and total soluble solids (Wang et al., 2001). The increased sugar accumulation of grapevines subjected to root restriction has been described by Xie, Li, et al. (2009), potentially as a result of increased acid invertase (AI) activity. Acid invertase operates within the vacuoles of cell walls, while neutral invertase (NI) occurs in the cytosol of cells. The invertases, along with sucrose synthase (SS) and sucrose phosphate synthase (SPS), are the enzymes primarily responsible for sugar accumulation and metabolism. Invertase and SS split sucrose into glucose and fructose once it reaches the grape berry.

Vines subjected to root restriction have also shown consistently higher total concentrations of anthocyanins than unrestricted vines (Wang et al., 2013; Wang et al., 2012). Anthocyanins are a class of flavonoids primarily responsible for the red, blue, and purple coloration in grapes and resultant wines. The composition and levels of individual anthocyanins vary among different grape cultivars and varieties, and the accumulation of these compounds can be modified by environmental factors and canopy management strategies. Anthocyanidins are stabilized by binding to one (3-glucoside) or two (3,5-diglucoside) glucose molecules, thus improving solubility and transforming into anthocyanins. The five main anthocyanidins are cyanidin, delphinidin, peonidin, petunidin, and malvidin. For red *V. vinifera* varieties, five to twenty anthocyanins are generally present, with malvidin-derived anthocyanins being the most abundant. Composition and content of anthocyanins, in combination with tannin and flavonol levels, ultimately determine color potential of a wine and color development during aging. Previous root restriction studies have demonstrated the increase in the number of individual anthocyanins, and notably higher percentages of delphinidin, cyanidin, and modified anthocyanidins (Wang et al., 2012). Furthermore, a follow-up study reported the genes

expressing the biosynthesis of anthocyanins were up-regulated in root restricted vines, explaining the enhanced anthocyanin profile (Wang et al., 2013).

Increased yield efficiency, fruit set, and in some cases quality, were reported in root restricted mango (Zaharah & Razi, 2009), mandarin (Mataa & Tominaga, 1998), peach (Boland et al., 2000b; Mandre et al., 1995), apple (Myers, 1992), and cherry (Webster et al., 1997).

Amelioration of exceedingly vigorous fruit-bearing plants using root restriction is viewed as an effective method of increasing efficiency and productivity per unit area, especially in the context of high-density planting (Bravdo et al., 1992).

The aim of our experiments is to manipulate the physiology of vines using different field treatments in order to produce a spectrum of vine sizes and evaluate the effects on fruit composition. Previous research on the same vines demonstrated that vine vegetative growth and water status would be altered (Hatch et al., 2011). The hypothesis of Experiment I was that vines with smaller canopies and more exposed fruit-zones would yield higher quality fruit (greater soluble solid accumulation, higher concentrations of anthocyanins and total phenolics). In Experiment II we hypothesized that there would be a positive linear response of canopy size to root volume. Additionally, fruit quality metrics would be improved with decreasing rooting volume.



## **Materials and Methods**

### **Objectives**

Research within the last two decades has established root restriction as an effective means of limiting above-ground growth while maintaining fruit quality and efficiency of many different fruit-bearing plant species. However, no studies to-date have investigated the use of root restriction on *Vitis vinifera* or any other wine grape species. Our primary objective was to study and evaluate the comprehensive effects of rooting restriction using fabric bags on canopy growth, canopy structure, plant nutritional status, components of yield, and fruit chemistry of *Vitis vinifera* cv. Cabernet Sauvignon. This research also studied the indirect effects of inhibited above-ground growth on canopy management labor time. The effects of root restriction were assessed in the presence of two separate under-trellis ground cover schemes (cover cropped or herbicide strip), and among three rootstocks (101-14, 420-A, or Riparia gloire).

Our secondary objective, explored in Experiment II, was to study different root bag volumes and their varying effects on the canopy growth and architecture, water status, plant nutrition, and fruit yield and chemistry. The purpose of this secondary project was to isolate the effects of rooting volume alone by keeping all other factors constant in order to determine the optimal rooting volume for Cabernet Sauvignon production.

### **Site and Weather**

Research was conducted exclusively at the Alson H. Smith Jr. Agricultural Research and Extension Center (AREC) experimental vineyard in Winchester, VA. Vineyard rows run

northeast/southwest on a 2% slope, at an approximate elevation of 300 m above sea level. The soil, generally described as a Frederick-Poplimento sandy loam, was specifically classified as a Poplimento-Hagerstown sandy loam in 2013 (Blackburn Consulting Services LLC) and has an estimated rooting depth of 0.75 to 1.50 meters. Meteorological data were collected from a weather station located within 100 m of the experimental vineyard.

## **Treatments and Design**

*Experiment I: Vitis vinifera* cv. Cabernet Sauvignon, clone ENTAV-INRA 337, vines were planted in 2006. These vines were planted 5-vines per panel spaced 1.5-m apart in 3-m wide rows. Vines were cordon trained, spur-pruned and trellised in a vertical shoot positioned (VSP) system. Inter-row cover crops were established prior to vineyard installation as a mix of tall fescue (*F. arundinacea*) and orchard grass (*D. glomerata*), managed with occasional mowing.

Experiment I was designed as a strip-split-split plot experiment and comprised three main effect treatments: under-trellis ground cover (2 levels), rootstock (3 levels), and root manipulation (2 levels). Experimental units were five-vine plots (7.5 m length) replicated in six blocks. Each block comprised the two under-trellis ground cover strips within the row length, the three rootstock subplots partitioned across three adjacent rows, and the two root manipulation techniques as sub-subplots. Border plots (7.5 m length) and border rows were used to spatially separate the six experimental blocks.

Rootstock treatments grafted to the Cabernet Sauvignon scion wood consisted of Riparia Gloire (*V. riparia*), 101-14 (*V. riparia*, x *V. rupestris*), or 420-A (*V. riparia* x *V. berlandieri*). In 2015 Riparia and 101-14 were the only rootstocks evaluated due the use of 420-A vines in a

separate experiment; all three rootstocks were assessed in 2016. Under-trellis ground cover treatments compared an 85-cm wide cover crop (CC) established in fall 2007, predominantly consisting of creeping red fescue (*F. rubra*) with some intermixed native weeds, with an herbicide strip (HERB) of the same width. Under trellis cover crops were maintained using a mechanical hand-held sickle-bar mower. Root volume manipulation treatments included root restriction bags (RBG) ( $\sim 0.015 \text{ m}^3$ ) (RootMaker, Stillwater, OK) and no root manipulation (NRM). Each combination of the three treatments was represented by a 5-vine panel (each 5-vine panel = experimental unit), which was replicated six times (one per block).

*Experiment II:* Vines were planted 4-vines per panel (each 4-vine panel = experimental unit) in 2009 (ENTAV-INRA 337) with identical spacing to Experiment I. Experiment II was designed as a randomized complete block, with each of the three treatments and the control represented in each of the four blocks. Plots were trained to a spur-pruned, VSP canopy on 101-14 rootstock. All plots were maintained using an 85 cm herbicide strip as the under-trellis ground cover. Treatment plots consisted of three different sized root restriction bags established at planting:  $0.026 \text{ m}^3$ ,  $0.035 \text{ m}^3$ , and  $0.058 \text{ m}^3$ , all compared to unrestricted control vines.

### **Plant Tissue Analysis**

In 2015, leaf petioles were collected at bloom and veraison and were analyzed for mineral nutrient levels (macronutrients and micronutrients). At bloom, 50 petioles were collected opposite an inflorescence amongst two plots of the same treatment in Experiment I (25 per plot; combining Blocks 1&2, 3&4, 5&6). Thus, each treatment was sampled in triplicate from the six block design. The petioles were collected evenly on each side of the canopies (East/West). In Experiment II, 40 petioles were pulled per individual plot using the same

technique. At veraison this process was repeated with mature, intact, and exposed leaves emerging from primary shoots above the top catch wire. In addition, whole leaf blade + petiole samples were also collected at veraison 2015 to provide a comparison of nutrient concentrations in the respective vegetative organs. In 2016, petiole samples were collected at veraison only using the same technique as the previous year. Samples were double rinsed with tap water and dried in an oven at 60°C prior to analysis at Waypoint Analytical Virginia, INC. (Richmond, VA).

### **Mid-day Stem Water Potentials**

Mid-day stem water potentials ( $\psi_{\text{md,stem}}$ ) were measured several times during the post-veraison period in Experiment II only. Two measurements were taken in 2015, and four measurements were taken in 2016. Foil bags were placed around two exposed primary leaves per panel between the first and second catch wires for at least an hour prior to  $\psi_{\text{md,stem}}$  measurements. When taking measurements, the bagged leaves were severed from the shoot and immediately placed in the chamber of the pressure bomb (Model 600 Pressure Chamber Instrument, PMS Instrument Co., Albany, OR). The chamber was then pressurized at a constant rate until xylem sap was observed bleeding from the stem. The pressure at the moment of sap exudation was recorded for each leaf and the chamber was depressurized.

### **Canopy Architecture**

At bloom and veraison the canopy architecture of each plot was assessed using Point Quadrat Analysis (PQA) (Smart & Robinson, 1991). PQA assessment was conducted at bloom when the only canopy management that had been performed was shoot thinning and positioning upright between catch wires. Using a measuring tape stretched across the length of an

experimental panel, a thin metal probe was then inserted into the canopy approximately every 30 cm (approximately 20 total insertions across the panel). Data was recorded for each insertion as a series of contacts representing leaves, clusters, or “gaps” in the canopy. Enhanced Point Quadrat Analysis (EPQA), and associated modelling software, was utilized to further study the level of sunlight exposure to the fruit (Meyers & Vanden Heuvel, 2008). To conduct these analyses, photosynthetic photon flux density (PPFD) was recorded using an AccuPAR ceptometer (AccuPAR80, Decagon Devices, Inc., Pullman, WA). Data were collected during clear skies within two hours of solar noon. An ambient PPFD was recorded in the row middle before every vine for each plot. The ceptometer was inserted into the fruiting zone of the canopy, parallel with the cordon/row orientation. East, vertical, and west measurements were recorded for each vine of the treatment plot and averaged. The percentages of average fruit zone-to-ambient PPF were then entered into the modelling software along with PQA data for subsequent analysis. All PPFD data are expressed as the % fruit-zone/ambient  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . Data were collected identically for both experiments.

### **Canopy Management**

Time trials were recorded for early-season canopy management in 2015 to determine how treatment impacted time to complete specific practices. Time measurements were taken on a per-panel basis in both experiments by the same individual. These trials were recorded for the first shoot hedging of the season and for leaf and lateral shoot removal around bloom.

Canopy management practices throughout the season included shoot thinning and positioning, fruit-zone leaf removal, lateral and basal shoot removal, and hedging. Shoot thinning was performed during the pre-bloom period while the shoots were approximately 20 to

40 cm in length. Shoot density was maintained at approximately 12-15 shoots per meter of cordon. Leaves, lateral shoots, and late-emerging basal shoots in the fruit zone were removed at the end of bloom as fruit began to set. Removal of vegetation at this time aimed to retain 1-2 leaf layers in the fruiting zone. Canopy height was managed using a mechanical hedger to hedge shoots above the top catch wire approximately every two weeks until growth ceased. Lateral shoots were trimmed as well throughout the growing season to prevent effects on fruit-zone light interception.

### **Components of Yield**

During 2015 and 2016 harvest yield weights and cluster counts were conducted in the field on a per-vine basis. Weights were measured using a calibrated hanger scale. Average cluster weights were then calculated from the values recorded. The number of berries per cluster was calculated from average cluster weight and average berry weight. Berry weights were assessed from 50-berry samples collected for primary chemistry analysis. Data collection was consistent with yield measurement in previous years.

### **Primary Fruit Chemistry**

At harvest, 50-berry samples were collected from all experimental units in Experiment I and II. Fresh samples were weighed for individual berry weights then hand-pressed for analysis of pH, titratable acidity (TA), and soluble solids (°Brix). Soluble solids were measured using a digital refractometer (Pocket PAL-1, ATAGO USA, Inc., Bellevue, WA) at room temperature. Titratable acidity was measured using an automatic titrator (848 Titrino Plus, Metrohm, Herisau, Switzerland) with 0.1 N NaOH to an endpoint detection of pH 8.2. Berry samples were collected in equal proportions among vines in each treatment panel for Experiments I and II.

## **Phenolics and Anthocyanins**

Separate 50-berry samples were collected from each plot and immediately frozen for future analysis. Berries were thawed and homogenized using an immersion blender. The homogenate was then separated into 1-g aliquots with 30 mL of 0.025 M KCl buffer (pH 1.0) and 0.4 M sodium acetate buffer (pH 4.5) added to each. The homogenate-buffer samples were shaken at 200 rpm for 20 min. and centrifuged for 5 min. at ~10,000 rpm. The supernatant was pipetted into a Macro Quartz Cuvette (14-958-116, Thermo Fisher Scientific Inc., Beverly, MA) with a path length of 10.0 mm, and the absorbance at 520 nm and 700 nm were measured in duplicate for anthocyanin concentration. Absorbance values were measured using the Genesis 8 ThermoSpectronic spectrophotometer (Cambridge, UK). Analysis by the pH differential method and calculation of anthocyanins in mg per g berry weight were performed as described in Lee et al. (2005).

An extra 0.5 g of the berry homogenate was combined with 30 mL of 0.025 M KCl buffer (pH 1.0) and was subjected to the same steps above, with absorbance measured in duplicate at 280 nm to determine total phenolic content.

## **Pruning Weights**

During the winter, the dormant canes were pruned and weighed in the field on a per-vine basis. These values were used to calculate the crop load (Yield weight : Pruning weight) and vine capacity (Yield weight + Pruning weight) for each vine. One-year-old canes were pruned such that 18-20 buds per vine were retained for the following year.

## **Juice Potassium Concentration and Yeast Assimilable Nitrogen**

Potassium ion concentrations were measured using an Orion Versa Star Advanced Electrochemistry Benchtop Meter (Thermo Fisher Scientific Inc., Beverly, MA) and connected potassium ion specific electrode (ISE) to record total  $K^+$  concentration in the unfermented grape juice. In 2015 samples prepared to a dilution factor of 10 with 2 mL of potassium strength adjuster (ISA). Samples in 2016 were diluted using a factor of 20 and 2 mL of ISA solution.

Total yeast assimilable nitrogen (YAN) was analyzed using Ammonia (Rapid) Assay and Primary Amino Nitrogen Assay kits (Megazyme International Ireland Limited; Wicklow, Ireland). Samples in 2015 were measured using the manual assay procedure using Fisherbrand Semimicro Methacrylite Cuvettes (Fisher Scientific; Fair Lawn, NJ) with a path length of 10.0 mm. Absorbance values were measured using the Genesys 10S UV-Vis spectrophotometer (Thermo Fisher Scientific Inc., Beverly, MA). In 2016 samples were analyzed using the microplate assay procedure in Corning UV-Transparent 96-well plates (Fisher Scientific, Fair Lawn, NJ). Absorbance measurements were taken at 340 nm and recorded using the Synergy H1 microplate reader (Bitotek; Winooski, VT).

## **Statistical Analysis**

Experiment I: Data were statistically analyzed using JMP Pro ver. 11 (SAS Institute, Cary, NC). Factorial analysis of variance (ANOVA) models were constructed and analyzed using standard least squares and an emphasis on effect leverage. Root manipulation (RM), under-trellis ground cover (UTGC), and rootstock were evaluated for each data set. Blocking was included as a possible model effect. Interactions between these effects were also considered and often needed to produce appropriate statistical tests. All tests assessed response variables for



significance at a 95% confidence level ( $\alpha = 0.05$ ). Significance ( $p > F$ )  $< 0.05$  were reported for each model effect and interaction, or listed as not significant (NS). Using the analysis of the fixed effect model, the separation of means was evaluated using Student's T-test (for pairwise comparisons of least square means) or Tukey's HSD (comparing all least square means). Linear regression analyses were used to evaluate the correlation of berry weight, crop load, vine capacity, and PPF as an indicator for color absorbance data (anthocyanin concentration and total phenolics). Further linear regression analysis assessed the value of veraison petiole K% as an indicator of juice K concentration.

Experiment II: Data were statistically analyzed using JMP Pro ver. 11 (SAS Institute, Cary, NC). One-way ANOVA was computed for the complete randomized block design for all response variables. Response variables included plant tissue analysis, EPQA and fruit-zone parameters, pruning weights, crop load, vine capacity, canopy management labor, primary chemistry, secondary chemistry, components of yield, and mid-day stem water potentials. All tests assessed response variables for significance at a 95% confidence level ( $\alpha = 0.05$ ). Significance ( $p > F$ )  $< 0.05$  were reported for treatment and block, or listed as not significant (NS). Separation of means was evaluated using Tukey's HSD (comparing all least square means).

### **Soil Nutrient Applications**

In both experiments nitrogen was applied at 11.2 kg N/ha among all treatments in the form of calcium nitrate ( $\text{Ca}(\text{NO}_3)_2$ ) at bloom and veraison during both years. In 2015 phosphorus ( $\text{P}_2\text{O}_5$ ) and magnesium were applied post-harvest at 83 kg/ha and 103 kg/ha respectively.

## Results

### Weather Metrics

Accumulated growing degree days (GDD) from April to October were comparable in 2015 and 2016 (Table 1). The early growing season (April-June) in 2015 was 131 GDD higher than 2016 (Figure 1). However mean temperatures were notably higher in 2016 during the phenological ripening months from July through September (Table 1). Cumulative rainfall was 82 mm greater in 2016, a 14% increase in precipitation during the active growing season compared to 2015. Precipitation during the ripening months (July-September) was 100 mm higher in 2016 (Figure 2).

Table 1. Seasonal temperature and rainfall data recorded at AHS Jr. AREC (Winchester, VA) experimental vineyard 2015-2016.

	<b>Bud Break Date</b>	<b>Harvest Dates (RBG, NRM)<sup>a</sup></b>	<b>GDD (base 10 °C) Apr-Oct</b>	<b>Precipitation (mm) Apr-Oct</b>			
2015	4-22	9-24, 10-6	2014	569.9			
2016	4-23	10-2, 10-13	2026	651.8			
<b>Mean Temperature (°C)</b>							
	<b>April</b>	<b>May</b>	<b>June</b>	<b>July</b>	<b>August</b>	<b>September</b>	<b>October</b>
2015	11.4	17.2	20.0	21.3	20.7	18.4	12.5
2016	12.1	15.5	21.8	24.4	24.4	21.1	14.3

<sup>a</sup> RBG = root bag, NRM = no root manipulation

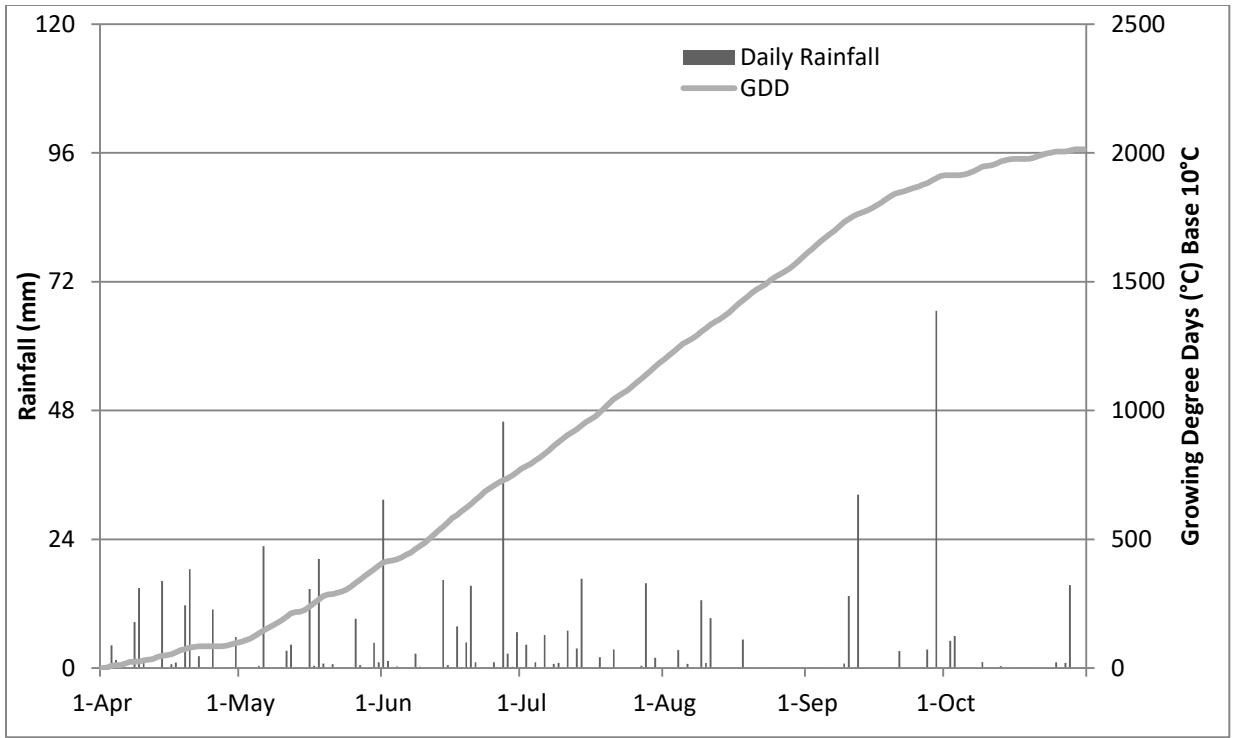


Figure 1. Daily rainfall and growing degree days (base 10°C) recorded at the Alson H. Smith Jr. AREC in Winchester, VA from April through October in 2015.

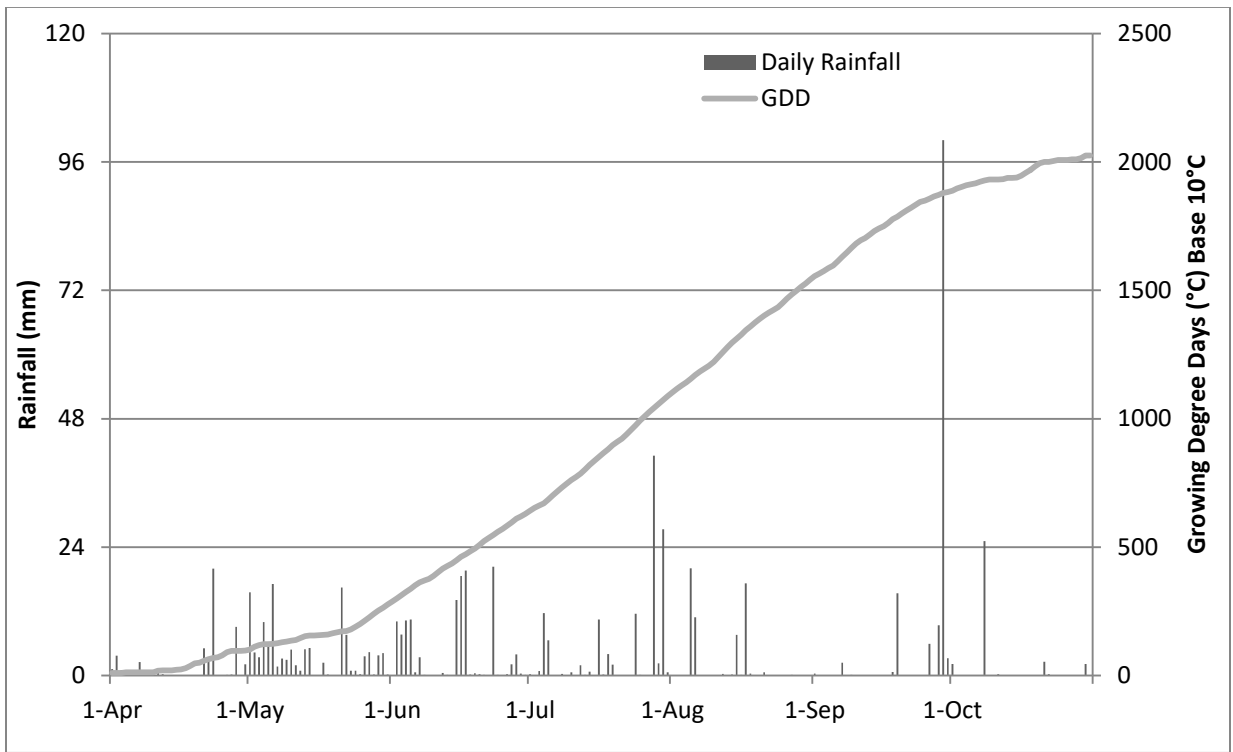


Figure 2. Daily rainfall and growing degree days (base 10°C) recorded at the Alson H. Smith Jr. AREC in Winchester, VA from April through October in 2016.

## **Experiment I**

### *Plant Tissue Analysis*

Nitrogen concentrations of grapevine petioles at veraison were reduced in RBG vines in 2015 and 2016 (Tables 2 & 6); however leaf blade + petiole nitrogen concentrations at veraison in 2015 and 2015 bloom petiole concentrations were not reduced compared with NRM vines (Tables 2 & 3). Potassium concentrations were reduced in RBG petioles at veraison 2015 and 2016, bloom 2015, and in leaf blade + petiole at veraison 2015 compared to NRM (Tables 2, 3, and 6). Conversely, magnesium concentrations were increased in petioles at veraison 2015 and 2016, and in leaf blade + petiole at veraison 2015 by the RBG treatment (Tables 2,3, and 6). Sulfur concentrations in petioles were reduced by the RBG treatment at veraison in 2015 and 2016 (Tables 2 & 6). Calcium concentrations were increased in RBG vines in veraison petioles in 2016 (Table 6).

There were no significant differences due to UTGC in the nitrogen concentrations of veraison plant tissues collected in 2015 and 2016 (Tables 2 & 6). Magnesium concentrations were reduced in petioles of CC vines at bloom 2015, veraison 2015 and 2016, and leaf blade + petiole at veraison 2015 (Tables 2,3, and 6). Potassium and calcium concentrations were reduced by CC in 2015 veraison petioles (Table 2).

Sulfur concentrations were highest in the 101-14 rootstock in petioles at bloom 2015, veraison 2015 and 2016, and leaf blade + petioles in 2015 (Tables 2,3, and 6). Calcium concentrations were highest in the Riparia rootstock in veraison petioles and leaf blade + petioles in 2015 (Tables 2 & 3). Phosphorous, magnesium, and calcium concentrations were all highest

in the 420-A rootstock in veraison 2016 petioles (Table 6). Conversely, potassium concentrations were lowest in the 420-A rootstock in veraison 2016 petioles (Table 6).

Table 2. Treatment effects on macronutrient concentrations (% dry weight) in petioles at bloom and veraison in 2015.

Treatment <sup>ab</sup>	Bloom						Veraison					
	N	S	P	K	Mg	Ca	N	S	P	K	Mg	Ca
<b>RM:</b>												
NRM	0.62 a	0.29 a	0.51 a	3.34 a	0.42 a	2.36 a	0.44 a	0.20 a	0.15 a	5.71 a	0.30 b	1.74 a
RBG	0.67 a	0.21 b	0.30 b	2.56 b	0.31 b	1.99 b	0.40 b	0.17 b	0.11 b	4.31 b	0.46 a	1.82 a
<b>UTGC:</b>												
Herb	0.73 a	0.28 a	0.41 a	2.75 b	0.40 a	2.18 a	0.44 a	0.19 a	0.15 a	5.30 a	0.43 a	1.87 a
CC	0.56 b	0.22 b	0.40 a	3.14 a	0.33 b	2.17 a	0.40 a	0.18 a	0.11 a	4.73 b	0.33 b	1.68 b
<b>Rootstock:</b>												
101-14	0.65 a	0.27 a	0.42 a	3.19 a	0.44 a	2.14 a	0.43 a	0.20 a	0.14 a	5.10 a	0.40 a	1.51 b
Riparia	0.64 a	0.23 b	0.39 a	2.71 b	0.29 b	2.22 a	0.41 a	0.16 b	0.12 a	4.93 a	0.36 a	2.05 a
<b>RM*UTGC:</b>												
NRM-Herb	0.73 a	0.32 a	0.53 a	3.15 b	0.44 a	2.34 a	0.45 a	0.20 a	0.13 ab	6.21 a	0.36 bc	1.93 a
NRM-CC	0.52 c	0.27 b	0.49 a	3.54 a	0.39 b	2.38 a	0.43 ab	0.20 ab	0.17 a	5.22 b	0.24 c	1.56 b
RBG-Herb	0.74 a	0.24 b	0.33 b	2.36 d	0.35 b	2.02 b	0.42 ab	0.17 bc	0.10 b	4.38 bc	0.50 a	1.82 a
RBG-CC	0.60 b	0.18 c	0.28 b	2.75 c	0.27 c	1.97 b	0.37 b	0.16 c	0.12 ab	4.24 c	0.42 ab	1.81 a
<b>Significance:<sup>c</sup></b>												
Block	0.0280	NS	0.0003	0.0327	<0.0001	0.0089	NS	NS	NS	NS	NS	NS
RM	NS	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0411	0.0001	0.0396	<0.0001	0.0003	NS
UTGC	<0.0001	<0.0001	NS	0.0007	<0.0001	NS	NS	NS	NS	0.0224	0.0134	0.0057
Rootstock	NS	<0.0001	NS	0.0001	<0.0001	NS	NS	<0.0001	NS	NS	NS	<0.0001
RM*UTGC	NS	NS	0.0228	NS	NS	NS	NS	NS	NS	NS	NS	0.0090
RM*Rootstock	NS	0.0114	<0.0001	0.0024	<0.0001	NS	NS	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	0.0001	<0.0001	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	0.0187	NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 3. Treatment effects on macronutrient concentrations (% dry weight) in petioles + leaf blades at veraison in 2015.

Treatment	Veraison					
	N	S	P	K	Mg	Ca
<b>RM:</b>						
NRM	1.92 a	0.28 a	0.16 a	2.54 a	0.21 b	2.05 a
RBG	2.04 a	0.26 a	0.14 a	1.92 b	0.29 a	1.96 a
<b>UTGC:</b>						
Herb	1.99 a	0.28 a	0.15 a	2.26 a	0.27 a	2.11 a
CC	1.97 a	0.26 a	0.15 a	2.20 a	0.23 b	1.90 a
<b>Rootstock:</b>						
101-14	1.96 a	0.29 a	0.16 a	2.37 a	0.28 a	1.86 b
Riparia	2.00 a	0.24 b	0.14 a	2.09 a	0.23 b	2.15 a
<b>RM*UTGC:</b>						
NRM-Herb	1.92 a	0.29 a	0.15 a	2.57 a	0.23 ab	2.14 a
NRM-CC	1.93 a	0.27 a	0.16 a	2.52 a	0.19 b	1.95 a
RBG-Herb	2.06 a	0.27 a	0.15 a	1.94 a	0.31 a	2.08 a
RBG-CC	2.02 a	0.25 a	0.14 a	1.89 a	0.27 a	1.84 a
<b>Significance:</b>						
Block	NS	NS	NS	NS	NS	NS
RM	NS	NS	NS	0.0055	0.0010	NS
UTGC	NS	NS	NS	NS	0.0468	NS
Rootstock	NS	0.0062	NS	NS	0.0207	0.0265
RM*UTGC	NS	NS	NS	NS	NS	NS
RM*Rootstock	NS	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 4. Treatment effects on micronutrient concentrations in petioles at bloom and veraison in 2015.

Treatment <sup>ab</sup>	Bloom							Veraison						
	Na (%dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)	Na (%dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)
<b>RM:</b>														
NRM	0.05 a	35 a	61 a	250 a	51 a	14 a	22 a	0.05 a	31 a	47 b	210 a	35 a	9 a	26 a
RBG	0.05 a	25 b	50 b	197 b	37 b	11 b	17 b	0.05 a	26 b	56 a	223 a	35 a	7 b	23 a
<b>UTGC:</b>														
Herb	0.05 a	29 b	52 b	202 b	41 b	13 a	19 a	0.05 a	29 a	52 a	215 a	36 a	9 a	22 a
CC	0.05 a	31 a	58 a	245 a	47 a	12 b	20 a	0.05 a	27 a	51 a	218 a	34 a	7 b	27 a
<b>Rootstock:</b>														
101-14	0.05 a	32 a	63 a	245 a	46 a	13 a	21 a	0.05 b	27 b	52 a	231 a	36 a	8 a	25 a
Riparia	0.05 a	29 b	47 b	203 b	42 a	12 b	18 a	0.06 a	29 a	51 a	202 a	34 a	8 a	24 a
<b>RM*UTGC:</b>														
NRM-Herb	0.05 a	34 b	58 ab	221 b	45 b	14 a	20 a	0.05 a	33 a	49 ab	225 a	37 a	10 a	23 a
NRM-CC	0.05 a	37 a	63 a	280 a	57 a	14 a	23 a	0.05 a	29b	46 b	194 a	33 a	8 ab	29 a
RBG-Herb	0.05 a	25 c	47 c	183 b	37 b	12 b	18 a	0.05 a	25 b	55 ab	205 a	35 a	8 ab	22 a
RBG-CC	0.05 a	25 c	53 b	211 b	37 b	10 c	17 a	0.06 a	26 b	57 a	241 a	36 a	7 b	24 a
<b>Significance:<sup>c</sup></b>														
Block	NS	0.0136	NS	NS	NS	NS	NS	NS	NS	NS	0.0180	NS	NS	NS
RM	NS	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0188	NS	<0.0001	0.0029	NS	NS	0.0032	NS
UTGC	NS	0.0106	0.0026	0.0004	0.0152	0.0149	NS	NS	NS	NS	NS	NS	0.0068	NS
Rootstock	NS	0.0001	<0.0001	0.0005	NS	0.0435	NS	0.0014	0.0439	NS	NS	NS	NS	NS
RM*UTGC	NS	0.0316	NS	NS	0.0152	NS	NS	0.0396	0.0067	NS	NS	NS	NS	NS
RM*Rootstock	NS	0.0011	NS	NS	NS	NS	0.0491	NS	0.0210	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)



Table 5. Treatment effects on micronutrient concentrations in petioles + leaf blades at veraison in 2015.

Treatment <sup>ab</sup>	Veraison						
	Na (%dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)
<b>RM:</b>							
NRM	0.04 a	31 a	60 b	452 a	89 a	11 a	27 a
RBG	0.04 a	22 b	71 a	452 a	78 a	9 a	23 a
<b>UTGC:</b>							
Herb	0.04 a	27 a	65 a	463 a	80 a	10 a	25 a
CC	0.04 a	26 a	66 a	441 a	87 a	10 a	26 a
<b>Rootstock:</b>							
101-14	0.04 a	27 a	73 a	496 a	92 a	10 a	23 a
Riparia	0.04 a	26 a	59 b	408 a	75 a	10 a	27 a
<b>RM*UTGC:</b>							
NRM-Herb	0.04 a	32 a	67 ab	500 a	80 a	11 a	27 a
NRM-CC	0.04 a	31 ab	54 b	403 a	98 a	11 a	28 a
RBG-Herb	0.04 a	23 b	64 ab	426 a	79 a	10 a	23 a
RBG-CC	0.04 a	22 b	79 a	478 a	77 a	9 a	24 a
<b>Significance:<sup>c</sup></b>							
Block	NS	NS	NS	0.0146	NS	NS	NS
RM	NS	0.0010	0.0467	NS	NS	NS	NS
UTGC	NS	NS	NS	NS	NS	NS	NS
Rootstock	NS	NS	0.0157	0.0139	NS	NS	NS
RM*UTGC	NS	NS	0.0157	0.0333	NS	NS	NS
RM*Rootstock	NS	NS	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS	0.0237
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 6. Treatment effects on macronutrient concentrations (% dry weight) in petioles at veraison in 2016.

Treatment <sup>ab</sup>	Veraison					
	N	S	P	K	Mg	Ca
<b>RM:</b>						
NRM	0.68 a	0.20 a	0.17 a	5.30 a	0.50 b	2.08 b
RBG	0.56 b	0.16 b	0.17 a	3.04 b	0.82 a	2.58 a
<b>UTGC:</b>						
Herb	0.64 a	0.18 a	0.15 b	4.15 a	0.72 a	2.40 a
CC	0.61 a	0.18 a	0.19 a	4.19 a	0.61 b	2.27 a
<b>Rootstock:</b>						
101-14	0.61 a	0.20 a	0.17 ab	4.87 a	0.67 b	2.03 c
Riparia	0.60 a	0.16 b	0.15 b	4.30 b	0.53 c	2.32 b
420-A	0.66 a	0.19 a	0.20 a	3.35 c	0.79 a	2.66 a
<b>RM*UTGC:</b>						
NRM-Herb	0.69 a	0.19 b	0.17 ab	5.38 a	0.54 b	2.18 b
NRM-CC	0.67 a	0.21 a	0.18 ab	5.21 a	0.47 b	1.99 b
RBG-Herb	0.58 b	0.16 c	0.14 b	2.91 b	0.89 a	2.62 a
RBG-CC	0.54 b	0.15 c	0.20 a	3.17 b	0.75 a	2.55 a
<b>Significance:<sup>c</sup></b>						
Block	NS	NS	0.0070	NS	NS	NS
RM	<0.0001	<0.0001	NS	<0.0001	<0.0001	<0.0001
UTGC	NS	NS	0.0111	NS	0.0079	NS
Rootstock	0.0409	<0.0001	0.0049	<0.0001	<0.0001	<0.0001
RM*UTGC	NS	0.0010	NS	NS	NS	NS
RM*Rootstock	NS	NS	NS	NS	NS	0.0177
UTGC*Rootstock	0.0489	<0.0001	NS	NS	NS	NS
RM*UTGC*Rootstock	0.0193	0.0453	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 7. Treatment effects on micronutrient concentrations in petioles at veraison in 2016.

Treatment <sup>ab</sup>	Veraison						
	Na (%dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)
<b>RM:</b>							
NRM	0.02 a	25 a	43 b	215 b	39 a	7 a	58 a
RBG	0.02 a	27 a	56 a	277 a	33 a	6 b	49 a
<b>UTGC:</b>							
Herb	0.02 a	27 a	49 a	249 a	38 a	7 a	52 a
CC	0.02 a	25 a	50 a	243 a	35 a	7 a	55 a
<b>Rootstock:</b>							
101-14	0.02 a	28 a	50 ab	254 a	39 a	7 a	59 a
Riparia	0.02 a	26 a	45 b	213 a	30 a	7 a	45 a
420-A	0.02 a	25 a	53 a	271 a	40 a	6 a	57 a
<b>RM*UTGC:</b>							
NRM-Herb	0.02 a	26 a	44 b	216 a	37 a	7 a	49 ab
NRM-CC	0.02 a	25 a	43 b	214 a	41 a	7 a	68 a
RBG-Herb	0.02 a	28 a	54 a	282 a	38 a	6 ab	56 ab
RBG-CC	0.02 a	26 a	57 a	272 a	29 a	6 b	42 b
<b>Significance:<sup>c</sup></b>							
Block	NS	NS	NS	0.0193	NS	NS	NS
RM	NS	NS	<0.0001	0.0133	NS	0.0005	NS
UTGC	NS	NS	NS	NS	NS	NS	NS
Rootstock	NS	NS	0.0171	NS	NS	NS	NS
RM*UTGC	NS	NS	NS	NS	NS	NS	0.0106
RM*Rootstock	NS	NS	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	0.0486	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

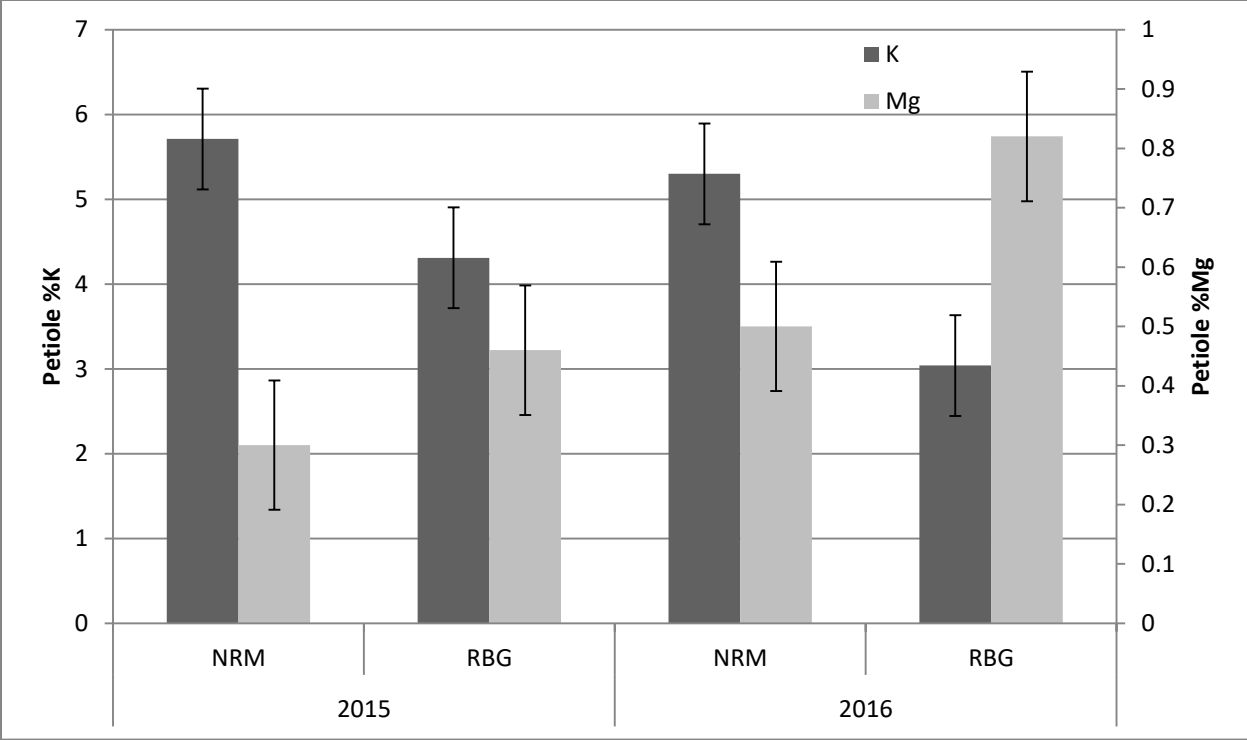


Figure 3. Veraison petiole K and Mg for NRM (no root manipulation) and RBG (root bag) in 2015 and 2016.

## *Canopy Metrics*

Root manipulation (RM) had the most consistent effects on all EPQA metrics at bloom and veraison. At bloom, occlusion layer number (OLN), cluster exposure layer (CEL), and leaf exposure layer (LEL) were reduced by the RBG in both years compared to NRM (Table 8). Cluster exposure flux availability (CEFA) and leaf exposure flux availability (LEFA) were both increased by the RBG factor at bloom in both years (Table 8). At veraison, the RBG factor had identical effects on all EPQA metrics in 2016 only (Table 9). UTGC treatment effects were observed at bloom 2015 only; CC reduced OLN, CEL, and LEL, while CEFA and LEFA were increased (Table 9).

Fruit-zone photosynthetic photon flux density (PPFD) at veraison was significantly affected by RM, UTGC, and Rootstock in both years (Table 10). RBG increased fruit-zone PPFD by an average of 234% over the two years. CC increased fruit-zone PPFD by an average of 62%. Rootstock 101-14 consistently had the lowest PPFD in 2015 and 2016. In 2016, 420-A had the highest PPFD of the three rootstocks. Interactive effects were also present in both years with RM-UTGC and RM-Rootstock (Table 10). RBG-CC consistently had the highest PPFD values and was on average 342% greater than NRM-HERB over the two years.

Table 8. Treatment effects on enhanced point quadrat analysis at bloom 2015-2016.

Treatment <sup>ab</sup>	Occlusion Layer Number (OLN)		Cluster Exposure Layer (CEL)		Leaf Exposure Layer (LEL)		Cluster Exposure Flux Availability (CEFA)		Leaf Exposure Flux Availability (LEFA)	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<b>RM:</b>										
NRM	3.15 a	2.23 a	0.73 a	0.28 a	0.35 a	0.16 a	0.22 b	0.41 b	0.38 b	0.47 b
RBG	2.67 b	1.98 b	0.53 b	0.18 b	0.24 b	0.11 b	0.35 a	0.47 a	0.46 a	0.51 a
<b>UTGC:</b>										
Herb	3.09 a	2.13 a	0.72 a	0.24 a	0.33 a	0.14 a	0.24 b	0.43 a	0.39 b	0.49 a
CC	2.73 b	2.08 a	0.54 b	0.21 a	0.25 b	0.13 a	0.33 a	0.45 a	0.44 a	0.49 a
<b>Rootstock:</b>										
101-14	2.86 a	2.20 a	0.63 a	0.23 a	0.28 a	0.16 a	0.28 a	0.42 a	0.42 a	0.47 b
Riparia	2.96 a	2.03 a	0.63 a	0.20 a	0.30 a	0.13 ab	0.30 a	0.47 a	0.42 a	0.50 ab
420-A	-	2.08 a	-	0.24 a	-	0.11 b	-	0.42 a	-	0.51 a
<b>RM*UTGC:</b>										
NRM-Herb	3.23 a	2.22 a	0.85 a	0.30 a	0.37 a	0.14 a	0.19 c	0.38 b	0.37 c	0.48 ab
NRM-CC	3.07 a	2.23 a	0.62 b	0.25 ab	0.33 ab	0.17 a	0.26 bc	0.43 ab	0.38 bc	0.46 b
RBG-Herb	2.94 a	2.04 ab	0.59 b	0.18 b	0.29 b	0.13 a	0.30 ab	0.47 a	0.41 b	0.50 ab
RBG-CC	2.39 b	1.93 b	0.46 b	0.17 b	0.18 c	0.10 a	0.40 a	0.47 a	0.50 a	0.52 a
<b>Significance:<sup>c</sup></b>										
Block	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM	<0.0001	0.0006	0.0007	0.0010	<0.0001	0.0406	<0.0001	0.0014	<0.0001	0.0009
UTGC	<0.0001	NS	0.0022	NS	<0.0001	NS	0.0032	NS	<0.0001	NS
Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	0.0387
RM*UTGC	0.0226	NS	NS	NS	0.0408	NS	NS	NS	<0.0001	NS
RM*Rootstock	NS	NS	0.0236	0.0382	NS	NS	0.0376	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	0.0339	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock (2015) and Tukey's HSD for RM\*UTGC and Rootstock (2016) ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 9. Treatment effects on enhanced point quadrat analysis at veraison 2015-2016.

Treatment <sup>ab</sup>	Occlusion Layer Number (OLN)		Cluster Exposure Layer (CEL)		Leaf Exposure Layer (LEL)		Cluster Exposure Flux Availability (CEFA)		Leaf Exposure Flux Availability (LEFA)	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<b>RM:</b>										
NRM	2.49 a	2.59 a	0.38 a	0.46 a	0.21 a	0.20 a	0.35 a	0.31 b	0.44 a	0.43 b
RBG	2.36 a	2.13 b	0.36 a	0.27 b	0.19 a	0.13 b	0.39 a	0.43 a	0.46 a	0.50 a
<b>UTGC:</b>										
Herb	2.45 a	2.39 a	0.39 a	0.37 a	0.19 a	0.16 a	0.36 a	0.37 a	0.45 a	0.46 a
CC	2.40 a	2.35 a	0.34 a	0.36 a	0.21 a	0.17 a	0.39 a	0.37 a	0.45 a	0.46 a
<b>Rootstock:</b>										
101-14	2.40 a	2.40 a	0.36 a	0.37 a	0.20 a	0.14 a	0.37 a	0.36 a	0.45 a	0.46 a
Riparia	2.45 a	3.38 a	0.38 a	0.36 a	0.20 a	0.18 a	0.37 a	0.38 a	0.45 a	0.46 a
420-A	-	2.32 a	-	0.36 a	-	0.17 a	-	0.37 a	-	0.47 a
<b>RM*UTGC:</b>										
NRM-Herb	2.48 a	2.63 a	0.41 a	0.46 a	0.20 a	0.20 a	0.34 a	0.31 a	0.45 a	0.43 b
NRM-CC	2.50 a	2.56 a	0.37 a	0.45 a	0.22 a	0.20 ab	0.36 a	0.31 b	0.42 a	0.43 b
RBG-Herb	2.42 a	2.14 b	0.35 a	0.27 b	0.19 a	0.14 bc	0.38 a	0.42 a	0.45 a	0.50 a
RBG-CC	2.31 a	2.13 b	0.34 a	0.27 b	0.20 a	0.12 c	0.41 a	0.44 a	0.47 a	0.50 a
<b>Significance:<sup>c</sup></b>										
Block	<0.0001	NS	0.0079	NS	<0.0001	NS	0.0118	NS	0.0003	NS
RM	NS	<0.0001	NS	<0.0001	NS	<0.0001	NS	<0.0001	NS	<0.0001
UTGC	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM*UTGC	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock (2015) and Tukey's HSD for RM\*UTGC and Rootstock (2016) ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 10. Treatment effects on fruit-zone PPFD at veraison 2015-2016.

<b>Treatment<sup>ab</sup></b>	<b>PPFD</b>	
	<b>2015</b>	<b>2016</b>
<b>RM:</b>		
NRM	1.45 b	1.05 b
RBG	4.34 a	4.01 a
<b>UTGC:</b>		
Herb	2.24 b	1.90 b
CC	3.55 a	3.16 a
<b>Rootstock:</b>		
101-14	2.27 b	2.11 b
Riparia	3.52 a	2.49 ab
420-A	-	2.98 a
<b>RM*UTGC:</b>		
NRM-Herb	1.47 c	0.99 c
NRM-CC	1.42 c	1.11 c
RBG-Herb	3.02 b	2.81 b
RBG-CC	5.67 a	5.21 a
<b>Significance:<sup>c</sup></b>		
Block	NS	0.0066
RM	<0.0001	<0.0001
UTGC	<0.0001	<0.0001
Rootstock	<0.0001	0.0034
RM*UTGC	<0.0001	<0.0001
RM*Rootstock	0.0007	0.0022
UTGC*Rootstock	NS	NS
RM*UTGC*Rootstock	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock (2015) and Tukey's HSD for RM\*UTGC and Rootstock (2016) ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)



### *Pruning Weights, Crop Load, Vine Capacity*

Dormant pruning weights were significantly affected by RM and UTGC in 2015. The largest separation was between RM factors, with RBG significantly reducing the weight of pruned canes (Table 11). CC also reduced pruning weights compared with HERB vines (Table 11). Vine capacity was also affected by RM and UTGC, with a larger separation among UTGC factors. Similarly, vine capacity was reduced by the RBG and CC respectively (Table 11). Differences in vine capacity were observed by rootstock in 2015, as capacity was increased in the Riparia rootstock. A RM-UTGC interaction was also observed with respect to vine capacity, such that the highest capacity was observed in NRM-HERB and the lowest in RBG-CC. As pruning weights and capacity were reduced by RBG and CC, crop load was subsequently increased (Table 11). The largest separation in crop load was observed with the RM treatment.

Table 11. Treatment effects on pruning weight/ m canopy, crop load, vine capacity in 2015.

<b>Treatment<sup>ab</sup></b>	<b>Pruning weight (kg)/ m canopy</b>	<b>Crop Load<sup>d</sup></b>	<b>Vine Capacity (kg)<sup>d</sup></b>
<b>RM:</b>			
NRM	1.12 a	2.68 b	5.97 a
RBG	0.54 b	6.60 a	5.13 b
<b>UTGC:</b>			
Herb	0.99 a	4.03 b	6.26 a
CC	0.66 b	5.25 a	4.84 b
<b>Rootstock:</b>			
101-14	0.84 a	4.36 a	5.34 b
Riparia	0.81 a	4.92 a	5.77 a
<b>RM*UTGC:</b>			
NRM-Herb	1.30 a	2.36 c	6.41 a
NRM-CC	0.95 b	3.01 c	5.53 b
RBG-Herb	0.69 c	5.71 b	6.10 ab
RBG-CC	0.38 d	7.50 a	4.16 c
<b>Significance:<sup>c</sup></b>			
Block	NS	NS	NS
RM	<0.0001	<0.0001	0.0001
UTGC	<0.0001	0.0003	<0.0001
Rootstock	NS	NS	0.0469
RM*UTGC	NS	NS	0.0138
RM*Rootstock	NS	NS	NS
UTGC*Rootstock	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

<sup>d</sup> Crop Load = Yield/vine:Pruning weight/vine; Vine Capacity = Yield/vine + Pruning weight/vine

*Canopy Management Labor*

RM and UTGC treatments affected both timed canopy management practices significantly. RBG vines required on average 48% less time to manually remove leaves in the fruiting zone compared to NRM vines (Table 12). CC vines required 22% less time for leaf removal compared with HERB vines (Table 12). Similarly, both RBG and CC factors reduced the time required to hedge shoots using a mechanical handheld trimmer by 62% and 46% respectively (Table 12).

Table 12. Treatment effects on timed manual labor: leaf removal and shoot hedging in 2015.

<b>Treatment<sup>ab</sup></b>	<b>Leaf removal (hrs./acre)</b>	<b>Hedging (min./acre)</b>
<b>RM:</b>		
NRM	19.9 a	34.5 a
RBG	9.5 b	11.9 b
<b>UTGC:</b>		
Herb	15.4 a	27.1 a
CC	11.8 b	14.7 b
<b>Rootstock:</b>		
101-14	14.9 a	22.5 a
Riparia	15.4 a	25.1 b
<b>RM*UTGC:</b>		
NRM-Herb	21.1 a	43.3 a
NRM-CC	18.7 a	25.7 b
RBG-Herb	11.7 b	16.3 b
RBG-CC	7.3 c	7.4 c
<b>Significance:<sup>c</sup></b>		
Block	<0.0001	0.0019
RM	<0.0001	<0.0001
UTGC	<0.0001	<0.0001
Rootstock	NS	NS
RM*UTGC	NS	NS
RM*Rootstock	NS	NS
UTGC*Rootstock	NS	NS
RM*UTGC*Rootstock	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

### *Primary Fruit Chemistry*

Treatment effects on soluble solids were the most consistent among primary chemistry metrics in 2015 and 2016. In both years soluble solids were increased by RBG and CC factors (Table 13). Inconsistent treatment effects were seen between 2015 and 2016 for pH and titratable acidity. Fruit of RBG vines had significantly higher pH in 2015 but not 2016; while RBG titratable acidity was significantly lower in 2016 but not 2015 (Table 13). In both years rootstock significantly affected pH, but had no significant effect on soluble solids or titratable acidity. 101-14 consistently produced higher pH fruit, while 420-A had the lowest pH in 2016 (Table 13). In both years UTGC had no effect on pH or titratable acidity.

Table 13. Treatment effects on soluble solids (°Brix), pH, titratable acidity (g/L) in 2015-2016.

Treatment <sup>ab</sup>	Soluble Solids (°Brix)		pH		Titratable Acidity (g/L)	
	2015	2016	2015	2016	2015	2016
<b>RM:</b>						
NRM	21.79 b	20.93 b	3.43 b	3.57 a	6.02 a	6.39 a
RBG	22.50 a	21.61 a	3.55 a	3.58 a	6.30 a	5.27 b
<b>UTGC:</b>						
Herb	21.92 b	21.07 b	3.51 a	3.57 a	6.09 a	5.94 a
CC	22.37 a	21.46 a	3.47 a	3.58 a	6.23 a	5.72 a
<b>Rootstock:</b>						
101-14	22.19 a	21.40 a	3.52 a	3.59 a	6.25 a	5.61 a
Riparia	22.10 a	21.40 a	3.46 b	3.59 a	6.08 a	5.88 a
420-A	-	21.00 a	-	3.54 b	-	5.99 a
<b>RM*UTGC:</b>						
NRM-Herb	21.63 b	20.69 b	3.46 bc	3.58 a	5.77 a	6.46 a
NRM-CC	21.95 b	21.16 ab	3.40 c	3.57 a	6.27 a	6.32 a
RBG-Herb	22.21 ab	21.44 a	3.57 a	3.57 a	6.40 a	5.42 b
RBG-CC	22.78 a	21.77 a	3.54 ab	3.59 a	6.20 a	5.11 b
<b>Significance:<sup>c</sup></b>						
Block	0.0339	NS	NS	0.0074	NS	<0.0001
RM	0.0001	0.0001	<0.0001	NS	NS	<0.0001
UTGC	0.0101	0.0205	NS	NS	NS	NS
Rootstock	NS	NS	0.0429	0.0060	NS	NS
RM*UTGC	NS	NS	NS	NS	NS	NS
RM*Rootstock	NS	NS	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock (2015) and Tukey's HSD for RM\*UTGC and Rootstock (2016) ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

## *Secondary Fruit Chemistry*

RM and UTGC treatment effects on berry skin color chemistry were consistent for 2015 and 2016. In both years RBG and CC factors increased the concentration of anthocyanins in berries by an average of 20% and 11% respectively (Table 14). Total phenolics, represented by the absorbance at 280 nm, was also increased in RBG and CC fruit in both years by an average of 19% and 14% respectively (Table 14). Rootstock effects on anthocyanin concentration were significant in both years. Anthocyanin concentrations were highest for 101-14 in both years, while Riparia was the lowest (Table 14). Absorbance values at 280 nm were also highest for 101-14 in 2015. RM-UTGC interaction was significant for total phenolics in both years, and anthocyanins in 2016. Consistently NRM-HERB resulted in the lowest values, while RBG-CC had higher values for both color metrics (Table 14).

Potassium concentration ( $K^+$ ) was significantly affected by RM in both years. Table 15 shows the significant reduction in potassium concentration in RBG fruit, analyzed in Riparia vines only in 2015. Potassium concentrations in 2016 were also reduced by the RBG factor, amongst all rootstocks (Table 16). In both years UTGC had no effect on fruit potassium concentration, nor did rootstock in 2016. Yeast assimilable nitrogen (YAN) was reduced in CC vines in 2015 by 23%, evaluated among Riparia vines. RM had no effect on YAN in 2015.

Table 14. Treatment effects on anthocyanin concentration and total phenolics (A280) in 2015-2016.

Treatment <sup>ab</sup>	Anthocyanin concentration (mg/g)		Absorbance at 280 nm	
	2015	2016	2015	2016
<b>RM:</b>				
NRM	0.81 b	0.88 b	1.15 b	1.18 b
RBG	1.00 a	1.03 a	1.34 a	1.43 a
<b>UTGC:</b>				
Herb	0.84 b	0.92 b	1.14 b	1.25 b
CC	0.97 a	0.99 a	1.35 a	1.37 a
<b>Rootstock:</b>				
101-14	0.95 a	1.00 a	1.28 a	1.32 a
Riparia	0.87 b	0.91 b	1.21 b	1.28 a
420-A	-	0.94 ab	-	1.33 a
<b>RM*UTGC:</b>				
NRM-Herb	0.76 c	0.87 c	1.09 b	1.18 b
NRM-CC	0.87 bc	0.89 bc	1.21 b	1.19 b
RBG-Herb	0.92 b	0.96 b	1.19 b	1.32 b
RBG-CC	1.07 a	1.09 a	1.48 a	1.54 a
<b>Significance:<sup>c</sup></b>				
Block	NS	NS	0.0114	NS
RM	<0.0001	<0.0001	<0.0001	<0.0001
UTGC	0.0004	0.0054	<0.0001	0.0047
Rootstock	0.0193	0.0216	0.0240	NS
RM*UTGC	NS	0.0321	0.0136	0.0072
RM*Rootstock	NS	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock (2015) and Tukey's HSD for RM\*UTGC and Rootstock (2016) ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

Table 15. Treatment effects on juice YAN and K<sup>+</sup> concentration in Riparia vines in 2015.

Treatment <sup>ab</sup>	YAN (mg/L)	K <sup>+</sup> (mg/L)
<b>RM:</b>		
NRM	141.77 a	930.25 a
RBG	133.03 a	701.33 b
<b>UTGC:</b>		
Herb	155.41 a	813.17 a
CC	119.39 b	818.42 a
<b>RM*UTGC:</b>		
NRM-Herb	153.75 a	926.17 a
NRM-CC	129.79 ab	934.33 a
RBG-Herb	157.07 a	700.17 b
RBG-CC	108.99 b	702.50 b
<b>Significance:<sup>c</sup></b>		
Block	NS	NS
RM	NS	<0.0001
UTGC	0.0002	NS
RM*UTGC	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM and UTGC and Tukey's HSD for RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)



Table 16. Treatment effects on juice YAN and K<sup>+</sup> concentration in 2016.

<b>Treatment<sup>ab</sup></b>	<b>YAN (mg/L)</b>	<b>K<sup>+</sup> (mg/L)</b>
<b>RM:</b>		
NRM	78.31 a	1997.39 a
RBG	72.50 a	1650.06 b
<b>UTGC:</b>		
Herb	83.46 a	1769.28 a
CC	67.35 b	1878.17 a
<b>Rootstock:</b>		
101-14	57.65 b	1943.33 a
Riparia	81.85 a	1769.33 a
420-A	86.71 a	1758.50 a
<b>RM*UTGC:</b>		
NRM-Herb	85.51 a	1910.11 ab
NRM-CC	71.11 a	2084.67 a
RBG-Herb	81.41 a	1628.44 b
RBG-CC	63.59 a	1671.67 b
<b>Significance:<sup>c</sup></b>		
Block	NS	NS
RM	NS	<0.0001
UTGC	0.0391	NS
Rootstock	0.0062	NS
RM*UTGC	NS	NS
RM*Rootstock	NS	NS
UTGC*Rootstock	NS	NS
RM*UTGC*Rootstock	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM and UTGC and Tukey's HSD for Rootstock and RM\*UTGC ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

### *Components of Yield*

RM had almost no effect on components of yield in either year, apart from a reduction in berry weight by RBG in 2016 compared to NRM. Under trellis cover cropping (CC) reduced cluster weights in both years, berry weight in 2015, berries per cluster in both years, and yield in both years (Table 17). Rootstock results were somewhat inconsistent between 2015 and 2016. However, berry weights were consistently greatest with the Riparia rootstock in both years, as were cluster weights in 2015 (Table 17). RM-UTGC interaction was consistently significant for cluster weights and yield in both years. Generally, RBG-CC had the lowest cluster weights and yields and RBG-HERB the highest.

Table 17. Treatment effects on components of yield in 2015-2016.

Treatment <sup>ab</sup>	Cluster count/vine		Cluster weight (g)		Berry weight (g)		Berries/cluster		Yield (kg/m canopy)	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<b>RM:</b>										
NRM	26.58 a	35.71 a	158.79 a	134.65 a	1.54 a	1.52 a	104.28 a	88.86 a	2.78 a	3.15 a
RBG	26.85 a	37.35 a	159.26 a	128.29 a	1.54 a	1.38 b	101.20 a	92.71 a	2.82 a	3.27 a
<b>UTGC:</b>										
Herb	26.75 a	37.41 a	174.62 a	141.94 a	1.58 a	1.45 a	110.72 a	98.16 a	3.09 a	3.52 a
CC	26.68 a	35.75 a	143.43 b	121.00 b	1.52 b	1.45 a	94.75 b	83.41 b	2.51 b	2.90 b
<b>Rootstock:</b>										
101-14	27.12 a	39.10 a	148.72 b	129.81 a	1.45 b	1.40 b	103.06 a	93.11 a	2.65 b	3.36 a
Riparia	26.32 a	35.90 b	169.32 a	136.96 a	1.65 a	1.55 a	102.41 a	88.42 a	2.94 a	3.25 a
420-A	-	34.75 b	-	127.63 a	-	1.41 b	-	90.82 a	-	3.03 a
<b>RM*UTGC:</b>										
NRM-Herb	25.93 a	35.11 b	167.63 a	140.39 ab	1.55 ab	1.50 a	109.34 ab	93.96 a	2.86 b	3.25 b
NRM-CC	27.23 a	36.31 ab	149.95 b	128.90 b	1.53 b	1.54 a	99.21 bc	83.76 b	2.69 bc	3.06 bc
RBG-Herb	27.57 a	39.71 a	181.61 a	143.48 a	1.62 a	1.40 b	112.11 a	102.36 a	3.31 a	3.79 a
RBG-CC	26.13 a	35.19 b	136.91 b	113.10 c	1.51 b	1.37 b	90.29 c	83.06 b	2.33 c	2.74 c
<b>Significance:<sup>c</sup></b>										
Block	NS	<0.0001	NS	NS	NS	NS	NS	NS	NS	0.0008
RM	NS	NS	NS	NS	NS	<0.0001	NS	NS	NS	NS
UTGC	NS	NS	<0.0001	<0.0001	0.0122	NS	<0.0001	<0.0001	<0.0001	<0.0001
Rootstock	NS	0.0037	<0.0001	NS	<0.0001	<0.0001	NS	NS	0.0173	NS
RM*UTGC	NS	0.0093	0.0029	0.0070	NS	NS	0.0500	NS	0.0012	0.0015
RM*Rootstock	NS	NS	NS	NS	<0.0001	NS	0.0066	NS	NS	NS
UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
RM*UTGC*Rootstock	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> RM: root manipulation (NRM = no root manipulation, RBG = root bag); UTGC: under-trellis ground cover (Herb = under-trellis herbicide strip, CC = under-trellis cover crop)

<sup>b</sup> Separation of means by Student's T-test for RM, UTGC, and Rootstock (2015) and Tukey's HSD for RM\*UTGC and Rootstock (2016) ( $\alpha=0.05$ )

<sup>c</sup> Significance of treatment effects on response variables using standard least squares with an emphasis on effect leverage ( $p>F$ ; NS = not significant)

### Linear Regression Analysis

Linear regression analyses were performed to determine correlation between various response variables and color absorbance metrics. Strong positive linear relationships were found between color metrics (anthocyanin concentration, A280) and average PPFD and crop load (Figures 4, 6, 8, 10, 12, 14). Strong negative relationships were found between color metrics and berry weight and vine capacity (Figures 5, 7, 9, 11, 13, 15). Vine capacity was the most highly correlated variable to color absorbance metrics, indicated by higher r-square values. Additionally, Figure 16 shows the strong positive correlation of veraison petiole K% to juice K concentration in 2016.

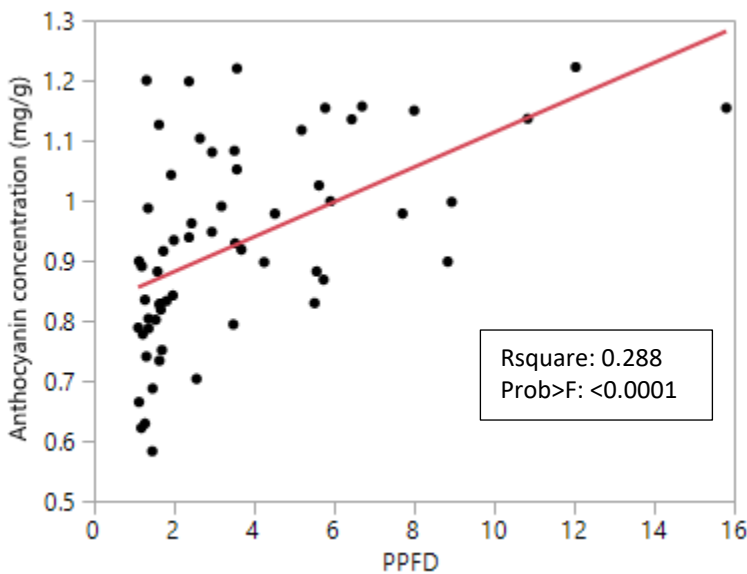


Figure 4. Regression analysis of anthocyanin concentration (mg/g) and average veraison PPFD in 2015.

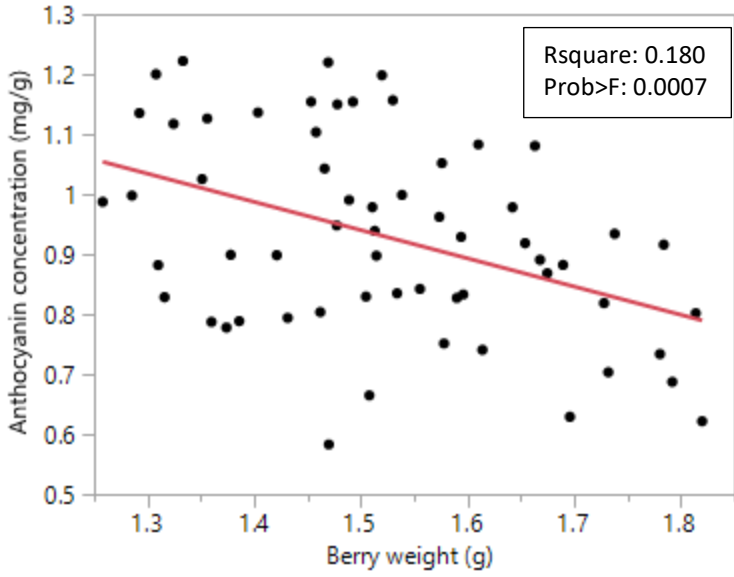


Figure 5. Regression analysis of anthocyanin concentration (mg/g) and average berry weight (g) in 2015.

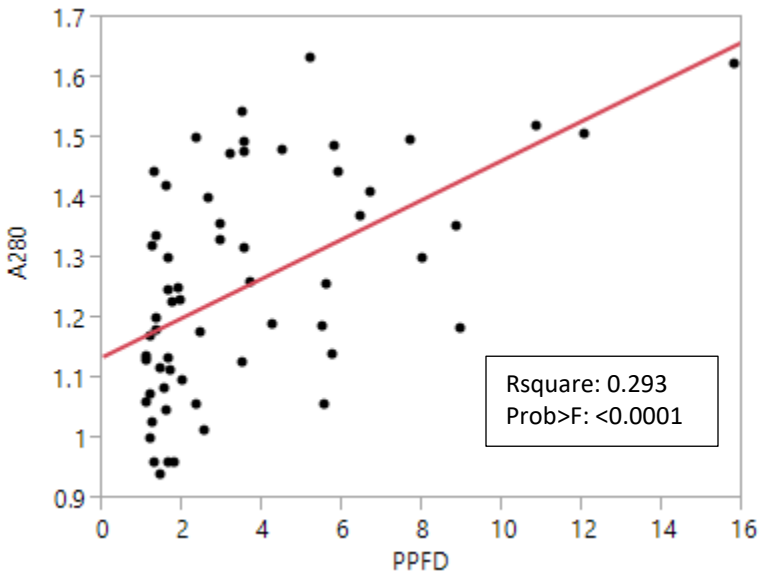


Figure 6. Regression analysis of A280 and average veraison PPFD in 2015.

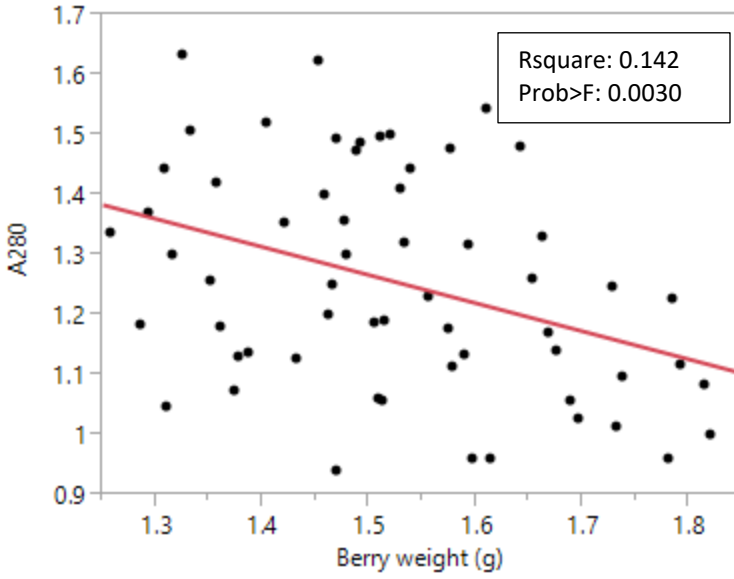


Figure 7. Regression analysis of A280 and average berry weight (g) in 2015.

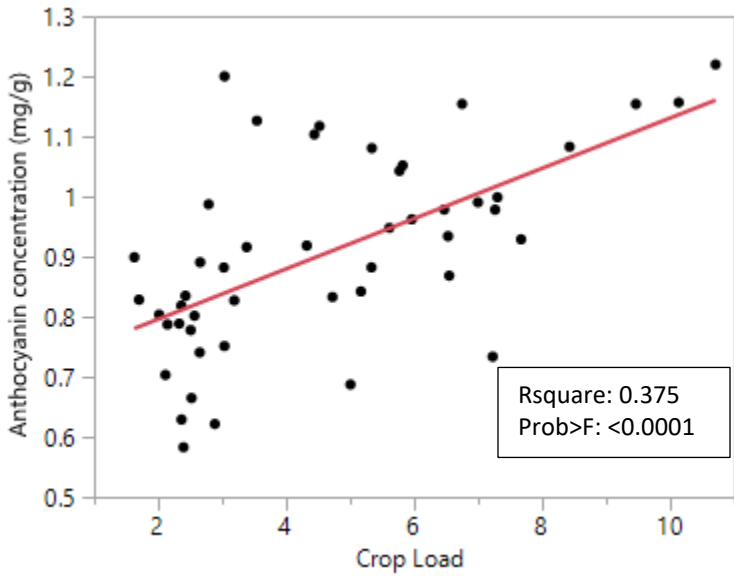


Figure 8. Regression analysis of anthocyanin concentration (mg/g) and average crop load in 2015.

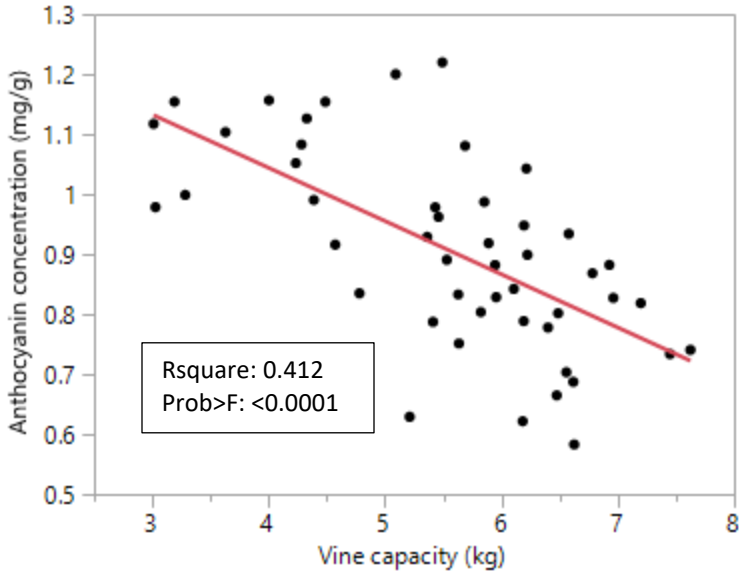


Figure 9. Regression analysis of anthocyanin concentration (mg/g) and average vine capacity (kg) in 2015.

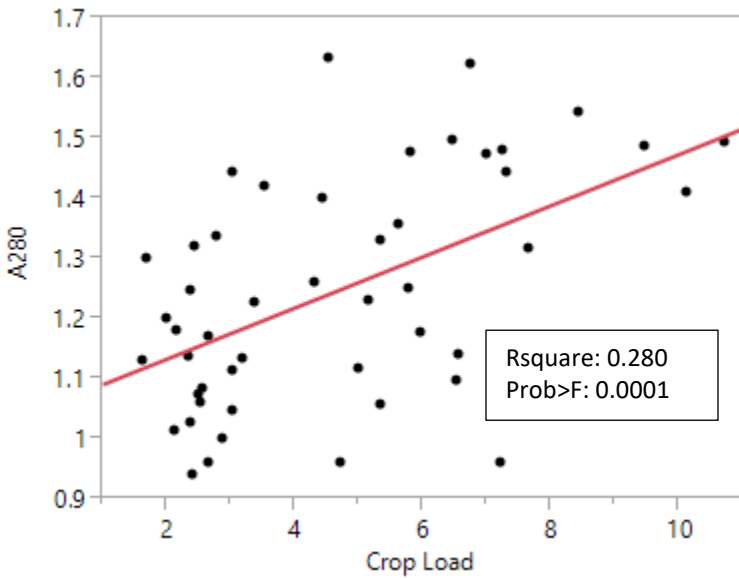


Figure 10. Regression analysis of A280 and average crop load in 2015.

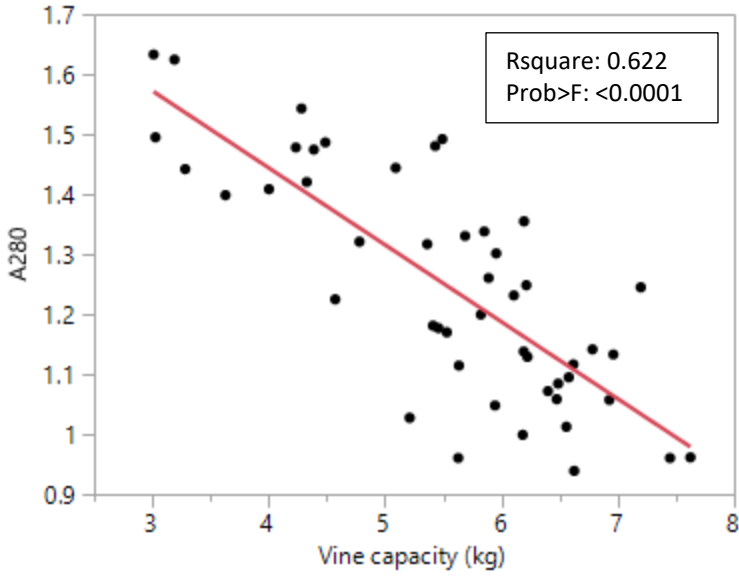


Figure 11. Regression analysis of A280 and average vine capacity (kg) in 2015.

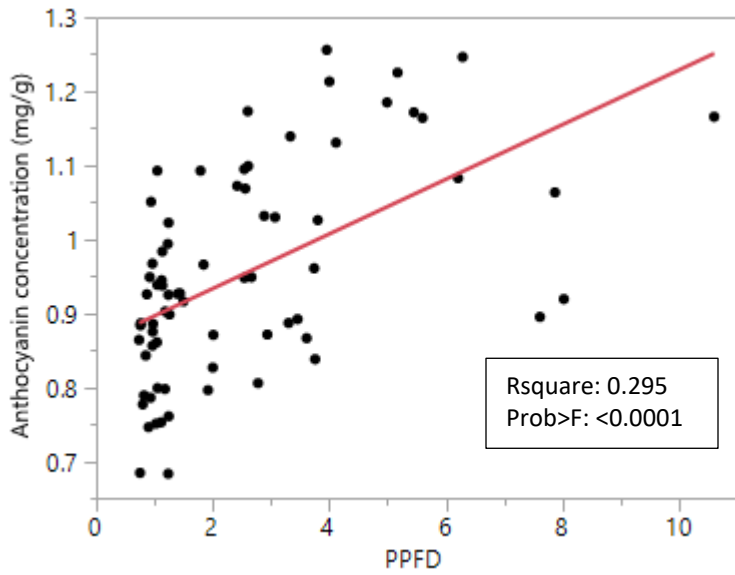


Figure 12. Regression analysis of anthocyanin concentration (mg/g) and average veraison PPFD in 2016.



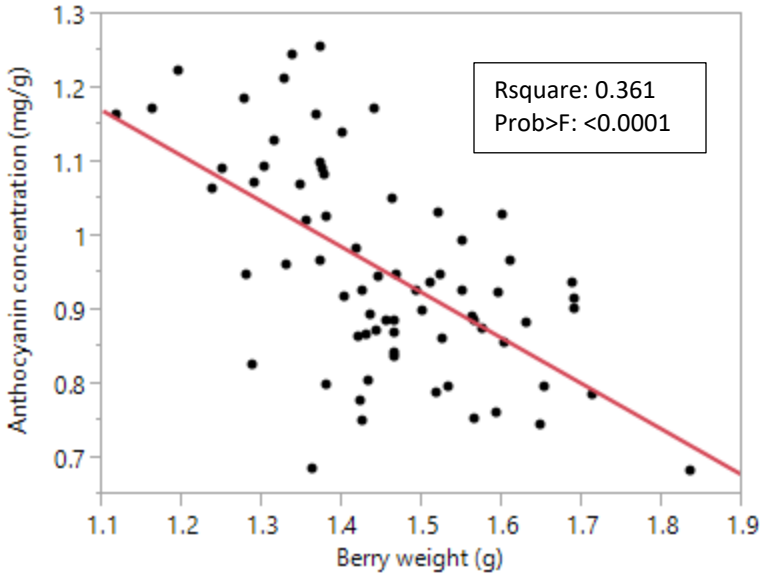


Figure 13. Regression analysis of anthocyanin concentration (mg/g) and average berry weight (g) in 2016.

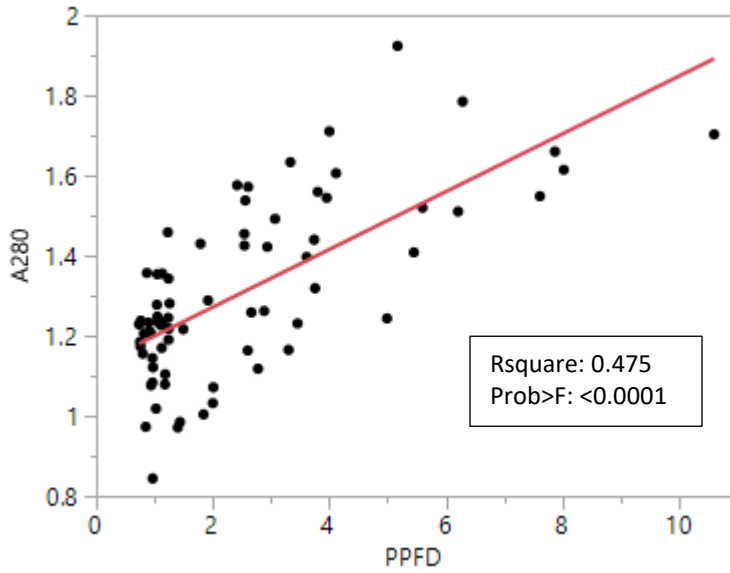


Figure 14. Regression analysis of A280 and average veraison PPFD in 2016.

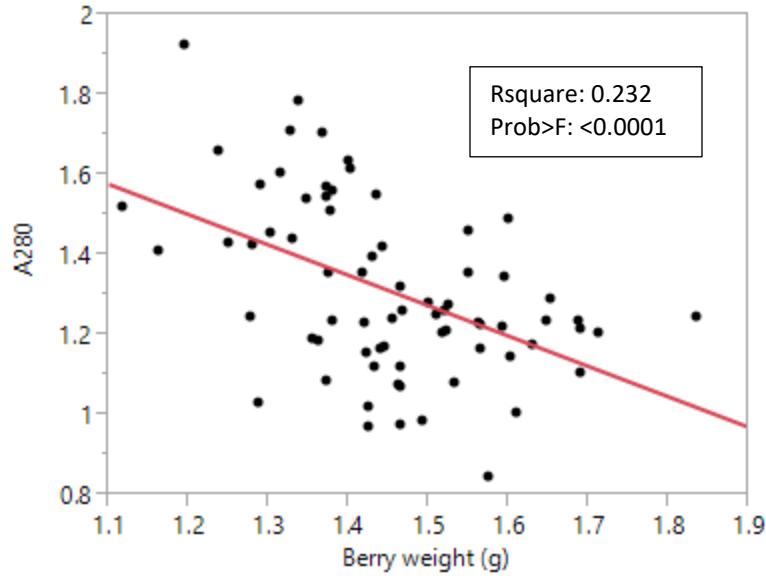


Figure 15. Regression analysis of A280 and average berry weight (g) in 2016.

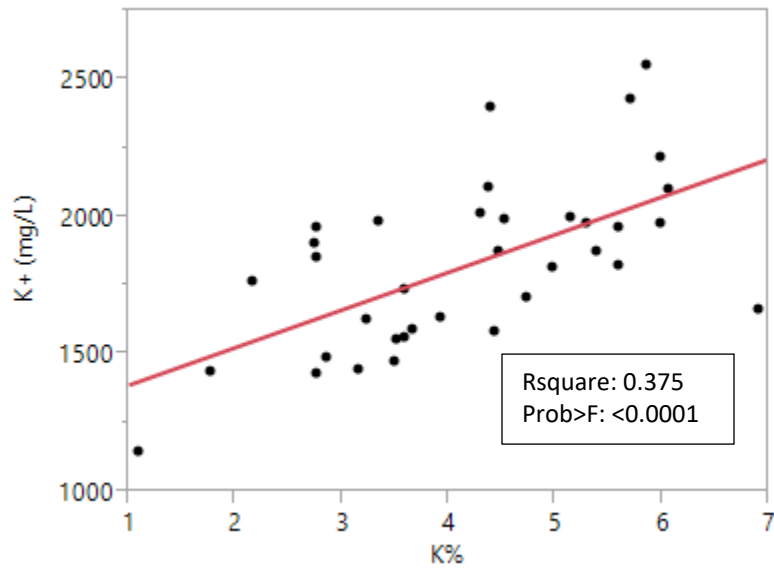


Figure 16. Regression analysis of juice K concentration (mg/L) and veraison petiole K% in 2016.

## **Experiment II**

### *Plant Tissue Analysis*

There was no treatment effect on the nitrogen concentration of plant tissues collected in either year (Tables 18,19, and 22). Potassium concentration of petioles at veraison in both years, and leaf blade + petiole concentrations was reduced by decreasing rooting volume (Tables 18,19, and 22). Conversely, magnesium concentrations were increased with decreasing rooting volume in veraison 2015 and 2016 petioles, and leaf blade + petioles at veraison 2015. Similarly, manganese concentrations were increased with decreasing root volume in bloom 2015 and veraison 2015 and 2016 (Tables 20 & 23).

Table 18. Treatment effects on macronutrient concentrations (% dry weight) in petioles at bloom and veraison in 2015.

	Bloom						Veraison					
Treatment <sup>ab</sup>	N	S	P	K	Mg	Ca	N	S	P	K	Mg	Ca
0.026 m <sup>3</sup>	0.67 a	0.26 a	0.33 c	3.79 a	0.30 a	1.82 a	0.40 a	0.19 a	0.10 a	4.23 b	0.48 a	1.44 a
0.035 m <sup>3</sup>	0.69 a	0.27 a	0.45 b	3.64 a	0.31 a	1.87 a	0.37 a	0.17 a	0.13 a	4.37 b	0.39 b	1.40 a
0.058 m <sup>3</sup>	0.71 a	0.27 a	0.48 b	3.34 a	0.31 a	2.14 a	0.33 a	0.20 a	0.17 a	5.74 a	0.37 b	1.66 a
NRM	0.72 a	0.30 a	0.61 a	3.67 a	0.34 a	2.16 a	0.36 a	0.20 a	0.15 a	5.99 a	0.26 c	1.63 a
Significance <sup>c</sup>	NS	NS	0.0003	NS	NS	NS	NS	NS	NS	0.0021	<0.0001	NS
Block	NS	NS	NS	NS	NS	NS	0.0360	0.0335	0.0201	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 19. Treatment effects on macronutrient concentrations (% dry weight) in petioles + leaf blades at veraison in 2015.

	Veraison					
Treatment <sup>ab</sup>	N	S	P	K	Mg	Ca
0.026 m <sup>3</sup>	1.97 a	0.27 a	0.14 a	1.77 b	0.28 a	1.56 a
0.035 m <sup>3</sup>	2.09 a	0.25 a	0.14 a	1.71 b	0.25 ab	1.60 a
0.058 m <sup>3</sup>	1.96 a	0.26 a	0.15 a	2.21 ab	0.22 bc	1.69 a
NRM	2.07 a	0.26 a	0.15 a	2.32 a	0.19 c	1.83 a
Significance <sup>c</sup>	NS	NS	NS	0.0115	0.0026	NS
Block	NS	NS	NS	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 20. Treatment effects on micronutrient concentrations in petioles at bloom and veraison in 2015.

Treatment <sup>ab</sup>	Bloom							Veraison						
	Na (%dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)	Na (% dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)
0.026 m <sup>3</sup>	0.05 a	29 b	57 a	225 a	34 a	12 a	13 a	0.04 a	25 a	70 a	353 a	31 a	7 a	24 a
0.035 m <sup>3</sup>	0.05 a	31 ab	55 a	222 a	40 a	12 a	16 a	0.04 a	24 a	57 a	251 ab	33 a	7 a	29 a
0.058 m <sup>3</sup>	0.05 a	32 ab	53 a	178 b	37 a	12 a	17 a	0.04 a	28 a	86 a	201 b	32 a	8 a	25 a
NRM	0.05 a	34 a	65 a	202 ab	38 a	13 a	17 a	0.04 a	27 a	59 a	180 b	32 a	8 a	24 a
Significance <sup>c</sup>	NS	0.0078	NS	0.0114	NS	NS	NS	NS	NS	NS	0.0084	NS	0.0433	NS
Block	NS	NS	0.0285	0.0053	NS	NS	NS	NS	NS	NS	0.0399	NS	NS	0.0447

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 21. Treatment effects on micronutrient concentrations in petioles + leaf blades at veraison in 2015.

Treatment <sup>ab</sup>	Veraison						
	Na (%dw)	B (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Cu (ppm)	Al (ppm)
0.026 m <sup>3</sup>	0.03 a	22 a	68 a	482 a	77 a	9 bc	26 a
0.035 m <sup>3</sup>	0.03 a	21 a	53 a	371 a	71 a	9 bc	25 a
0.058 m <sup>3</sup>	0.03 a	26 a	58 a	360 a	75 a	10 ab	24 a
NRM	0.03 a	26 a	50 a	370 a	73 a	11 a	22 a
Significance <sup>c</sup>	NS	NS	NS	NS	NS	0.0012	NS
Block	NS	NS	NS	0.0336	0.0180	0.0233	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 22. Treatment effects on macronutrient concentrations (% dry weight) in petioles at veraison in 2016.

<b>Veraison</b>						
<b>Treatment<sup>ab</sup></b>	<b>N</b>	<b>S</b>	<b>P</b>	<b>K</b>	<b>Mg</b>	<b>Ca</b>
0.026 m <sup>3</sup>	0.48 a	0.18 a	0.31 a	4.40 c	0.62 a	1.91 a
0.035 m <sup>3</sup>	0.47 a	0.18 a	0.25 a	4.63 c	0.55 a	1.88 a
0.058 m <sup>3</sup>	0.46 a	0.18 a	0.30 a	5.27 b	0.45 b	1.96 a
NRM	0.49 a	0.19 a	0.31 a	5.90 a	0.40 b	1.98 a
Significance <sup>c</sup>	NS	NS	NS	<0.0001	0.0001	NS
Block	NS	NS	NS	0.0071	0.0091	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 23. Treatment effects on micronutrient concentrations in petioles at veraison in 2016.

<b>Veraison</b>							
<b>Treatment<sup>ab</sup></b>	<b>Na</b>	<b>B</b>	<b>Zn</b>	<b>Mn</b>	<b>Fe</b>	<b>Cu</b>	<b>Al</b>
	<b>(%dw)</b>	<b>(ppm)</b>	<b>(ppm)</b>	<b>(ppm)</b>	<b>(ppm)</b>	<b>(ppm)</b>	<b>(ppm)</b>
0.026 m <sup>3</sup>	0.02 a	26 a	69 a	423 a	90 a	7 a	13 ab
0.035 m <sup>3</sup>	0.02 a	25 a	57 a	318 ab	31 a	7 a	21 a
0.058 m <sup>3</sup>	0.02 a	23 a	46 a	186 b	25 a	6 a	4 b
NRM	0.02 a	25 a	59 a	200 b	31 a	7 a	6 b
Significance <sup>c</sup>	NS	NS	NS	0.0034	NS	NS	0.0080
Block	NS	NS	NS	0.0355	NS	0.0361	0.0009

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

### *Canopy Metrics*

At bloom, decreasing rooting volume increased CEFA values in 2015 and LEFA values in both years (Table 24). Other EPQA metrics at bloom were not significantly affected. Veraison EPQA metrics were inconsistent between years. In 2016 OLN and CEL were reduced by decreasing rooting volume, and CEFA and LEFA were both increased. CEFA and LEFA values were increased in the 0.026 m<sup>3</sup> treatment by 76% and 28% respectively compared to NRM vines in 2016 (Table 25).

Fruit-zone PPFD was increased by decreasing rooting volume in both years. The 0.035 m<sup>3</sup> treatment had the highest fruit-zone PPFD in 2015, while the 0.026 m<sup>3</sup> volume was highest in 2016. The 0.026 m<sup>3</sup> rooting volume increased fruit-zone PPFD by an average of 207% over the two years (Table 26).

Table 24. Treatment effects on enhanced point quadrat analysis at bloom 2015-2016.

	Occlusion Layer Number (OLN)		Cluster Exposure Layer (CEL)		Leaf Exposure Layer (LEL)		Cluster Exposure Flux Availability (CEFA)		Leaf Exposure Flux Availability (LEFA)	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<b>Treatment<sup>ab</sup></b>										
0.026 m <sup>3</sup>	2.71 a	1.91 a	0.53 a	0.32 a	0.27 a	0.11 a	0.33 a	0.43 a	0.45 a	0.55 a
0.035 m <sup>3</sup>	2.85 a	1.65 a	0.58 a	0.18 a	0.25 a	0.17 a	0.28 ab	0.46 a	0.43 a	0.46 ab
0.058 m <sup>3</sup>	2.79 a	2.23 a	0.53 a	0.32 a	0.26 a	0.13 a	0.30 ab	0.39 a	0.42 a	0.48 ab
NRM	3.18 a	2.21 a	0.63 a	0.28 a	0.32 a	0.19 a	0.21 b	0.41 a	0.36 b	0.45 b
Significance <sup>c</sup>	NS	NS	NS	NS	NS	NS	0.0209	NS	0.0017	0.0388
Block	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 25. Treatment effects on enhanced point quadrat analysis at veraison 2015-2016.

	Occlusion Layer Number (OLN)		Cluster Exposure Layer (CEL)		Leaf Exposure Layer (LEL)		Cluster Exposure Flux Availability (CEFA)		Leaf Exposure Flux Availability (LEFA)	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
<b>Treatment<sup>ab</sup></b>										
0.026 m <sup>3</sup>	2.34 a	2.16 b	0.33 a	0.23 b	0.25 a	0.16 a	0.42 a	0.44 a	0.46 a	0.50 a
0.035 m <sup>3</sup>	2.13 a	2.39 ab	0.25 a	0.38 ab	0.12 a	0.15 a	0.46 a	0.39 a	0.52 a	0.47 ab
0.058 m <sup>3</sup>	2.25 a	2.75 a	0.27 a	0.45 ab	0.18 a	0.22 a	0.43 a	0.33 ab	0.45 a	0.42 bc
NRM	2.37 a	2.93 a	0.35 a	0.57 a	0.15 a	0.26 a	0.35 a	0.25 b	0.46 a	0.39 c
Significance <sup>c</sup>	NS	0.0067	NS	0.0492	NS	NS	NS	0.0068	NS	0.0033
Block	0.0185	NS	0.0073	NS	0.0247	NS	0.0161	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).



Table 26. Treatment effects on fruit-zone PPFD at veraison 2015-2016.

Treatment <sup>ab</sup>	PPFD	
	2015	2016
0.026 m <sup>3</sup>	3.41 ab	5.16 a
0.035 m <sup>3</sup>	3.77 a	4.52 a
0.058 m <sup>3</sup>	2.31 ab	2.41 b
NRM	1.61 b	1.18 b
Significance <sup>c</sup>	0.0097	<0.0001
Block	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

*Pruning Weights, Crop Load, Vine Capacity*

The weights of dormant pruned canes were reduced by decreasing rooting volume in 2015 (Table 27). Crop load was significantly increased by decreasing rooting volume. Crop loads in the 0.026 m<sup>3</sup> treatment were 130% higher than NRM in 2015. Total vine capacity was not affected by treatment in 2015.

Table 27. Treatment effects on pruning weight, crop load, and vine capacity in 2015.

Treatment <sup>ab</sup>	Pruning weight (kg)/ m canopy	Crop Load <sup>d</sup>	Vine Capacity (kg) <sup>d</sup>
0.026 m <sup>3</sup>	0.49 c	7.18 a	6.00 a
0.035 m <sup>3</sup>	0.58 c	6.73 a	6.56 a
0.058 m <sup>3</sup>	0.77 b	4.28 b	5.89 a
NRM	1.08 a	3.12 b	6.63 a
Significance <sup>c</sup>	<0.0001	<0.0001	NS
Block	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

<sup>d</sup> Crop Load = Yield/vine:Pruning weight/vine; Vine Capacity = Yield/vine + Pruning weight/vine

### *Canopy Management Labor*

Time required for canopy management practices was reduced by decreasing rooting volume. Manual fruit-zone leaf removal was reduced by 48% in the 0.026 m<sup>3</sup> treatment compared with NRM (Table 28). Similarly, hedging was reduced by 62% in the 0.026 m<sup>3</sup> treatment compared with NRM vines (Table 28).

Table 28. Treatment effects on timed manual labor: leaf removal and hedging in 2015.

	<b>Leaf removal (hrs./acre)</b>	<b>Hedging (min./acre)</b>
<b>Treatment<sup>ab</sup></b>		
0.026 m <sup>3</sup>	10.1b	9.0 b
0.035 m <sup>3</sup>	14.4 ab	13.9 ab
0.058 m <sup>3</sup>	14.2 b	20.0 a
NRM	19.5 a	23.7 a
Significance <sup>c</sup>	0.0031	0.0091
Block	NS	0.0243

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

### *Primary Fruit Chemistry*

Titrateable acidity (TA) was consistently reduced by decreasing rooting volume in 2015 and 2016 (Table 29). Treatment effects on soluble solids and pH were inconsistent over the two seasons. NRM soluble solids were significantly lowest in 2016, and pH highest in 2015 (Table 29).

Table 29. Treatment effects on soluble solids (°Brix), pH, and titratable acidity (g/L) in 2015-2016.

Treatment <sup>ab</sup>	Soluble Solids (°Brix)		pH		Titratable Acidity (g/L)	
	2015	2016	2015	2016	2015	2016
0.026 m <sup>3</sup>	23.10 a	22.15 a	3.27 b	3.61 a	6.88 b	5.35 b
0.035 m <sup>3</sup>	22.80 a	21.73 a	3.26 b	3.59 a	7.67 a	5.57 b
0.058 m <sup>3</sup>	23.08 a	22.28 a	3.29 b	3.61 a	7.65 a	5.91 ab
NRM	22.18 a	20.35 b	3.34 a	3.56 a	8.12 a	6.82 a
Significance <sup>c</sup>	NS	0.0010	0.0041	NS	0.0025	0.0138
Block	0.0117	NS	<0.0001	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

### *Secondary Fruit Chemistry*

Smaller rooting volume consistently increased anthocyanin concentration in berries in both years (Table 30). The 0.026 m<sup>3</sup> treatment increased anthocyanin concentration by an average of 42% over the two years compared with NRM. Total phenolic content, represented by the absorbance at 280 nm, was significantly affected by rooting volume in 2016. Absorbance values at 280 nm were increased in the 0.026 m<sup>3</sup> treatment by 28% over the two years compared with NRM (Table 30).

Neither yeast assimilable nitrogen (YAN) or juice potassium concentration were significantly affected by treatment in 2015 and 2016 (Table 31).

Table 30. Treatment effects on anthocyanin concentration of berries and total phenolics (A280) in 2015-2016.

Treatment <sup>ab</sup>	Anthocyanin concentration (mg/g)		Absorbance at 280 nm	
	2015	2016	2015	2016
0.026 m <sup>3</sup>	1.25 a	1.30 a	1.60 a	1.68 a
0.035 m <sup>3</sup>	1.10 ab	1.04 ab	1.46 a	1.50 ab
0.058 m <sup>3</sup>	1.05 ab	1.04 ab	1.45 a	1.56 a
NRM	0.96 b	0.84 b	1.32 a	1.24 b
Significance <sup>c</sup>	0.0155	0.0045	NS	0.0106
Block	0.0245	0.0432	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 31. Treatment effects on YAN and K<sup>+</sup> concentration in 2015-2016.

Treatment <sup>ab</sup>	YAN (g/L)		K <sup>+</sup> (mg/L)	
	2015	2016	2015	2016
0.026 m <sup>3</sup>	80.55 a	38.60 a	677.75 a	1763.50 a
0.035 m <sup>3</sup>	92.48 a	42.97 a	688.75 a	1778.00 a
0.058 m <sup>3</sup>	94.10 a	39.00 a	744.75 a	2252.00 a
NRM	108.82 a	68.85 a	809.00 a	1958.00 a
Significance <sup>c</sup>	NS	NS	NS	NS
Block	NS	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

### *Components of Yield*

Treatments had no strong consistent effect on components of yield in 2015 and 2016.

One-way ANOVA determined weak significance of treatment on berry weight in 2015, although there was no separation of means by Tukey's HSD (Table 32).

Table 32. Treatment effects on components of yield in 2015-2016.

Treatment <sup>ab</sup>	Cluster count/vine		Cluster weight (g)		Berry weight (g)		Berries/cluster		Yield (kg)/m canopy	
	2015	2016	2015	2016	2015	2016	2015	2016	2015	2016
0.026 m <sup>3</sup>	33.25 a	40.13 a	161.85 a	146.71 a	1.47 a	1.38 a	110.27 a	106.24 a	3.44 a	3.83 a
0.035 m <sup>3</sup>	33.88 a	41.50 a	168.91 a	166.95 a	1.47 a	1.43 a	114.88 a	117.00 a	3.72 a	4.57 a
0.058 m <sup>3</sup>	30.88 a	41.81 a	155.92 a	152.20 a	1.46 a	1.45 a	106.99 a	105.03 a	3.10 a	4.09 a
NRM	30.69 a	40.19 a	163.06 a	166.00 a	1.33 a	1.44 a	123.14 a	115.45 a	3.27 a	4.36 a
Significance <sup>c</sup>	NS	NS	NS	NS	0.0484	NS	NS	NS	NS	NS
Block	0.0002	<0.0001	0.0006	NS	NS	NS	0.0019	NS	NS	0.0003

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

### Mid-day Stem Water Potentials

Treatments had significant effects on mid-day stem water potentials ( $\psi_{\text{md,stem}}$ ) on each day measured in 2015 and 2016. Decreasing rooting volume consistently reduced stem water potentials. NRM vines had on average 57% and 64% higher stem water potentials than 0.026 m<sup>3</sup> in 2015 and 2016 respectively (Tables 33 & 34). Figure 17 shows stem water potential readings in 2016 along with daily rainfall during the same period.

Table 33. Treatment effects on stem water potential in 2015.

Treatment <sup>ab</sup>	$\psi_{\text{md,stem}}$ (MPa) <sup>a</sup>	
	13 Aug.	24 Aug.
0.026 m <sup>3</sup>	-0.68 b	-1.22 c
0.035 m <sup>3</sup>	-0.60 b	-1.15 c
0.058 m <sup>3</sup>	-0.45 a	-0.85 b
NRM	-0.38 a	-0.44 a
Significance <sup>c</sup>	<0.0001	<0.0001
Block	0.0022	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes; MPa = megapascal.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

Table 34. Treatment effects on stem water potential in 2016.

Treatment <sup>ab</sup>	$\psi_{\text{md,stem}}$ (MPa) <sup>a</sup>			
	12 Aug.	22 Aug.	29 Aug.	6 Sep.
0.026 m <sup>3</sup>	-0.68 c	-0.79 c	-1.08 c	-0.94 c
0.035 m <sup>3</sup>	-0.61 c	-0.65 bc	-0.94 c	-0.88 c
0.058 m <sup>3</sup>	-0.49 b	-0.46 ab	-0.66 b	-0.66 b
NRM	-0.31 a	-0.27 a	-0.29 a	-0.38 a
Significance <sup>c</sup>	<0.0001	<0.0001	<0.0001	<0.0001
Block	0.0002	NS	NS	NS

<sup>a</sup> NRM = no root manipulation; 0.026 m<sup>3</sup>, 0.035 m<sup>3</sup>, and 0.058 m<sup>3</sup> indicate root bag volumes; MPa = megapascal.

<sup>b</sup> Separation of means by Tukey's HSD ( $\alpha=0.05$ ).

<sup>c</sup> Significance of treatment effects on response variables using one-way ANOVA ( $p>F$ ; ns= not significant).

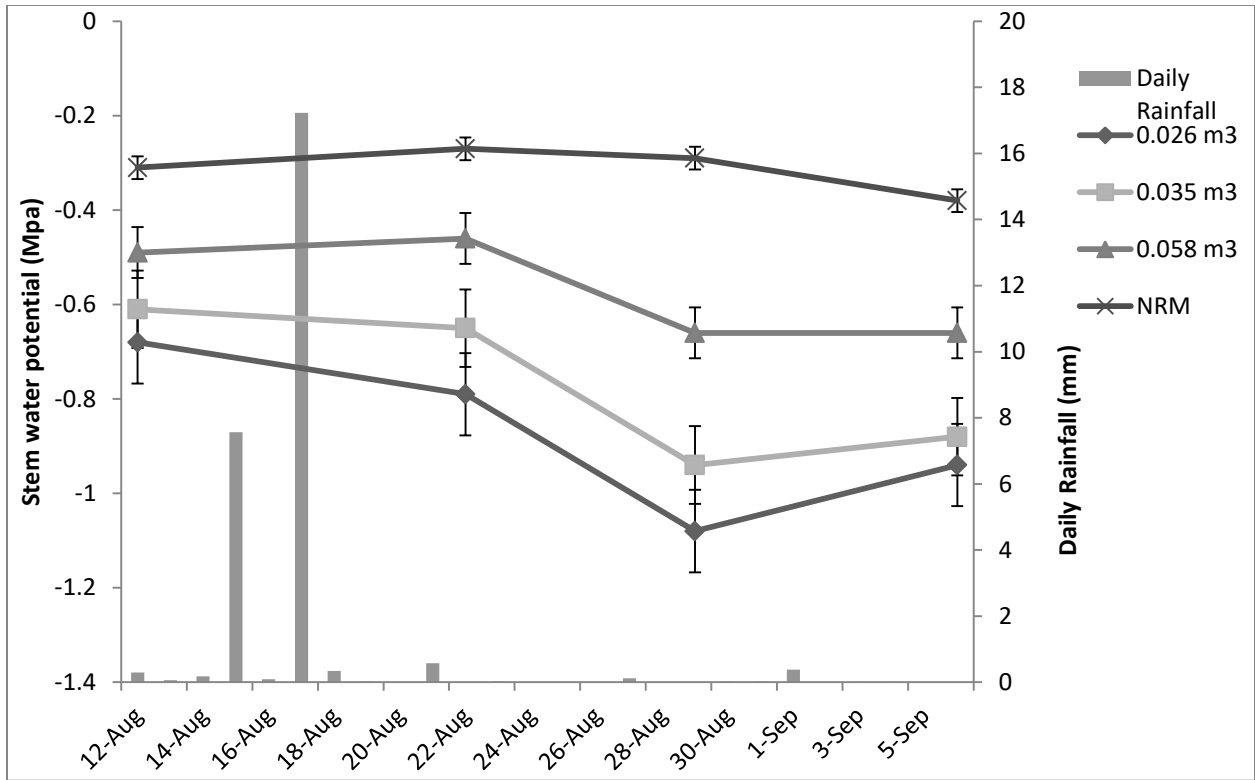


Figure 17. Stem Water Potential measurements by treatment (MPa) and daily rainfall (mm) between 12 Aug. and 6 Sep. of 2016.

## Discussion

The primary constraint to producing high quality wines in the Mid-Atlantic region is the climate. Variable and often superfluous rainfall and high humidity in the growing season, combined with occasionally cold winters creates a challenging environment. The consequence of this environment is very vigorous grapevines that have poor balance between vegetative and reproductive structures, dense and overly crowded canopies, poor fruit-zone exposure, and poor fruit quality. Vigor is primarily determined by an interaction between the genotype and climatic and soil-driven factors, but can be partially mitigated by cultural management practices. Cabernet Sauvignon illustrates the adversity of these growing conditions due to its characteristic late maturation, inherent vigor, and difficulty to ripen in the Mid-Atlantic. Common grapevine training, pruning, and canopy management practices alone are often inadequate to achieve optimal fruit maturity for quality red wine production. The objective of these studies was to evaluate alternative growth management treatments that would simultaneously improve vine balance, reduce management inputs, and optimize fruit quality and yield.

Root volume manipulation using fabric root bags can be considered the most radical treatment used in these studies, and was generally the most effective treatment in all vegetative and reproductive metrics. Included in both experiments as a research tool, the root bags demonstrated the ability to consistently reduce vegetative growth rate and total vegetative production. Vine vigor was limited through inhibition of plant available water and nutrient uptake, which are the primary drivers of early season vegetative growth rates (Poorter & Nagel, 2000). The lack of resources available via the root system likely induces signaling to the above-ground portion of the vine, particularly actively growing shoots and leaves, to slow growth and prompt cessation of active shoot-tip meristems. This signaling can occur hormonally through



water-deficit induced abscisic acid (ABA) production in the roots and movement into xylem sap (Lovisolo et al., 2010), or through hydraulic signaling that can induce ABA production in canes and leaves (Christmann et al., 2007). ABA is known to prevent cell wall loosening necessary for cell expansion during rapid growth, and induce stomatal closure to avoid dehydration.

Experiment II demonstrated that decreased rooting volume caused lower  $\psi_{\text{md,stem}}$  values, indicating a higher degree of water stress. Although petiole and leaf nitrogen concentrations were not consistently lower in RBG compared with NRM, this does not reflect the difference in total plant nitrogen uptake. The similar nutrient concentrations between RM treatment factors may be caused by the difference in size of plant vegetative parts (i.e. leaves, petioles, etc.) and thus a concentrative effect by smaller RBG foliage. Leaf area and petiole size were not quantitatively measured in these studies, but our field notes indicate consistently smaller leaves and petioles on RBG vines. The density of the root system contained inside the root bag in the top portion of the soil would likely make RBG vines more efficient at capturing mobile  $\text{NO}_3^-$  as well (Dunbabin et al., 2003). Nonetheless, there were no effects by root restriction on juice yeast assimilable nitrogen (YAN) at harvest.

Enhanced point quadrat analysis data at veraison were inconsistent among RM treatment factors between 2015 and 2016, but was likely due to the fruit-zone canopy management performed among all treatments. Leaf and lateral removal were conducted on both RBG and NRM vines, thus creating a generally uniform occlusion layer number (OLN) in the fruit-zone. Despite comparable relative openness and layer numbers in the fruit-zone between RBG and NRM, this did not accurately reflect whole-canopy density and size. Insertions recorded for EPQA analysis were performed in the fruit-zone, and thus do not express contact density or width in the upper portion of the canopy. Photosynthetic photon flux density (PPFD)

measurements were more indicative of the relative exposure among treatments. PPFD measurements clearly indicated more exposed fruit-zones in RBG vines compared to NRM vines. Timed canopy management practices in 2015 revealed a marked reduction in time required for executing specific canopy management practices. Manual fruit-zone leaf and lateral removal on NRM vines demanded twice the time of RBG vines. Hedging in commercial settings is often executed using tractor-mounted hedgers that uniformly cut vertically growing shoots to a specific height. Hedging in our experimental vineyard was performed using a handheld gas-powered trimmer, thus we were able to express the differences in vertical growth by timing hedging by treatment panel. In a commercial setting these results may be considered negligible because rows are often managed uniformly, but it should be noted that these data are more a reflection of the need to hedge or not. Our field notes describe the intensive need to hedge NRM vines bi-weekly from June to September, while RBG vines required much less attention. In addition, shoot-tip meristems of RBG vines consistently died and ceased active growth much sooner than NRM vines (Hatch et al., 2011). The differences in canopy density of RBG and NRM vines can be seen in Figures 18a and 18b below.



Figure 18a and 18b. Post-veraison differences between NRM (left) and RBG (right) canopy densities.

Vine phenology can be affected by a number of interactive factors including weather, variety, vine age, nutrition, interaction with other organisms, rootstocks, crop load, and viticultural practices. It can be difficult to assess the individual effects of any given factor due to their simultaneous influence throughout the year. RBG fruit at veraison and harvest was consistently more developmentally advanced in both years, and this can be attributed to multiple effects. Experiment II demonstrated that as rooting volume decreased the degree of water stress increased, resulting in reduced vegetative growth and altered canopy microclimate. Canopy metrics illustrated consistently smaller canopies in RBG vines and greater fruit-zone light interception; although the effects of light and subsequent radiant heating on grape clusters cannot be separated based on our data. Past studies have observed that abundant water supply to grapevines delays veraison and slows the rate of sugar accumulation (Bravdo et al., 1985). In addition, more plant available water has been shown to result in fruit with less sugar, high acidity, and less color (Jackson & Lombard, 1993; Salón et al., 2005). Our berry acidity data were inconsistent between years, but root restriction has the potential to reduce total titratable acidity significantly. Decreased acidity is likely a result of smaller canopies, and thus more light interception and subsequent berry temperature increase causing malate degradation (Ginestar et al., 1998). Soluble solids accumulation was consistently increased in RBG fruit. Our annual field notes indicate RBG vines routinely reach veraison sooner than NRM vines, and express full color change more rapidly. Berry skin color data, determined by absorbance related metrics, expressed higher concentrations of anthocyanins and total phenolics in RBG fruit. The increased ripening in RBG fruit, expressed by soluble solids and color, was in agreement with past studies on root restriction (Mataa & Tominaga, 1998; Wang et al., 2012). Experiment II demonstrated color concentrations were increased as rooting volume decreased. In both experiments the

increased concentrations of color compounds may be at least partially attributed to a decrease in berry size. Differences in berry size are mostly due to a reduction in the mesocarp, while skins and seeds tend to be less affected (Roby & Matthews, 2004). Linear regression analyses show that as PPFD and crop load increases, there is a concurrent increase in anthocyanin and total phenolic concentration; while increasing vine capacity and berry weight result in lower color absorbance values. The concentrative differences in color compounds caused by decreasing berry weight may be compounded by the roles of ABA in ripening and development. Studies have shown that ABA generates changes that induce sugar accumulation, berry softening and anthocyanin biosynthesis (Castellarin et al., 2007; Gambetta et al., 2010; Jeong et al., 2004; Jia et al., 2011).

High potassium (K) concentration in grape berries can impact finished red wine quality in several negative ways. Elevated levels of K (> 1,500 mg/L) in musts can result in increased precipitation of tartrate, decreased free acids, and increased pH (Boulton, 1980). The precipitation of tartaric acid out of solution raises the malate:tartrate ratio, and the increased pH generates high microbial instability, both of which are considered undesirable for high quality wine production. Amendment of pH is possible through the addition of tartaric acid during winemaking but these adjustments can be costly and unsatisfactory, thus an understanding of effective vineyard treatments to reduce berry K accumulation is desired.

Current understanding of berry K content suggests that rootstock selection, canopy management, and water supply may be the most effective means of manipulating K content at harvest (Mpelasoka et al., 2003). Canopy management supporting low K is in agreement with practices that promote other compositional proxies (soluble solids, acidity, color compounds, ect.); sparse, open canopies with high effective leaf area, decreased shading, increased fruit

exposure, and relatively higher crop loads (Hepner & Bravdo, 1985). Water supply may be more related to K concentration than is crop load, although crop load is strongly affected by water supply.

Veraison petiole K concentration was consistently reduced by root restriction in both experiments; Experiment II demonstrated that petiole K decreased with decreasing rooting volume. Magnesium (Mg) concentrations were inversely related, such that increasing rooting volume decreased the concentration of Mg in petioles. This antagonistic relationship between K and Mg has been previously well demonstrated and seems to be affected differently by water supply and crop load (Hepner & Bravdo, 1985; Jakobsen, 1993; Morris et al., 1983). The associated K/Mg uptake relationship seems to be related to the relative availability of these cations in soils. High K and Ca availability in soils is known to reduce plant Mg due to competition for root uptake among the cations. The reduced soil K availability in RBG vines, combined with the application of Mg (and Ca in the form of calcium nitrate) in 2015 is likely responsible for the shift in K/Mg uptake. One previous study showed that high Mg in grapevines protected anthocyanins from degradation in the cell vacuoles of grape skins, resulting in higher concentrations (Sinilal et al., 2011). Juice K concentration at harvest was also decreased by RBG in both years. Petiole K at veraison had a strong positive correlation with juice K measured at harvest. Differences in relative juice K concentrations between 2015 and 2016 were attributed to a difference in the dilution factor chosen for ISE measurement. Although trends among treatments were unaffected, the dilution factor was changed from 10 to 20 to measure K concentration in the appropriate calibration range. Juice K concentrations analyzed using the ISE method with a dilution factor of 20 provided accurate results in agreement with values presented in earlier studies (Threlfall et al., 2006). Despite apparent differences in mean juice K of

Experiment II, the lack of statistical significance can be attributed to the collection of berry samples by panel, thus limiting the sample size.

The establishment of cover crops as a ground cover management system can be an effective method of reducing erosion, improving soil structure, reducing vine vigor, increasing soil organic matter, suppressing weeds, improving microbial biomass, and optimizing polyphenol composition in grape berries (Baumgartner et al., 2008; Celette et al., 2009; Ingels et al., 2005; Perez-Alvarez et al., 2015; Steenwerth & Belina, 2008). Cover crops in vineyard row alleys can be permanent or semi-permanent depending on the location and associated climate. In the mid-Atlantic inter-row cover crops are often permanent to avoid soil erosion caused by high rainfall and heavy mechanical traffic on steep vineyard sites. Intra-row cover crops, or under trellis ground covers (UTGC), are becoming increasingly utilized rather than a typical herbicide strip due to their benefits in high vigor environments and highly erodible sites.

In accordance with past studies, cover-cropping (CC) reduced vine vigor during the growing season, decreased canopy density, decreased yield, and improved fruit maturity compared with vines grown in the presence of an herbicide strip (HERB) (Beslic et al., 2015; Hatch et al., 2011; Tesic et al., 2007). Fruit-zone PPFD was significantly higher in CC canopies, while pruning weights and timed canopy management labor were reduced. Lower yields in CC were due to reduced cluster weights as a function of fewer berries per cluster. The reduction of berries per cluster was likely a combined effect of increased vine-cover crop competition for both water and N during the key periods of crop yield development (between flowering and berry enlargement), resulting in decreased fruit set (Keller et al., 2001; Tesic et al., 2007). Although yield and total vine capacity were reduced, crop load was increased in cover cropped vines. Fruit maturation was enhanced in CC, exhibiting a greater degree of soluble solids and

increased anthocyanin and phenolic concentration, corroborating earlier experiments (Perez-Alvarez et al., 2015; Xi et al., 2010). Despite relatively comparable petiole N concentrations at veraison, CC fruit had significantly lower yeast assimilable nitrogen (YAN). Many studies have reported decreased N uptake and subsequent decreased YAN in cover-cropped systems (Keller et al., 2001; Perez-Alvarez et al., 2015; Tesic et al., 2007). In contrast, one study found that cover cropping had no effect on YAN, while rootstock significantly affected berry YAN (Lee & Steenwerth, 2011). Disparities in the effects of cover-cropping on YAN may relate to differences in the length of cover-crop establishment and environmental factors (humid vs. arid).

Rootstocks grafted onto scion varieties may not necessarily have a genetic influence on the berry composition of the scion fruit. However, rootstocks can indirectly affect fruit quality due to the effects on water and nutrient uptake and translocation, overall growth, and components of yield (Keller et al., 2001; Koblet et al., 1994; Koundouras et al., 2009). Although there were no significant differences in EPQA metrics among rootstocks at veraison, fruit-zone PPFDF was consistently lowest in the 101-14 rootstock. Surprisingly, anthocyanin concentrations were greatest in 101-14 in both years. Previous research on the same vines indicate that phenolics and anthocyanins of 101-14 and Riparia were inconsistently increased compared to 420-A (Hickey et al., 2016). Berry weights were considerably increased in the Riparia rootstock as previously reported (Hickey et al., 2016), although no consistent rootstock effects on total yield were observed. Measured juice YAN in 2016 was greatest in 420-A and Riparia, while 101-14 was significantly decreased. These results are in contrast to a previous study that suggested more vigorous rootstocks will result in increased YAN, although this may be a product of environmental differences and their effect on root distribution (Lee & Steenwerth, 2011).

Many studies have shown that rootstocks vary in the uptake and redistribution of K and may be utilized for the manipulation of fruit pH, although results differed based on rootstock/scion combination and environmental factors (Kodur et al., 2013; Ruhl, 1989; Walker & Blackmore, 2012). Differences in K uptake and translocation between own-rooted and grafted vines appear to be greater than among grafted rootstocks (Walker et al., 1998). Our ranking of K concentration among rootstocks by mean separation suggests that 420-A consistently reduces petiole K content. The reduced concentrations in veraison petioles coincide with lower fruit pH at harvest and mean juice K concentration in 420-A. The antagonistic relationship of K/Mg uptake, and differences in nutrient uptake in general, have been observed among different rootstocks (Dalbo et al., 2011). The differences in nutrient uptake among rootstocks can vary based on its graft partner and the soil nutrient status as well. The K/Mg inverse relationship was exhibited among the three rootstocks evaluated, such that lower K concentrations in 420-A corresponded with higher Mg and Ca concentrations in petioles.

Commercial application of root restriction using fabric root bags is still inhibited by logistical and financial uncertainty. For application to be justified, the resultant grape and/or wine quality would require increased product compensation in return for installation. The cost of a fabric root bag is comparable to the cost of a grapevine, thus the cost per acre for installation would increase by roughly 20-30%. The added labor of filling each bag with soil and appropriate planting would warrant consideration as well. In addition, the durability and persistence of these bags must be further assessed. We estimate that there is some degree of root escape from the root bags, but our data do not quantitatively examine this. Our analysis of pruning weights over time suggest that about 10-15% of RBG vines may have significant root escape. A separate issue is the over-stressing of vines, observed to a lesser extent in RBG-CC



treatment combinations. Figure 19 below displays the symptoms of this over-stressing, with micro-canopies and substantially reduced yields. This vegetative stunting and consequent yield reduction was observed in about 3% of RBG-CC vines, or approximately 1% of all RBG vines.



Figure 19. Stunted canopy in RBG-CC treatment (right) vs. healthy canopy (left). Both vines are RBG-CC (Root bag restricted and under-trellis cover crop).

## Conclusion

Our results demonstrated that the three treatments used in this comprehensive study have the ability to reduce overall vine size and vigor. The reduction of vegetative growth created more open canopies that required less management labor, increased fruit sunlight exposure, stimulated the rate of fruit development, and increased harvest fruit quality. Root restriction was the most effective of these treatments, while under-trellis ground cover and rootstock were less significant. Root restriction using fabric root bags consistently reduced canopy growth, decreased vine water status, decreased time necessary for common canopy management tasks, and increased the concentration of soluble solids and color compounds in Cabernet Sauvignon. It is important to note that while canopy size and total vine capacity were reduced, total yield was not affected, thus crop load was significantly increased. Under-trellis cover cropping was also effective at reducing vegetative growth and increasing the quality of harvested fruit, exhibiting greater soluble solid concentration, anthocyanins, and total phenolics. In addition to these effects, under-trellis cover crops also reduced yeast assimilable nitrogen and total crop yield, which is likely a result of reduced water supply and associated N uptake. Rootstock was much less significant in altering canopy size and fruit composition. However, 420-A was identified as having significantly reduced potassium uptake, resulting in lower concentrations in harvest juice. Future work relating to root restriction should focus on designing a more feasible way to implement root restricting effects. The cost of installing vines in fabric root bags may be too high to justify commercial application and the long-term durability (>10 yrs.) of these bags is unknown. Additionally these effects need to be observed on multiple varieties planted at different sites. Nevertheless, the treatment effects here may provide options and strategies for vineyards on potentially high-vigor sites.

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