

Evaluation of Nitrogen Management Schemes upon Vine Performance in Cover Cropped Vineyards

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ABSTRACT

Vineyards in the Eastern United States are often prone to excessive vegetative growth. In order to suppress excessive vine vigor, many viticulturists have employed cover cropping strategies. Cover crops provide a myriad of agronomic benefits, however they are known to compete with the vine for water and nutrients. Due to the widespread use of cover crops in Eastern vineyards, many vineyards experience nitrogen (N) deficiencies in both the vegetative vine tissue and yeast assimilable nitrogen (YAN) in the juice. Soil applications of calcium nitrate and foliar applications of urea were assessed as a means of vineyard N amelioration at cover cropped sites comprised of Petit Manseng and Sauvignon blanc (*Vitis vinifera* L.). Perennial White and Crimson clover cover crops and foliar urea applications were also used in a Vidal blanc (*Vitis spp.*) vineyard. Treatments were imposed in the Sauvignon blanc vineyard for five years. The Petit Manseng and Vidal blanc vineyards were subjected to treatments for two years. Soil-applied N at bloom was most effective at increasing leaf petiole N at véraison, season-long chlorophyll content index (CCI), vine capacity and fruit yield. Fruit yield was increased due to more berries per cluster and greater berry weights. Increased rates of soil-applied N decreased the fruit weight:pruning weight ratio. Clover cover crops offered little to no benefit as a N source in the two-year period of evaluation. None of the N management schemes negatively impacted canopy density, fruit zone light interception, or botrytis bunch rot incidence. The combination of both a soil-applied and foliar-applied N fertilizer may be the most effective means to increase both vine capacity and YAN in vineyards where vineyard floor cover crops are compromising vine N status.

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List of abbreviations

AREC – Alson H. Smith Jr. AREC (Petit Manseng)

CCI – Chlorophyll Content Index

CEFA – Cluster exposure Flux Availability

CEL – Cluster Exposure Layer

EPQA – Enhanced Point Quadrante Analysis

GDD – Growing Degree Days

GMV – Glen Manor Vineyards

ISV – Indian Springs Vineyard

LEFA – Leaf Exposure Flux Availability

LEL – Leaf Exposure Layer

OLN – Occlusion Layer Number

% PPF – Photosynthetic photon flux

Introduction

Excessive vine vigor is a common characteristic of vineyards in the Eastern United States. This excessive vegetative growth can lead to deleterious effects summarized in table 1.

Table 1. Deleterious effects of excessive vegetative growth of grapevines

Impact upon fruit	Increase/decrease	Reference
fungal disease pressure	increase	(Austin et al. 2011; English et al. 1993)
phenolic concentration	decrease	(Diago et al. 2012; Verzera et al. 2016)
methoxy-pyrazines	increase	(Šuklje et al. 2014)
thiols	decrease	(Šuklje et al. 2014)
pH	increase	(Bledsoe et al. 1988)
potassium concentration	increase	(Bledsoe et al. 1988)
malic acid:tartaric acid ratio	increase	(Hunter et al. 2004)
monoterpenes	decrease	(Skinkis et al. 2010)

Excessive vegetative growth has led vintners to employ various methods to reduce the vigor within their farming systems. In recent years, many vintners have begun using cover crops to reduce vine vigor (Giese et al. 2014; Morlat and Jacquet 2003; Tesic et al. 2007; Wheeler et al. 2005). Cover cropping has been demonstrated to have many beneficial effects upon the farming system, including acting as a nitrogen (N) and carbon (C) source (Ranells and Waggoner 1996) and providing alternative food source for mealybugs (Clearwater 2000). More benefits of cover crops have been summarised in table 2.

Table 2. Benefits of cover crops in farming systems

Impact upon soil	Increase/decrease	Reference
soil erosion	decrease	(Gaffney et al. 1991)
soil compaction during wet periods	decrease	(Louw and Bennie 1991)
water infiltration	increase	(Celette et al. 2005)
weed suppression	increase	(Baumgartner et al. 2008)
microbial biodiversity within the soil	increase	(Ingels et al. 2005)

Not all aspects of cover cropping are beneficial, however. The use of a cover crop can have significant drawbacks such as lower yields (Tesic et al. 2007), potential frost risk (Derr 2008), decreased

perennial N reserves (Celette et al. 2009), and depressed concentrations of Yeast Assimilable Nitrogen (YAN) (Gouthu et al. 2012).

Reductions in YAN concentrations represents a significant issue for vintners. Low YAN concentrations can lead to issues such as stuck and sluggish fermentations (Mendes-Ferreira et al. 2004), off odors (H₂S) (Jackson 2008), increased concentrations of higher alcohols (Webster et al. 1993) and lower concentrations of esters (Garde-Cerdán and Ancín-Azpilicueta 2008). A high YAN is not always beneficial either. If berry nitrogen is excessive it can lead to decreased anthocyanin concentrations (Keller and Hrazdina 1998), increased risk of botrytis infection (R'Houma et al. 1998), potential for atypical aging by indirectly increasing indole-3-acetic acid (Linsenmeier et al. 2004) and increased levels of ethyl carbamate (Bell and Henschke 2005). It is important for the vintner to achieve a YAN “balance” in the must in order to attain the highest wine quality.

Much of the previous viticulture research in relation to N has focused upon vine physiology as well as berry and must YAN; however, more research is needed to determine the impact of vine N supply upon secondary metabolites, such as flavor and aroma (Bell and Henschke 2005).

The widespread use of competitive crops in the Eastern United States has presented challenges to vintners as far as supplying adequate N to the vine and resulting must. The viticultural aim of this study was to examine methods by which vintners might attain adequate vine and must N through vineyard management schemes, while still maintaining the benefits of a cover crop and gaining an understanding as to how these treatments impact wine flavor and aroma. The methods employed in the current study included the use of leguminous cover crops, foliar applications of urea and traditional soil applications of calcium nitrate.

Petit Manseng and Sauvignon blanc represent the third and fifth most planted white wine grapes in Virginia respectively. On average, Petit Manseng and Sauvignon blanc are farmed at higher tonnages per acre and sold for higher prices per ton than the state’s most widely planted white variety,

Chardonnay (Virginia Wine Marketing Office 2014). Sauvignon blanc is also the third most planted white grape variety in the United States and is the fourth most popular white varietal wine in the country (California Department of Food and Agriculture 2014; Wine Institute 2014). The regional importance of Petit Manseng and the national and international economic significance of Sauvignon blanc warrant further investigation into how one might impact secondary metabolites within these varieties through N management schemes in the vineyard.

Varietal aromas (e.g. terpenes, norisoprenoids, pyrazines and thiols) have been demonstrated to be influenced by nitrogenous inputs to the musts and vines (Bell and Henschke 2005). Petit Manseng and Sauvignon blanc grapes and wines have been found to have significantly high concentrations of bound and free thiols (Darriet et al. 1995; Tominaga et al. 2000; Tominaga et al. 1996) These thiols contribute to the distinctive tropical fruit aromas of these wines and have been shown to be impacted by must nitrogen concentrations (Choné et al. 2006; Dufourcq et al. 2009; Lacroux et al. 2008; Peyrot des Gachons et al. 2005).

As was previously mentioned, the vegetative growth is relatively high in the Eastern United States. Higher leaf area and the fruit shading that results can increase concentrations of methoxypyrazines and depress the concentration and/or perception of the tropical fruit aromas of the volatile thiols (Šuklje et al. 2014; Van Wyngaard 2013). Although a survey of thiol concentrations in Virginia wines has not been carried out, one has been conducted in New York and the researchers found that the Sauvignon blanc wines from New York had considerably lower concentrations of volatile thiols compared to wines from other regions, including New Zealand (Musumeci et al. 2015). New Zealand Sauvignon blanc represents ~30% of the US Sauvignon blanc market, it sells for a premium price point and the market equity of these wines has been increasing dramatically in recent years (New Zealand Winegrowers 2013, 2014).

One of the distinguishing features of New Zealand Sauvignon blanc is its high concentration of thiols (Benkwitz et al. 2012b). There are many excellent Sauvignon blanc wines with low concentrations of thiols. If the concentration of these compounds is too high, it may be considered unpleasant to some consumers. However, it may be to the advantage of American vintners to devise strategies to increase the concentration of thiols in their Sauvignon blanc wines in order to remain qualitatively competitive with the wines from New Zealand.

Review of Literature

Varietal aroma

Wine flavor originates from many different sources. These can be considered “primary” or “varietal” aromas which arise as either free or bound compounds within the grape itself, “secondary” or fermentation aromas which develop during fermentation and “tertiary aromas” which develop as a wine ages in barrel and bottle. Secondary aromas were previously reviewed by the author and will not be covered here (Moss 2014a, 2014b, 2014c).

Varietal aroma includes classes of compounds such as terpenes, C13-norisoprenoids, methoxypyrazines and thiols. Grapes do not have exclusive aromatic compounds which are specific to the cultivar, rather each grape has a multitude of flavor and aroma compounds in a complex matrix which interact to deliver the unique aromatic character of the cultivar. Varietal aroma does not seem to be directly associated with sugar accumulation but is influenced by vintage weather and viticultural practices (González-Barreiro et al. 2015). Many of these aromatic compounds are glycosidically bound or form amino acid conjugates in the grape itself and are released during alcoholic fermentation (Park et al. 1991; Peyrot des Gachons et al. 2000).

The biosynthesis and effect of N nutrition upon terpenes and methoxypyrazines have been previously discussed by the author and will not be mentioned here (Moss 2016).

C₁₃-Norisoprenoids

Norisoprenoids are formed from the degradation of carotenoids, which are formed via the non-mevalonate pathway (MEP), and are responsible for a great diversity of aromas ranging from those perceived as flowery and fruity to petrol and kerosene (see table 4) (Robinson et al. 2014).

Norisoprenoids are found in nearly all grape varieties and are known to be important contributors to the aromas of wines made from grapes including Riesling, Chardonnay, Merlot, Syrah, Cabernet Sauvignon, Sémillon and Sauvignon blanc (Arrhenius et al. 1996; Benkwitz et al. 2012a; Ferreira et al. 2000; Gürbüz et al. 2006; Mayr et al. 2014; Sefton et al. 1996; Simpson and Miller 1984).

β -Damascenone and β -ionone are largely considered to be the most important C₁₃-Norisoprenoids due to their low sensory thresholds and relatively high concentrations in wines (González-Barreiro et al. 2015).

Table 3. C₁₃-norisoprenoids and their corresponding aromatic descriptors

Compound	Aroma descriptors
β -Ionone	Violet ^a
β -Damascenone	Bark, canned peach, baked apple, dry plum ^b
1,1,6-trimethyl-1,2-dihydronaphthalene (TDN)	Petrol ^c , Kerosene ^d
(<i>E</i>)-1-(2,3,6-trimethylphenyl)buta-1,3-diene (TPB)	Floral, geranium, tobacco, insecticide ^e

^a(Fretz et al., 2005) ^b(Li et al., 2008) ^c(Sacks et al., 2012) ^d(Ross et al., 2014) ^e(Janusz et al., 2003)

The norisoprenoids can be formed through several different mechanisms. β -ionone is known to arise from thermal degradation and photo-oxygenation of carotenoids (Isoe et al. 1969; Kanasawud and Crouzet 1990). A carotenoid cleavage dioxygenase, CCD1 has been found to cleave β -carotene at carbons 9,10 and 9',10' and form β -ionone in other crops (Ibdah et al. 2006; Lashbrooke et al. 2013; Simkin et al. 2004). However, some researchers found that CCD1 was not able to use β -carotene as a substrate in grapes to form β -ionone (Gunata 2013; Mathieu et al. 2005). Another carotenoid cleavage dioxygenase, CCD4, has been found to cleave β -carotene and yield β -ionone in other plants (Huang et al. 2009; Rubio et al. 2008). More research into the functional characterization of CCDs may be necessary to attain a better understanding of their action and specificity.

The biosynthesis of β -damascenone is extraordinarily complex and varied. The synthesis, sensory impact, occurrence and fate of β -damascenone has been reviewed by Sefton et al. (2011). β -damascenone has been found to be directly synthesized from neoxanthin via thermal oxidation (Bezman et al. 2005). The synthesis of β -damascenone is the result of bio-oxidative cleavage of a carotenoid substrate (neoxanthin) followed by a series of enzymatic transformations and finally an acid-catalyzed conversion to form β -damascenone (Sefton et al. 2011; Winterhalter and Gok 2013). 1,1,6-Trimethyl-1,2-dihydronaphthalene (TDN) and (*E*)-1-(2,3,6-trimethylphenyl)buta-1,3-diene (TPB) are also formed through a degradation of a parent carotenoid followed by further chemical conversion (Mendes-Pinto 2009; Winterhalter and Gok 2013).

It is interesting to note that although β -damascenone has an extremely low aroma threshold (0.05 $\mu\text{g/L}$), it may only play an indirect role in overall wine aroma even when its concentration is greater than the sensory threshold (Bindon et al. 2014; Guth 1997; Pineau et al. 2007). In a study upon interaction effects of fruity aromas in wine, it was found that norisoprenoids can enhance yeast derived esters at low concentrations and can provide an aromatic impact of raisin or dried plums at high concentrations (Escudero et al. 2007). Further study and clarification of the interactions between the wine matrix and aromatic compounds may allow for more reliable viticultural recommendations based upon aromatic optimization.

Vine nutrition and C_{13} -norisoprenoids

Little is known about the influence of vineyard N upon the formation of norisoprenoids (Burin et al. 2015; Linsenmeier and Lohnertz 2007). N fertilization has been demonstrated to increase carotenoids in grape leaves and this may also apply to fruit (Keller and Torres-Martinez 2002). It has been found that increased concentrations of leaf N correlate with an increased concentration of carotenoids in the leaf tissue (Chen and Cheng 2003a). Linsenmeier and Lohnertz (2007) evaluated norisoprenoid concentrations in wines made from Riesling vines which had been subjected to 0, 60 and 150 kg N/ha for

over a decade. Over the course of this study, increased N fertilization in the vineyard led to a decrease in TDN concentrations in the wine. Conversely, an increase of β -damascenone in response to increased N fertilization was reported by Linsenmeier and Lohnertz (2007). However, across vintages, only β -damascenone was found to be positively correlated with must N ($\alpha = 0.1\%$). β -damascenone was also found to significantly increase with the foliar application of a commercial N fertilizer in two applications at véraison which resulted in a total N application of 900g/ha (Garde-Cerdán et al. 2015). Increasing YAN concentrations in must through the addition of di-ammonium phosphate has also been found to correlate with higher concentrations of C_{13} -norisoprenoids in wine (Vilanova et al. 2012). The increase in must N status may increase the activity of yeast glycosidases which can result in a greater release of glycoconjugated norisoprenoids.

Thiols

Thiols are any organic compound which contain an –SH group. To date, research on thiols has focused primarily on their negative impact upon wine quality in the form of secondary odors formed by yeast (e.g. H_2S) and tertiary odors (e.g. mercaptans, dimethyldisulfide and thioacetic acid esters). These aromas are considered deleterious to wine quality and can impart aromas which have been described as rotten egg, onion, cabbage, burnt match and cabbage. However, not all thiols are associated with negative wine aromas.

The existence of positive volatile thiols in wine is a relatively new discovery, with the first being 4-mercapto-4-methylpentan-2-one (Darriet et al. 1995). Three positive volatile thiols have been identified. These are 4-mercapto-4-methylpentan-2-one (4MMP), 3-mercaptohexan-1-ol (3MH) and an acetate of 3MH, 3-mercaptohexyl acetate (3MHA) (Tominaga et al. 1996; Tominaga et al. 1998).

The varietal thiols are important to the aromas of wines made from white varieties such as Sauvignon blanc, Petit Manseng, Gewürztraminer, Riesling, Colombard, Semillon, Koschu, Niagra and Cayuga White (Kobayashi et al. 2010; Musumeci et al. 2015; Tominaga et al. 2000) Although most of the research has

focused upon thiols in Sauvignon blanc, due its significantly high concentrations of thiols, they are also present in red grapes above sensory thresholds. Negrette, Cabernet Sauvignon, Merlot, Syrah and Grenache are all red grapes in which the volatile thiols have been identified in concentrations greater than the sensory threshold (Murat et al. 2001; Rigou et al. 2014; Rodriguez-Bencomo et al. 2009). The aromatic descriptors of the varietal thiols have been summarized in table 6.

Table 4. Aroma descriptors of key varietal thiols found in grapes and wine

Compound	Aroma description
4-mercapto-4-methylpentan-2-one (4MMP)	box tree ^a , broom ^a , passion fruit ^b , black currant ^b
3-mercapto-hexan-1-ol (3MH)	passion fruit ^c , gooseberry ^b , grapefruit ^c and guava ^b
3-mercapto-hexylacetate (3MHA)	passion fruit ^a , box tree ^a

^a(Dubourdieu et al. 2006) ^b(Coetzee and du Toit 2012) ^c(Tominaga et al. 1998)

3MH and 4MMP are found within the juice as non-volatile cysteinylated (Cys-3MH and Cys-4MMP) or glutathionylated conjugates (Glu-3MH and Glu-4MMP) (Darriet et al. 1995; Tominaga et al. 1998). 3MHA is a product of fermentation as a result of an enzymatic esterification of 3MH by acetyltransferase with acetic acid and is not found in the grape (Swiegers et al. 2007). The uptake of the conjugated thiols is thought to occur via amino acid uptake pathways in yeast (Subileau et al. 2008). Once inside the yeast cell, the amino-thiol junction is cleaved by the carbon-sulfur β -lyase enzyme that is present in some wine yeasts (Coetzee and du Toit 2012).

The exact mechanisms by which amino acid-thiol conjugates are formed is not fully understood. Capone et al. 2010 postulate that the formation of Glu-3MH is likely due to conjugation of glutathione (a derivative of glutamic acid, cysteine and glycine) and (*E*)-2-hexenal. It has also been postulated that the Glu-3MH is a precursor to Cys-3MH.

S-glutathione conjugates are known to be involved in the detoxification of exogenous compounds in plants, including herbicides such as 2,4-D and atrazine (Anders et al. 1988). In the plant, the toxic compound is bound to glutathione and then degraded by γ -glutamyltranspeptidase. Through the removal of glutamic acid, a carboxypeptidase and glycine, a cysteinylated thiol conjugate is formed

(Peyrot des Gachons et al. 2002). The enzymes responsible for this transformation have been identified in grapes and the genes which regulate them are known to be up regulated by environmental stresses such as UV-C irradiation, water deficit, and cold/heat shock (Kobayashi et al. 2011).

The conversion of the glutathionylated and cysteinylated precursors of volatile 3MH by yeast has been evaluated by adding the conjugated form of each to a synthetic grape juice medium and fermenting it with a known thiol releasing yeast, *Saccharomyces Cerevisiae* VL 3 (Laffort) (Winter et al. 2011). The researchers then quantified the molar conversion of the conjugated form of 3MH into the volatile forms. They found that the molar conversion of cys-3MH was 2 times greater than that of Glu-3MH. However, it has been found that Glu-3MH can be present in musts in concentrations up to 37 times greater than Cys-3MH (Capone et al. 2010), attaining a more thorough understanding of the enzymatic conversion of Glu-3MH to Cys-3MH and how viticultural and enological factors influence this reaction might allow for managerial activities which could increase the concentration of the volatile thiol in the resulting wine.

Volatile thiols in wine are not just related to their conjugated precursors in the juice. A study was conducted in which the concentrations of Glu-3MH and Cys-3MH were determined and related to the final concentration of the corresponding volatile thiols in the finished wine (Pinu et al. 2012). Of the 55 wines made, the researchers found no correlation between the thiol conjugates found in the juice and the volatile thiols in the resulting wines. Capone et al. (2011) also found there to be no significant relationship between 3MH conjugated precursors and volatile 3MH. This suggests that there are other means by which volatile thiols are formed during alcoholic fermentation.

It has been demonstrated that volatile 3MH and 4MMP can be formed from other carbonyl compounds such as *E*-2-hexenal and mesityl oxide (Schneider et al. 2006). These researchers hypothesize that 3MH and 4MMP can be formed indirectly by the bonding of the carbonyl with a cysteine molecule followed by a conversion to the volatile thiol by yeast. They further theorized that

thiols may be formed directly by a 1, 4 addition of H₂S to conjugated carbonyls followed by a reduction step to form the volatile thiol.

Viticultural practices are known to influence the concentrations of thiols. The conjugated thiols seem to increase from véraison until harvest in Sauvignon blanc (Peyrot des Gachons et al. 2005; Roland et al. 2010). It is not understood what impacts the rate of formation of thiol precursors during ripening, as research has demonstrated this to be highly variable. Roland et al. (2010) and Capone et al. (2011) both found dramatic increases in thiol precursor concentrations from 1 to 2 weeks before commercial harvest of Sauvignon blanc. However, Roland et al. (2010) found that while Cys-3MH increased dramatically 7 days before and after harvest for Sauvignon blanc, Glu-3MH concentration differed slightly. The researchers also demonstrated that one of the vineyard sites had its lowest concentration of thiols on the same day that another vineyard site had its highest concentration of thiols. Peyrot des Gachons et al. (2005) found that conjugated 3MH did not change its concentration very dramatically from véraison until harvest, but that conjugated 4MMP increased steadily from véraison onward and decreased prior to harvest. More research is needed to understand the mechanisms which impact the rate of thiol development, so as to provide vintners with the ability to make management decisions from véraison to harvest that will have a positive influence upon potential flavor and aroma.

Vine water status has also been shown to have an impact upon thiol precursors in the fruit (Peyrot des Gachons et al. 2005). Peyrot des Gachons et al. (2005) found limited thiol production under severe water deficits, but an increase in thiol conjugates within the fruit under moderate water stress.

One of the defining characteristic aromas of Sauternes (wines made from botrytized Semillon, Sauvignon blanc and Muscadelle) is grapefruit and tropical fruits. Thibon et al. (2009) found that there was up to a 275-fold increase in the concentration of Cys-3MH in Sauvignon blanc juice from botrytized fruit when compared to uninfected fruit. The authors suggested that *Botrytis cinerea* was not responsible for the production of Cys-3MH, but rather that it stimulated its formation via metabolic

pathways in the grape itself. The 3-MH precursors in botrytised Sauterne juice was also correlated to significantly higher volatile 3MH in the resulting wine (Sarrazin et al. 2007). Sarrazin et al. (2007) found a 12 – 60 fold increase of 3MH concentration in wines made from botrytised fruit. The amount of increase was dependent upon the stage of botrytis infection. The researchers suggested that the increase was due to the increased concentration of the thiol precursors in the juice of botrytised Sauvignon blanc and Semillon. The correlation between precursors and volatile thiols in the wine was in direct contradiction with other research on this subject and warrants further study.

Vine nutrition and thiols

Nitrogenous fertilization in the vineyard has been demonstrated to have a significant impact upon the resulting volatile thiol concentration in the wine and the thiol conjugates found in the fruit. Choné et al. (2006) applied 60 kg of N/ha to a Bordeaux vineyard that was historically low in N. The fruit of the control had a YAN concentration of 29 ppm at harvest, whereas the fruit from the treated vines had a YAN concentration of 174ppm. All of the cys- conjugate thiols increased, but not in the same fashion. Cys-4MMP increased by 76% and Cys-3MH increased by 341%. The researchers found a 30% reduction in phenols with a dramatic 670% increase in gluathione. Polyphenol quinones can react with thiols, resulting in their oxidation and loss of aromatic potential (Blanchard et al. 2004). Glutathione can prevent oxidation of thiols by reacting with quinones to form a stable glutathionylated quinone adduct (Ugliano et al. 2011). Therefore, thiols are less likely to oxidize in wines made from juices of higher N concentrations.

Foliar applications of N in the form of urea have been found to be an effective means of increasing berry and must YAN concentrations when applied immediately around véraison (Dufourcq et al. 2009; Hannam et al. 2016; Hannam et al. 2014; Lacroux et al. 2008; Lasa et al. 2012). This methodology can increase berry YAN without the deleterious effects of increased vegetative growth. In their study of foliar nitrogen applied to Sauvignon blanc, Colombard, Gros Manseng and Négrette,

Dufourcq et al. (2009) observed significant increases in berry YAN with applications of urea. However, the modulations were variable and in some cases modulation was not demonstrated, illustrating the unpredictable variability of plant response to fertilization. Dufourcq et al. (2009) found that musts with higher YAN concentrations correlated with higher 3MH and 3MHA concentrations in the wine.

Dufourcq et al. (2009) also evaluated the impact of applying micronized sulphur (S) and urea together as foliar sprays. A synergistic effect between foliar N and S applications in wheat has been found (Tea et al. 2007). Tea et al. (2007) found that when foliar N and S were applied together, the uptake of both nutrients was better than when each was applied alone. Dufourcq et al (2009) did not find an increase in YAN concentrations with the N + S foliar spray treatment in relation to just an application of N. They found a 3 – 12 fold increase in total thiols in the plots sprayed with N and S compared to the control.

Lacroux et al (2008) applied the following treatments to a Sauvignon blanc vineyard in Bordeaux:

1. Control: no fertilization
2. 30 kg of N/ha applied to the soil after flowering
3. 10 kg of N/ha as a foliar urea spray in two applications before véraison
4. 10 kg of N/ha as a foliar urea spray + 5 kg S/ha as micronized sulphur in two applications prior to véraison

Wines were made from these treatments and evaluated both chemically and sensorially.

Lacroux et al. (2008) found that glutathione concentrations were significantly higher in foliar N and foliar N + S treatments than in the soil treatment or control. However, the foliar N + S treatment did not yield higher glutathione concentrations than foliar N alone. Nitrogen assimilation was not enhanced by N and S co-application. Volatile thiols in the wine did not increase from the soil N application when compared to the control. 4MMP increased in the foliar N treatment when compared to the control, but

this was the only volatile thiol to increase in the foliar N treatment when compared to the control. However, 3MH, 3MHA and 4MMP were all at the highest concentrations in wines produced from the N + S treatment. A tasting panel found that the N + S treatment was more aromatically intense when compared to the foliar N alone and the foliar N treatment was more intense than the soil N treatment and control. Soil N and control wines could not be differentiated by the tasting panel in that study, which was only conducted in one vineyard and with only one variety. A more robust study with more sites and varieties should be conducted to verify these results.

An increased tropical fruit aroma was described in wines made from Petit Manseng to which a foliar nitrogen and sulfur treatment was applied. However, in this study, volatile thiols were not measured and it could not be determined whether the increased tropical fruit aromas were coming from the volatile thiols and/or esters (Kelly 2013).

The mechanisms behind the apparent increase in volatile thiols associated with N+S foliar treatments has not been elucidated. It would seem that the conjugated thiols do not form a major source of the volatile thiols found in the resulting wine (Pinu et al. 2012). However, cysteine plays an important role in the formation of the volatile thiols during fermentation (Schneider et al. 2006). It may be possible that the foliar application of N and S through foliar fertilization increases the concentration of cysteine and/or other carbonyl compounds which are able to be transformed by yeast to form volatile thiols during fermentation. To date, the studies which have demonstrated increased thiol concentrations with N + S foliar applications have not provided an explanation as to the mechanism behind this result.

Enological factors which influence the concentrations of conjugated precursors and volatile thiols have been reviewed by the author in a previous article and will not be reviewed here (Moss 2015).

Materials and methods

Sites and treatments

Three vineyard sites with a history of low vine and juice nitrogen were chosen for this study. Two of the sites, Glen Manor Vineyards (GMV) and Indian Springs Vineyard (ISV), were commercially managed. The third site, The Alson H. Smith Jr. Agricultural Research and Extension Center (AREC), was managed as a commercial vineyard throughout the course of the study. AREC was the location of two separate experiments, AREC 1 and AREC 2.

GMV

GMV was located in Front Royal, VA (38°50'23.8"N 78°13'42.2"W). The site was planted in 1995 with Sauvignon blanc (*V. vinifera*) grafted to 3309C (*V. riparia* × *V. rupestris*) supported by an open Lyre trellis. The site was on a deep and well drained Myersville-Catoctin silt loam complex with a 15% South-West facing slope. The rows were in a North-South orientation with a spacing of 3.35 meters between the rows and 2.13 meters between plants within the row, resulting in a planting density of 1401 vines/ha. The intra-row and inter-row was cover-cropped with red fescue (*Festuca rubra*) and tall fescue (*Festuca arundinacea*), respectively.

Treatments have been annually imposed since the 2010 growing season. Three treatments and an unfertilized control were imposed upon six replicates of three vine plots with three-vine border plots separating treatments. Treatments included nitrogenous fertilizer applied to the soil in the form of calcium nitrate at rates of 30 kg N/ha (30 N soil) and 60 kg N/ha (60 N soil). Both soil N treatments were applied at bloom, however the 60 kg N/ha treatment was split equally and the second dose was applied approximately 4 weeks later to minimize N lost to leaching. The third treatment consisted of 30 kg N/ha applied as a foliar urea spray (30 N foliar). The foliar urea treatment was split into six equal applications beginning at bloom and implemented every 7-10 days. Each foliar treatment was applied with a backpack sprayer at the equivalent of 900 liters of water/ha.

AREC 1 and AREC 2

The AREC vineyard was located near Winchester, VA (39°06'43.4"N 78°17'04.9"W). AREC 1 and AREC 2 were planted with Petit Manseng (*V. vinifera*) on 420 A rootstock (*V. berlandieri* × *V. riparia*) in 2007 and 2009, respectively. Vines at AREC were supported by a Vertical Shoot Positioned (VSP) trellis. The soil was a deep and well drained Frederick-Poplimento sandy loam on a 2% easterly grade. Rows were in a northeast-southwest orientation. The vine rows were separated by 3 meters with 1.5 meters between plants within the row, resulting in a vine density of 2,222 vines/ha. The inter-rows were sown in tall fescue (*F. arundinacea*) and orchard grass (*Dactylis glomerata*), maintained by mowing throughout the growing season. The intra-row of AREC 1 was cover-cropped in creeping red fescue (*F. rubra*). The intra-row of AREC 2 was bare and maintained with glyphosate.

In both 2014 and 2015, four treatments and an unfertilized control were imposed at AREC 1 over five replicates in a completely randomized design. Experimental units in both AREC 1 and 2 consisted of five-vine plots. Treatments at AREC 1 were separated by five border vines. Treatments consisted of soil-applied nitrogen in the form of calcium nitrate at rates of 30 kg N/ha (30 N soil), 45 kg N/ha (45 N soil) and 60 kg N/ha (60 N soil). Each soil treatment was imposed at bloom. The 60 N soil treatment was split into equal applications, the first being applied at bloom and the second application was imposed about 4 weeks later in order to avoid leaching. The fourth treatment consisted of 45 kg N/ha as calcium nitrate applied at bloom and a split application, separated by 7-10 days of 15 kg N/ha at véraison as a foliar urea spray at a rate of 900 liters of water/ha (45 N soil + 15 N foliar).

AREC 2 was established in 2014 with five replicates of two treatments and an unfertilized control in a randomized complete block design. One treatment consisted of 15 kg N/ha applied as foliar urea two weeks prior to véraison in two equivalent split applications separated by 7-10 days (15 N foliar). The second treatment was 15 kg N/ha applied as foliar urea, in conjunction with 5 kg S/ha as micronized sulfur applied two weeks prior to véraison in two equivalent split applications separated by

7-10 days (15 N foliar + 5 S foliar). Each foliar treatment was applied with a backpack sprayer at a water rate that was equivalent to 900 liters/ha.

ISV

ISV was located in Shenandoah County, VA (38°55'55.0"N 78°33'42.3"W) and was planted with own-rooted Vidal blanc (*Vitis ssp.*) with an inter-row spacing of 2.74 meters and an inter-plant spacing of 2.13 meters, resulting in a density of 1712 vines/hectare. The vines were trellised to VSP running in a slightly northwest-southeasterly direction on a southern facing slope with a 2-7% gradient. The soil at ISV was a well-drained Edom silty clay loam. The inter-rows were cover-cropped with a regularly mown voluntary sward. Historically, the vines have had an intra-row cover crop, but this was removed for this study and replaced with clover or maintained bare with herbicide. Due to the results from soil samples taken at the site, triple superphosphate (P_2O_5) was applied at a rate of 50 kg P/ha prior to the imposition of treatments in 2014.

Each experimental unit consisted of four vines replicated four times in a randomized complete block design. Experimental units were separated by four vine border plots. Four treatments and two controls were established at ISV. Crimson clover (*Trifolium incarnatum*) and Dutch White clover (*T. repens*), inoculated with NITRO-COAT® rhizobium (Outsidelands.com Inc., Independence, OR), were sown in all treatments at rates of 33.6 kg/ha and 15.7kg/ha, respectively. In 2014, due to project timing, seeds were sown in May. Due to difficulty in cover crop establishment in 2014, seeds were re-sown in 2015. In 2015, the cover crop was frost-seeded in March in order to assist with soil incorporation. Seeds for each treatment were broadcast by hand onto bare soil and incorporated down to a depth of approximately one centimeter using rakes.

The following treatments were applied at ISV: under-vine Crimson clover (Crimson), under-vine White clover (White), Crimson clover + 10 kg N/ha as foliar urea (Crimson + 10 N foliar), White clover + 10 kg N/ha as foliar urea (White + 10 N foliar).

Clover plots were maintained throughout the season as needed through spot applications of Fluzifop-P-butyl and glyphosate.

Two controls were established at ISV to evaluate the efficacy of the cover cropping strategies in relation to industrially standard practices. The two controls at ISV were as follows: bare intra-row and 15 kg N/ha applied to soil (15 N soil), bare intra-row and 15 kg N/ha applied to soil and 10 kg N/ha applied to the foliage (15 N soil + 10 N foliar).

The intra-rows of the 15 N soil and 15 N soil + 10 N foliar treatments were kept bare with the application of glyphosate throughout the season. All soil-applied nitrogen at ISV was in the form of calcium nitrate and was imposed at bloom. Foliar N treatments were applied with a backpack sprayer in the form of urea at the start of véraison in two equal split applications separated by 7-10 days at an equivalent water rate of 900 liters/ha.

Plant tissue analysis

Plant tissue analysis was conducted at each site in 2014 and 2015. Sixty petioles (thirty from each side of the canopy) attached to the first fully expanded leaf from the end of a count shoot were collected from each experimental unit at 90-100% véraison. The petioles were dried in an oven at 60°C for 24 hr and sent to the Pennsylvania State University Agricultural Services Laboratory (University Park, PA) in 2014 and to Waypoint Analytical (Richmond, VA) in 2015 for an analysis of mineral nutrients.

Chlorophyll content index

A chlorophyll meter (CCM-200 plus, Apogee Instruments, Logan, UT) was used to measure the chlorophyll content index (CCI). CCI was calculated by measuring the ratio of radiation transmitted via a 0.71mm² aperture through the leaf at wavelengths of 653nm (red) and 931nm (near-infrared). The red wavelength was strongly absorbed by chlorophyll *a* and *b* and the near-infrared wavelength was used to compensate for physical differences in leaf tissue, such as tissue thickness.

CCI measurements were taken at GMV, ISV and AREC 1 in 2014 and 2015. CCI measurements were not taken at AREC 2, as foliar urea sprays were unlikely to significantly affect leaf chlorophyll content.

CCI measurements were taken three times through the growing season (pre-véraison, véraison and post-harvest) separated by approximately 30 days between measurements. Due to an equipment malfunction, no post-harvest measurement was made at GMV and ISV in 2014 and only a post-véraison measurement was made at ISV in 2014.

CCI readings were made on fully expanded leaves on count shoots, which were between the 5th and 8th nodal positions. Leaves were tagged upon first measurement and the same leaf was used in subsequent readings. Measurements were taken on the right side of the leaf in the third interveinal space from the leaf tip. These measurements were made once upon five random leaves throughout the experimental unit and readings were averaged.

Components of yield

Crop yield data was collected at all sites in all years. All clusters were harvested, counted and weighed on a per vine basis. From this information, total vine yield and the number of clusters per vine was determined. Cluster weights were calculated by dividing the mass of the harvested clusters by the number of clusters per vine. Berry weights were determined by dividing the mass of the berries collected for primary fruit chemical analysis by the number of berries within the sample. The number of berries per cluster was calculated by dividing cluster weight by berry weight. All data collected on a per-vine basis was averaged across each experimental unit.

Pruning weights

Pruning weights were collected in the winters of 2015 and 2016 for all sites. Pruning weights were recorded on a per vine basis using a hand held scale and averaged across each experimental unit.

Canopy architecture

The Enhanced Point Quadrat Analysis (Meyers and Vanden Heuvel 2008) was performed on every site at 90-100% véraison in 2014 and 2015. Canopy insertions were made with a metal rod every 30 cm within each experimental unit. Data were recorded as “gaps”, “leaves” or “fruit” as the rod contacted those features. Photosynthetic photon flux density (PPFD) was measured on cloudless days at solar noon (± 1 hr) using an AccuPAR ceptometer (AccuPAR80, Decagon Devices, Inc., Pullman, WA). The ceptometer readings were taken by placing the instrument within the fruit zone parallel to, and just above, the cordon. Once in the canopy, a vertical, east and west recording of photon flux was made and averaged. PPFD was recorded on a per vine basis. Unobstructed ambient light readings were taken in the inter-row prior to taking each set of canopy PPFD readings. Data were analyzed using a specially designed EPOA software package (Meyers and Vanden Heuvel 2008).

Cover crop measurements

Cover crop performance was assessed qualitatively and quantitatively at ISV in 2015. Each cover cropped experimental plot was assessed. Therefore, the 15 N soil and 15 N soil + 10 N foliar treatments were not assessed, as they were kept bare through the application of herbicide.

Square quadrats with an internal area of 0.25m^2 were placed equidistant between every vine in each experimental unit. A turf-grass stand density scale was modified (Morris 2001) and used for the visual assessment of stand density by three people, as used elsewhere (Giese et al. 2014). Stand density was ranked on a scale from one to six with the numerical ranks as follows: 1 = 76-100% invasive species/bare ground; 2 = 51-75% invasive species/bare ground; 3 = 26-50% invasive species or bare ground; 4 = 10-25% invasive species/bare ground; 5 = <10% invasive species/bare ground; 6 = 100% ground cover by cover crop.

Above ground biomass was sampled from the same quadrats that were used for visual stand density estimation. Sampling took place by cutting the vegetation within the quadrat with shears about

5 mm above the soil surface. The vegetation was then sorted into “weeds” and “cover crop” and dried in an oven for 48 hours at 60 °C. The dry weight of the biomass was then taken and averaged across each experimental unit, as used elsewhere (Giese et al. 2014).

Data analysis

Data were analyzed using JMP pro version 11 (SAS; Cary, NC). Two-way analysis of variance (ANOVA) was carried out on all data sets, where applicable. Model effects tested at AREC 2, GMV and ISV were treatment, block, year and the treatment*year interaction. Model effects tested at AREC 1 were treatment, year and treatment*year interaction.

One-way ANOVAs were carried out on all appropriate data sets in order to assess within year treatment differences. Model effects tested at AREC 2, GMV and ISV were treatment and block. The model effect tested at AREC 1 was treatment. A confidence level of 95% was used in all statistical analyses.

The means of all analyses containing three or more treatments were separated using Tukey’s HSD ($\alpha = 0.05$). The means of all analyses containing two treatments were separated using Student’s T-test ($\alpha = 0.05$).

Results

Temperature and rainfall

Total rainfall and heat summation for the 2015 growing season were slightly greater than in the 2014 season (Table 5). At the end of the growing season, the 2015 season had accumulated 156 more growing degree days than had accrued in 2014. The 2015 season also accrued 32mm more rain than the 2014 season (Table 5).

Table 5. Precipitation and heat accumulation at AREC vineyard site 2014-2015

Parameter/year	Apr	May	Jun	Jul	Aug	Sep	Oct	Total Apr-Oct
Rainfall (mm)								
2014	77	102	87	78	74	10	91	519
2015	87	92	122	54	31	96	68	551
GDD (<10°C)^a								
2014	83	249	363	397	356	286	124	1858
2015	98	300	363	424	409	316	103	2014

^a GDD (<10 °C) = growing degree days above 10 °C

Plant tissue analysis

GMV: Véraison petiolar N and Mg concentration at GMV were significantly increased by soil-applied N, with the highest N and Mg concentrations from the highest rate of fertilization (60 N soil) (Table 6 and 7). Phosphorus and K concentration in the petioles at véraison were slightly depressed by soil-applied N (Table 6 and 7). However, differences in petiolar K were only detected by the combined model (Table 6) and petiolar K was not significantly different between treatments within 2014 and 2015 (Table 7).

Petiolar P concentration was only significantly depressed by soil-applied N (60 N soil) in 2014 ($P < 0.05$). (Table 7). The higher soil treatment (60 N soil) depressed petiolar Mn and B concentrations (Table 8). However, within each year, N fertilization had no effect upon Mn concentrations and only significantly depressed B concentration in 2015 (Table 9). Year had a significant effect upon petiolar nutrient status for every nutrient except boron ($p < 0.05$) (Table 6 and 8).

AREC 1: Soil-applied N increased petiolar N concentrations at véraison (Table 10 and 11). The highest petiolar N concentration was found in the 45 N soil treatment in both 2014 and 2015 (Table 11). N applications depressed petiolar P at véraison (Table 10), but this reduction in P was only significant in 2015 ($p < 0.05$) (Table 13). The treatment-year interaction for P at AREC 1 was significant ($p < 0.05$). The 45 N soil + 15 N foliar treatment increased petiolar Ca concentration (Table 10). However, a significant increase in Ca concentration was not detected within each year (Table 11). Treatment did not have a

significant effect upon micronutrients at AREC 1 (Table 12 and 13). Year significantly affected all petiolar nutrients, except N ($p < 0.05$) (Table 10 and 12).

AREC 2: Petiolar nutrient status was not significantly affected by treatment (Table 14-17). Year had a significant effect upon petiolar nutrient concentrations of K, Mn, Fe, Cu, B, Zn and Na ($p < 0.05$).

ISV: Treatments at ISV did not have a significant effect upon petiolar N at véraison (Table 18 and 19).

Foliar N and clover cover crops significantly increased petiolar P concentrations at véraison ($p < 0.05$) (Table 18). However, significance between treatments with regards to petiolar P was not detected within each year (Table 19). The Crimson + 10 N foliar treatment significantly increased petiolar K concentrations relative to the 15 N soil + 10 N foliar and White + 10 N foliar treatments (Table 18).

However, this result was only significant in 2014 (Table 19). Treatment did not have a significant effect upon micronutrients (Table 20 and 21). Year had a significant effect upon every nutrient, aside from Mg ($p < 0.05$) (Table 18 and 20).

Table 6. GMV: Macronutrient composition of petioles at véraison in response to soil and foliar N fertilization (2014 – 2015)

Treatment ^{ab}	N%	P%	K%	Ca%	Mg %
Control	0.64 bc	0.38 a	4.81 a	1.21	0.52 bc
30 N soil	0.66 ab	0.23 bc	4.46 ab	1.26	0.59 b
60 N soil	0.67 a	0.18 c	4.03 b	1.21	0.67 a
30 N foliar	0.63 c	0.32 ab	4.58 a	1.19	0.49 c
Trt ^c	0.0457	0.0001	0.0017	ns ^d	<0.0001
Yr	<0.0001	0.0240	<0.0001	<0.0001	<0.0001
Trt × Yr	ns	ns	ns	Ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha = 0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of Year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 7. GMV: Macronutrient composition of petioles at véraison in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	N%		P%		K%		Ca%		Mg %	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	0.72 ab	0.56 ab	0.33 a	0.43	5.48	4.14	1.08	1.35	0.39 c	0.65 ab
30 N soil	0.75 ab	0.58 ab	0.20 bc	0.27	4.91	4.01	1.11	1.40	0.48 b	0.69 ab
60 N soil	0.76 a	0.59 a	0.16 c	0.20	4.48	3.59	1.07	1.35	0.58 a	0.77 a
30 N foliar	0.71 b	0.55 b	0.29 ab	0.35	5.07	4.10	1.05	1.34	0.40 c	0.57 b
Trt ^c	0.0254	0.0113	0.0007	ns ^d	ns	ns	Ns	ns	<0.0001	0.0152

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 8. GMV: Micronutrient composition of petioles at véraison in response to soil and foliar N fertilization (2014 – 2015)

Treatment ^{ab}	Mn ppm	Fe ppm	Cu ppm	B ppm	Al ppm	Zn ppm	Na ppm
Control	297 ab	23	18	31 a	6	55	47
30 N soil	300 ab	23	17	30 ab	4	54	46
60 N soil	258 b	23	16	28 b	5	55	47
30 N foliar	338 a	24	17	29 ab	5	65	46
Trt ^c	0.0115	ns ^d	ns	0.0021	ns	Ns	ns
Yr	<0.0001	<0.0001	<0.0001	ns	0.0015	0.0003	<0.0001
Trt × Yr	ns	ns	ns	ns	ns	Ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 9. GMV: Micronutrient composition of petioles at véraison in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Mn ppm		Fe ppm		Cu ppm		B ppm		Al ppm		Zn ppm		Na ppm	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	192	401	10	36	26	9	30	31 a	4	9	43	67	59	35
30 N soil	204	396	10	35	24	9	30	30 ab	4	4	47	61	56	35
60 N soil	171	345	10	37	23	9	29	27 b	4	6	43	67	57	37
30 N foliar	213	464	12	37	25	10	29	29 ab	4	6	45	84	59	33
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	0.0193	ns	ns	ns	ns	ns	Ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 10. AREC 1: Macronutrient composition of petioles at véraison in response to soil and foliar N fertilization (2014-2015)

Treatment ^{ab}	N%	P%	K%	Ca%	Mg %
Control	0.59 c	0.15 a	6.06	1.97 b	0.32
30 N soil	0.67 b	0.11 b	5.61	2.10 ab	0.33
45 N soil	0.74 a	0.09 b	5.72	1.96 b	0.39
60 N soil	0.67 b	0.10 b	5.47	1.98 b	0.35
45 N soil + 15 N foliar	0.69 ab	0.10 b	5.37	2.32 a	0.38
Trt ^c	<0.0001	<0.0001	ns	0.0055	ns
Yr	ns ^d	<0.0001	<0.0001	<0.0001	0.0004
Trt × Yr	ns	0.0070	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 11. AREC 1: Macronutrient composition of petioles at véraison in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	N%		P%		K%		Ca%		Mg %	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	0.60 b	0.59 b	0.10	0.20 a	6.49	5.64	1.75	2.20	0.29	0.35
30 N soil	0.65 ab	0.68 ab	0.09	0.15 ab	6.05	5.18	1.89	2.30	0.30	0.37
45 N soil	0.73 a	0.75 a	0.08	0.10 b	6.20	5.24	1.75	2.17	0.35	0.43
60 N soil	0.66 ab	0.68 ab	0.09	0.12 b	5.88	5.05	1.76	2.21	0.31	0.39
45 N soil + 15 N foliar	0.68 a	0.69 ab	0.08	0.12 b	5.90	4.85	2.08	2.56	0.33	0.44
Trt ^c	0.0025	0.0032	ns ^d	0.0010	ns	Ns	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 12. AREC 1: Micronutrient composition of petiole at véraison in response to soil and foliar N fertilization (2014 – 2015)

Treatment ^{ab}	Mn ppm	Fe ppm	Cu ppm	B ppm	Al ppm	Zn ppm	Na ppm
Control	177	24	7	26	7	59	105
30 N soil	177	19	7	27	5	56	104
45 N soil	189	20	7	27	6	57	111
60 N soil	187	19	7	25	4	56	101
45 N soil + 15 N foliar	206	18	7	26	6	59	110
Trt ^c	ns ^d	ns	ns	ns	Ns	ns	ns
Yr	<0.0001	<0.0001	<0.0001	<0.0001	0.0462	<0.0001	<0.0001
Trt × Yr	ns	ns	ns	ns	Ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 13. AREC 1: Micronutrient composition of petiole at véraison in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Mn ppm		Fe ppm		Cu ppm		B ppm		Al ppm		Zn ppm		Na ppm	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	124	231	14	34	3	12	21	30	7	7	47	72	151	60
30 N soil	131	223	8	31	4	11	24	31	6	4	48	64	152	56
45 N soil	137	241	8	33	4	11	23	31	6	6	47	67	160	62
60 N soil	145	228	10	28	3	11	22	29	6	3	43	70	149	54
45 N soil + 15 N foliar	163	250	7	29	3	11	22	30	6	5	47	70	159	62
Trt ^c	ns ^d	ns	ns	ns	ns	ns	Ns	ns	ns	ns	ns	ns	Ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 14. AREC 2: Macronutrient composition of petioles at véraison in response to foliar N fertilization with and without micronized sulfur (2014-2015)

Treatment ^{ab}	N%	P%	K%	Ca%	Mg %
Control	0.57	0.20	5.94	1.90	0.15
15 N foliar	0.58	0.23	5.93	1.93	0.15
15 N foliar + 5 S foliar	0.58	0.20	5.75	1.83	0.14
Trt ^c	ns ^d	ns	ns	ns	ns
Yr	ns	ns	<0.0001	ns	ns
Trt × Yr	ns	ns	ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 15. AREC 2: Macronutrient composition of petioles at véraison in response to foliar N fertilization without and without micronized sulfur by year (2014 and 2015)

Treatment ^{ab}	N%		P%		K%		Ca%		Mg %	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	0.58	0.56	0.19	0.22	6.85	5.02	1.87	1.94	0.14	0.15
15 N foliar	0.59	0.58	0.21	0.26	6.85	5.02	1.86	1.99	0.15	0.15
15 N foliar + 5 S foliar	0.59	0.58	0.21	0.20	6.60	4.90	1.76	1.90	0.14	0.15
Trt ^c	ns ^d	ns								

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 16. AREC 2: Micronutrient composition of petioles at véraison in response to foliar N fertilization with and without micronized sulfur (2014 – 2015)

Treatment ^{ab}	Mn ppm	Fe ppm	Cu ppm	B ppm	Al ppm	Zn ppm	Na ppm
Control	197	19	6	38	6	44	117
15 N foliar	172	24	7	37	6	47	113
15 N foliar + 5 S foliar	198	21	7	36	6	48	118
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns
Yr	<0.0001	<0.0001	<0.0001	0.0227	ns	0.0004	<0.0001
Trt × Yr	ns	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 17. AREC 2: Micronutrient composition of petioles at véraison in response to foliar N fertilization with and without micronized sulfur by year (2014 and 2015)

Treatment ^{ab}	Mn ppm		Fe ppm		Cu ppm		B ppm		Al ppm		Zn ppm		Na ppm	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	134	259	7	32	2	10	37	39	6	6	37	50	182	52
15 N foliar	114	231	7	41	3	11	35	38	5	7	42	51	174	52
15 N foliar + 5 S foliar	145	252	9	33	3	11	35	38	6	7	38	58	183	52
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 18. ISV: Macronutrient composition of Vidal blanc petioles at véraison in response to treatments (2014-2015)

Treatment ^{ab}	N%	P%	K%	Ca%	Mg %
15 N soil	0.66	0.30 b	1.81 abc	2.88	0.87
15 N soil + 10 N foliar	0.69	0.34 ab	1.54 bc	2.91	0.93
Crimson	0.65	0.42 a	1.69 abc	2.78	0.78
Crimson + 10 N foliar	0.64	0.41 a	2.11 a	2.97	0.80
White	0.65	0.37 ab	2.00 ab	2.82	0.88
White + 10 N foliar	0.67	0.40 a	1.28 c	2.88	0.90
Trt ^c	ns ^d	0.0056	0.0007	ns	ns
Yr	0.0001	0.0058	0.0060	<0.0001	ns
Trt × Yr	ns	ns	ns	ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 19. ISV: Macronutrient composition of Vidal blanc petioles at véraison in response to treatments by year (2014 and 2015)

Treatment ^{ab}	N%		P%		K%		Ca%		Mg %	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
15 N soil	0.68	0.65	0.34	0.26	1.70 ab	1.93	2.64	3.12	0.78 ab	0.96
15 N soil + 10 N foliar	0.71	0.68	0.37	0.31	1.41 ab	1.67	2.76	3.07	0.85 ab	1.01
Crimson	0.67	0.63	0.45	0.39	1.65 ab	1.74	2.6	2.96	0.84 ab	0.71
Crimson + 10 N foliar	0.68	0.61	0.41	0.40	1.92 a	2.31	2.8	3.14	0.73 b	0.86
White	0.69	0.61	0.4	0.33	1.84 ab	2.17	2.66	2.97	0.78 ab	0.98
White + 10 N foliar	0.68	0.65	0.43	0.38	1.01 b	1.56	2.67	3.10	0.98 a	0.83
Trt ^c	ns ^d	ns	ns	ns	0.0474	ns	ns	ns	0.0303	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 20. ISV: Micronutrient composition of Vidal blanc petioles at véraison in response to treatments (2014-2015)

Treatment ^{ab}	Mn ppm	Fe ppm	Cu ppm	B ppm	Al ppm	Zn ppm	Na ppm
15 N soil	67.68	20.59	6.11	35.68	5.53	65.91	68.14
15 N soil + 10 N foliar	65.59	22.69	6.74	38.96	6.73	70.68	67.48
Crimson	70.79	22.48	6.21	37.18	5.52	75.63	68.19
Crimson + 10 N foliar	70.21	22.71	6.77	37.99	5.87	75.44	64.00
White	74.82	20.31	5.93	37.74	3.86	73.90	68.45
White + 10 N foliar	81.54	23.49	6.57	39.88	4.95	75.09	70.02
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns
Yr	<0.0001	<0.0001	<0.0001	<0.0001	0.0202	<0.0001	0.0090
Trt × Yr	ns	ns	ns	ns	ns	ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 21. ISV: Micronutrient composition of Vidal blanc petioles at véraison in response to treatments by year (2014 and 2015)

Treatment ^{ab}	Mn ppm		Fe ppm		Cu ppm		B ppm		Al ppm		Zn ppm		Na ppm	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
15 N soil	37	98	9	32	4	8	39	32	5	7	56	76	69	68
15 N soil + 10 N foliar	43	89	11	35	4	9	44	34	4	9	61	81	72	63
Crimson	40	101	9	36	4	9	41	34	4	7	62	89	69	68
Crimson + 10 N foliar	40	101	10	36	4	10	41	35	4	7	64	87	68	60
White	48	102	10	31	4	8	41	34	4	3	62	86	74	63
White + 10 N foliar	51	112	9	38	4	10	45	35	4	6	66	84	78	63
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	Ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Chlorophyll content index

GMV: Soil-applied N at GMV significantly increased the season long CCI (Table 24). This result was significant in both 2014 and 2015 ($p < 0.05$) (Table 23). The highest rate of soil-applied N (60 N soil) resulted in the highest CCI values. Year had a significant effect upon CCI ($p < 0.05$) (Table 22).

AREC 1: The 45 N soil and 45 N soil + 15 N foliar treatments had significantly higher season-long CCIs than the control ($p < 0.05$) (Table 24). However, the season-long CCI was only statistically significant in the first year of the study (Table 25). In general, soil-applied N increased the season-long CCI values. Year had a significant effect upon CCI ($p < 0.05$) (Table 24).

ISV: Treatments had no significant effect upon the season-long CCI at ISV in 2015 (Table 26).

Table 22. GMV: Season long chlorophyll content index in response to soil and foliar N fertilization (2014-2015)

Treatment ^{ab}	Season long CCI
Control	14.13 c
30 N soil	16.09 b
60 N soil	18.35 a
30 N foliar	15.85 b
Trt ^c	<0.0001
Yr	0.0062
Trt × Yr	ns ^d

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha = 0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 23. GMV: Season long chlorophyll content index in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Season long CCI	
	2014	2015
Control	13.85 c	14.4 c
30 N soil	15.83 b	16.33 b
60 N soil	17.58 a	19.12 a
30 N foliar	15.37 bc	16.35 b
Trt ^c	0.0001	<0.0001

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 24. AREC 1: Season long chlorophyll content index in response to soil and foliar N fertilization (2014-2015)

Treatment ^{ab}	Season long CCI
Control	19.01 b
30 N soil	20.91 ab
45 N soil	23.02 a
60 N soil	20.45 ab
45 N soil + 15 N foliar	22.03 a
Trt ^c	0.0021
Yr	0.0008
Trt × Yr	ns ^d

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 25. AREC 1: Season long chlorophyll content index in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Season long CCI	
	2014	2015
Control	18.14 b	19.89
30 N soil	19.55 ab	22.28
45 N soil	22.07 a	23.97
60 N soil	19.32 ab	21.58
45 N soil + 15 N foliar	20.82 ab	23.25
Trt ^c	0.0269	ns ^d

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 26. ISV: Season long chlorophyll content index in response to treatments (2015)

Treatment ^{ab}	Season long CCI
15 N soil	20.20
15 N soil + 10 N foliar	20.53
Crimson	18.45
Crimson + 10 N foliar	18.32
White	19.60
White + 10 N foliar	19.60
Trt ^c	ns ^d

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Linear regression analysis

No correlation between petiolar N and season-long CCI was detected at GMV (Figure 1). There was a significant positive relationship between véraison petiolar N and season-long CCI at AREC 1 and ISV ($p < 0.05$) (Figure 2 and 3).

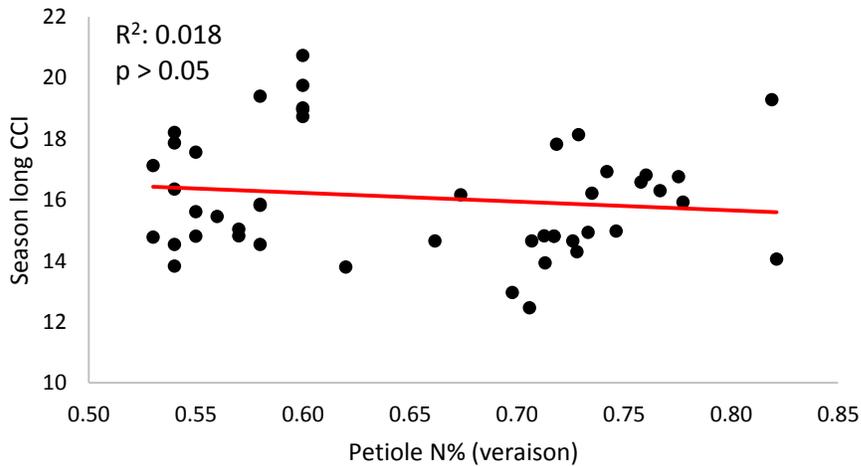


Figure 1. GMV: Regression analysis of season-long CCI and Petiole N% at véraison (2014-2015)

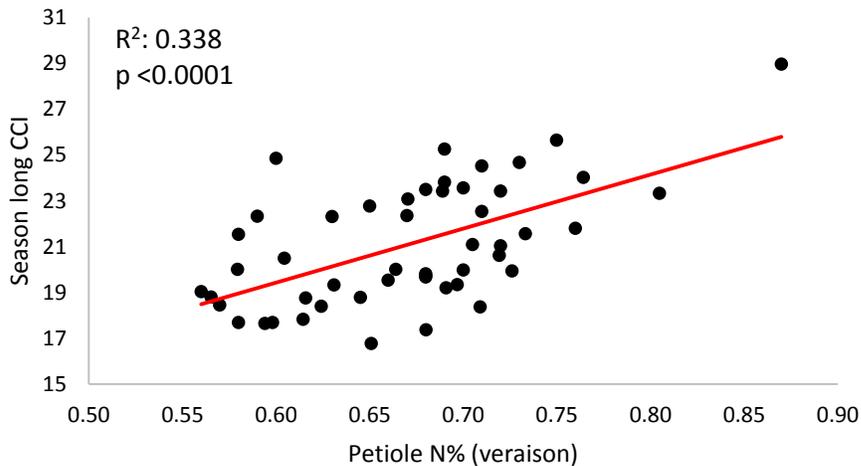


Figure 2. AREC 1: Regression analysis of season-long CCI and petiole N% at véraison (2014-2015)

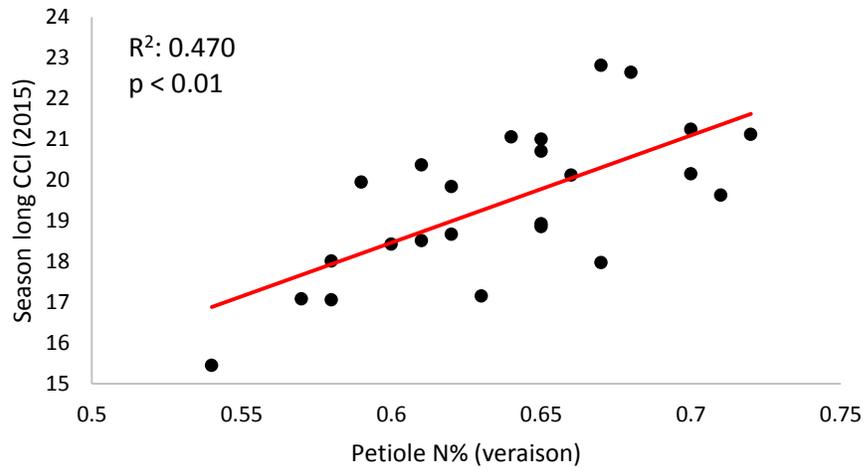


Figure 3. ISV: Regression analysis of Season long CCI and petiole N% at véraison (2015)

Components of yield and pruning weights

GMV: Relative to the control, soil and foliar-applied N significantly increased total vine yield, cluster weight, berry weight, berries per cluster and the yield to pruning weight ratio ($p < 0.05$) (Table 27). The most dramatic increase in total yield, cluster weight, berry weight and pruning weights came from the 60 N soil treatment (Table 27). The 60 N soil treatment also significantly reduced the yield to pruning weight ratio relative to the control and 30 N foliar treatment ($p < 0.05$) (Table 27). Total vine yield, cluster weight, berry weight, berries per cluster and pruning weights were significantly affected by treatment each year ($p < 0.05$) (Table 28). The 60 N soil treatment significantly lowered ($p < 0.05$) the yield to pruning weight ratio relative to the control and 30 N foliar treatment in 2014, but not 2015 (Table 28). Year had a significant effect upon total vine yield, clusters per vine and pruning weights ($p < 0.05$) (Table 27).

AREC 1: The highest berry weight came from the 60 N soil treatment (Table 29 and 30). However, this result was only significant in 2015 and there was a significant treatment-year interaction ($p < 0.05$) (Table 29 and 30). No other component of yield was affected by treatment. Pruning weights and the yield to pruning weight ratio was unaffected by treatment. Year had a significant effect upon total vine yield, clusters per vine, berry weight, berries per cluster, pruning weights and the yield to pruning weight ratio (Table 29).

AREC 2: Treatments did not significantly affect any component of yield, pruning weights or the yield to pruning weight ratio (Table 31 and 32). Year had a significant effect upon clusters per vine, pruning weights and the yield to pruning weight ratio ($p < 0.05$) (Table 31).

ISV: Treatments had no significant effect upon any component of yield, pruning weights or the yield to pruning weight ratio (Table 33 and 34). Treatment had no effect upon pruning weights or the yield to pruning weight ratio (Table 35). Year had a significant effect upon total vine yield, clusters per vine, cluster weight, berry weight and berries per cluster ($p < 0.05$) (Table 33).

Table 27. GMV: Components of yield and pruning weights in response to soil and foliar N fertilization (2014-2015)

Treatment ^{ab}	Yield (kg/vine)	Clusters per vine	Cluster weight (g)	Berry weight (g)	Berries per cluster	Pruning weights (kg/vine)	Y:P ^e
Control	11.19 c	61.19	183.39 c	1.56 c	117.36 b	0.98 c	11.58 a
30 N Soil	13.76 b	64.44	214.43 b	1.65 b	130.14 a	1.34 b	10.53 ab
60 N soil	14.96 a	64.46	233.25 a	1.71 a	136.29 a	1.75 a	8.77 b
30 N foliar	12.80 b	62.97	206.44 b	1.62 b	127.26 a	1.23 bc	10.98 a
Trt ^c	<0.0001	ns	<0.0001	<0.0001	<0.0001	<0.0001	0.0034
Yr	<0.0001	<0.0001	ns	ns	ns	0.0047	ns
Trt × Yr	ns ^d	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

^eY:P = Yield to pruning weight ratio

Table 28. GMV: Components of yield and pruning weights in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Yield (kg/vine)		Clusters per vine		Cluster weight (g)		Berry weight (g)		Berries per cluster		Pruning weights (kg/vine)		Y:P ^e	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	10.33 b	12.06 c	55.5	66.89	186.43 b	180.35 c	1.55 b	1.58 c	120.41 b	114.31 b	0.90 c	1.06 b	12.71 a	11.54
30 N soil	12.69 a	14.84 ab	57.39	71.5	221.27 a	207.60 b	1.65 a	1.64 b	133.87 a	126.41 ab	1.26 ab	1.49 ab	10.35 ab	10.16
60 N soil	13.34 a	16.58 a	57.97	70.95	230.51 a	235.99 a	1.69 a	1.73 a	136.18 a	136.41 a	1.55 a	1.95 a	8.99 b	8.75
30 N foliar	11.61 ab	14.09 b	55.07	69.56	211.47 ab	202.30 bc	1.66 a	1.59 c	127.69 ab	127.65 ab	1.06 bc	1.37 ab	13.07 a	10.81
Trt ^c	0.0007	<0.0001	ns ^d	ns	0.0004	0.0001	0.0017	<0.0001	0.0167	0.0050	0.0004	0.0053	0.0258	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

^eY:P = Yield to pruning weight ratio

Table 29. AREC 1: Components of yield and pruning weights in response to soil and foliar N fertilization (2014 -2015)

Treatment ^{ab}	Yield (kg/vine)	Clusters per vine	Cluster weight (g)	Berry weight (g)	Berries per cluster	Pruning weights (kg/vine)	Y:P ^e
Control	3.42	31.53	106.68	1.17 a	93.04	1.09	3.29
30 N soil	3.72	31.51	117.27	1.15 ab	102.20	1.17	3.17
45 N soil	3.78	33.29	113.10	1.13 ab	100.57	1.11	3.63
60 N soil	3.24	30.90	104.34	1.18 a	90.24	1.09	3.11
45 N soil + 15 N foliar	3.59	33.44	108.00	1.08 b	100.63	1.09	3.49
Trt ^c	ns ^d	ns	ns	0.0084	ns	ns	ns
Yr	0.0041	0.0019	ns	<0.0001	<0.0001	0.0062	<0.0001
Trt × Yr	ns	ns	ns	0.0257	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

^eY:P = Yield to pruning weight ratio

Table 30. AREC 1: Components of yield and pruning weights in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Yield (kg/vine)		Clusters per vine		Cluster weight (g)		Berry weight (g)		Berries per cluster		Pruning weights (kg/vine)		Y:P ^e	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	4.00	2.85	34.65	28.40	114.06	99.30	1.05	1.28 ab	108.33	77.76 b	1.07	1.12	3.82	2.76
30 N soil	3.95	3.49	32.58	30.43	118.69	115.85	1.08	1.22 ab	109.51	94.89 a	1.11	1.23	3.46	2.87
45 N soil	4.10	3.45	35.78	30.78	113.85	112.35	1.05	1.22 ab	108.75	92.40 ab	1.00	1.20	4.26	3.01
60 N soil	3.31	3.19	31.28	30.52	104.35	104.32	1.04	1.33 a	101.34	79.15 b	1.02	1.15	3.25	2.98
45 N soil + 15 N foliar	3.68	3.50	34.52	32.36	107.33	108.67	1.02	1.14 b	105.34	95.93 a	1.01	1.16	3.89	3.09
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	0.0136	ns	0.0016	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

^eY:P = Yield to pruning weight ratio

Table 31. AREC 2: Components of yield and pruning weights in response to foliar N fertilization (2014 -2015)

Treatment ^{ab}	Yield (kg/vine)	Clusters per vine	Cluster weight (g)	Berry weight (g)	Berries per cluster	Pruning weights (kg/vine)	Y:P ^e
Control	2.55	34.35	74.60	1.25	59.78	0.68	3.90
15 N foliar	2.56	33.66	77.19	1.24	62.53	0.75	3.53
15 N foliar + 5 S foliar	2.48	35.02	70.32	1.23	57.09	0.68	3.85
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns
Yr	ns	0.0070	ns	ns	ns	<0.0001	<0.0001
Trt × Yr	ns	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

^eY:P = Yield to pruning weight ratio

Table 32. AREC 2: Components of yield and pruning weights in response to foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Yield (kg/vine)		Clusters per vine		Cluster weight (g)		Berry weight (g)		Berries per cluster		Pruning weights (kg/vine)		Y:P ^e	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	2.78	2.31	37.1	31.6	76.09	73.11	1.23	1.27	61.81	57.75	0.63	0.73	4.53	3.26
15 N foliar	2.72	2.41	35.84	31.48	77.02	77.35	1.23	1.24	62.74	62.33	0.68	0.82	4.08	2.99
15 N foliar + 5 S foliar	2.5	2.45	37.28	32.76	66.28	74.37	1.2	1.26	55.03	59.15	0.61	0.75	4.28	3.41
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

^eY:P = Yield to pruning weight ratio

Table 33. ISV: Components of yield in response to treatments (2014 -2015)

Treatment^{ab}	Yield (kg/vine)	clusters per vine	cluster weight (g)	berry weight (g)	berries per cluster
15 N soil	10.55	52.17	207.40	1.96	106.53
15 N soil + 10 N foliar	10.65	53.09	203.49	1.96	103.79
Crimson	10.81	56.88	196.56	1.93	102.24
Crimson + 10 N foliar	10.42	51.35	207.91	1.95	106.93
White	10.90	57.34	195.74	1.94	100.92
White + 10 N foliar	11.67	57.66	211.45	1.98	107.65
Trt ^c	ns ^d	ns	ns	ns	ns
Yr	0.0016	<0.0001	0.0093	<0.0001	<0.0001
Trt × Yr	ns	ns	ns	ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 34. ISV: Components of yield in response to treatments by year (2014 and 2015)

Treatment ^{ab}	Yield (kg/vine)		Clusters per vine		Cluster weight (g)		Berry weight (g)		Berries per cluster	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
15 N soil	11.08	9.86	57.81	44.64	195.11	223.78	2.04	1.86	95.86	120.75
15 N soil + 10 N foliar	12.20	9.10	60.19	46.00	208.18	198.80	2.05	1.86	101.48	106.10
Crimson	11.51	10.11	63.00	50.75	187.49	205.63	2.05	1.82	91.63	112.86
Crimson + 10 N foliar	11.07	9.54	57.81	42.72	195.83	224.03	2.01	1.88	97.57	119.40
White	11.70	10.11	63.88	50.81	190.55	200.93	1.98	1.91	96.36	105.49
White + 10 N foliar	12.69	10.66	68.38	46.94	192.03	230.88	2.06	1.89	93.22	122.07
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	ns	ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 35. ISV: Pruning weights in response to treatments (2015)

Treatment^{ab}	Pruning weights (kg/vine)	Y:P^e
15 N soil	1.08	10.73
15 N soil + 10 N foliar	1.09	8.78
Crimson	0.97	11.53
Crimson + 10 N foliar	0.94	10.92
White	0.90	11.71
White + 10 N foliar	1.00	11.47
Trt ^c	ns ^d	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

^eY:P = Yield to pruning weight ratio

Canopy architecture

Treatment had no effect upon any metric of canopy architecture at any site, except ISV (Table 36 – 43). At ISV, significantly more ambient light was able to reach the fruit zone (%PPF) in the Crimson + 10 N foliar treatment than the 15 N soil + 10 N foliar treatment, ($p < 0.05$) (Table 42). However, the significance of the relationship between treatment and %PPF was not found within each year (Table 43). The Crimson + 10 N foliar treatment had a significantly higher leaf exposure layer number (LEL) in 2015 in relation to the 15 N soil treatment. Year had a significant effect upon cluster exposure layer (CEL) at GMV and AREC 1 ($p < 0.05$) (Table 36 and 38). Year had a significant effect upon occlusion layer number (OLN) at GMV and AREC 2 ($p < 0.05$) (Table 36 and 40). Cluster exposure flux layer (CEFA) and leaf exposure flux availability (LEFA) were significantly affected by year at GMV, AREC 1 and AREC 2 ($p < 0.05$) (Table 36, 38 and 40). Year had a significant effect upon photosynthetic photon flux at all four experimental sites ($p < 0.05$) (Table 36, 38, 40 and 42).

Table 36. GMV: Select EPQA metrics in response to soil and foliar N fertilization (2014-2015)

Treatment ^{ab}	Occlusion layer number (OLN)	Cluster exposure layer (CEL)	Leaf exposure layer (LEL)	Cluster exposure flux availability (CEFA)	Leaf exposure flux availability (LEFA)	Photosynthetic photon flux (%PPF)
Control	1.71	0.21	0.11	0.55	0.55	4.58%
30 N soil	1.88	0.26	0.14	0.49	0.51	3.74%
60 N soil	1.81	0.30	0.14	0.47	0.53	3.74%
30 N foliar	1.83	0.27	0.12	0.52	0.54	4.38%
Trt ^c	ns ^d	ns	ns	ns	ns	ns
Yr	0.0002	0.0048	ns	0.0005	0.0103	<0.0001
Trt × Yr	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 37. GMV: Select EPQA metrics in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Occlusion layer number (OLN)		Cluster exposure layer (CEL)		Leaf exposure layer (LEL)		Cluster exposure flux availability (CEFA)		Leaf exposure flux availability (LEFA)		Photosynthetic photon flux (%PPF)	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	1.62	1.77	0.21	0.23	0.11	0.12	0.61	0.48	0.57	0.53	6.04%	3.05%
30 N soil	1.72	2.03	0.20	0.29	0.13	0.15	0.56	0.44	0.52	0.50	4.89%	2.73%
60 N soil	1.60	2.01	0.16	0.44	0.13	0.16	0.61	0.38	0.56	0.50	4.87%	2.53%
30 N foliar	1.74	1.93	0.23	0.33	0.13	0.11	0.55	0.43	0.56	0.52	5.90%	2.88%
Trt ^c	ns ^d	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = application of 30 kg N/ha as calcium nitrate at bloom and véraison; 30 N foliar = six applications of 5 kg N/ha as urea starting at bloom and separated by 7-10 days

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 38. AREC 1: Select EPQA metrics in response to soil and foliar N fertilization (2014-2015)

Treatment^{ab}	Occlusion layer number (OLN)	Cluster exposure layer (CEL)	Leaf exposure layer (LEL)	Cluster exposure flux availability (CEFA)	Leaf exposure flux availability (LEFA)	Photosynthetic photon flux (%PPF)
Control	2.43	0.50	0.18	0.37	0.49	15.64%
30 N soil	2.38	0.43	0.17	0.39	0.50	14.28%
45 N soil	2.30	0.41	0.17	0.38	0.49	12.33%
60 N soil	2.33	0.45	0.17	0.40	0.51	14.76%
45 N soil + 15 foliar	2.25	0.42	0.17	0.41	0.50	13.61%
Trt ^c	ns ^d	ns	ns	ns	ns	ns
Yr	ns	0.0004	ns	0.0065	<0.0001	<0.0001
Trt × Yr	ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 39. AREC 1: Select EPQA metrics in response to soil and foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Occlusion layer number (OLN)		Cluster exposure layer (CEL)		Leaf exposure layer (LEL)		Cluster exposure flux availability (CEFA)		Leaf exposure flux availability (LEFA)		Photosynthetic photon flux (%PPF)	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	2.46	2.40	0.50	0.50 a	0.19	0.17	0.43	0.31	0.53	0.45	26.52%	2.05%
30 N soil	2.48	2.28	0.57	0.29 b	0.16	0.18	0.38	0.39	0.54	0.46	24.13%	1.96%
45 N soil	2.47	2.12	0.47	0.34 b	0.21	0.13	0.40	0.36	0.49	0.48	18.58%	1.91%
60 N soil	2.37	2.30	0.50	0.40 ab	0.18	0.16	0.45	0.34	0.55	0.47	27.26%	2.25%
45 N soil + 15 N foliar	2.28	2.22	0.49	0.34 b	0.16	0.18	0.43	0.40	0.55	0.45	25.18%	2.04%
Trt ^{c+}	ns ^d	ns	ns	0.0060	ns	ns	Ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 30 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom; 45 N soil = 45 kg N/ha applied to soil as calcium nitrate at bloom; 60 N soil = 30 kg N/ha applied to soil as calcium nitrate at bloom and véraison; 45 N soil + 15 N foliar = 45 kg N/ha applied to soil as calcium nitrate at bloom and two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 40. AREC 2: Select EPQA metrics in response to foliar N fertilization (2014-2015)

Treatment ^{ab}	Occlusion layer number (OLN)	Cluster exposure layer (CEL)	Leaf exposure layer (LEL)	Cluster exposure flux availability (CEFA)	Leaf exposure flux availability (LEFA)	Photosynthetic photon flux (%PPF)
Control	2.5	0.4	0.22	0.38	0.45	10.77%
15 N foliar	2.5	0.5	0.21	0.31	0.44	7.93%
15 N foliar + 5 S foliar	2.68	0.56	0.23	0.3	0.45	10.19%
Trt ^c	ns ^d	ns	ns	Ns	ns	ns
Yr	0.0192	ns	ns	0.0038	0.0006	<0.0001
Trt × Yr	ns	ns	0.0412	Ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 41. AREC 2: Select EPQA metrics in response to foliar N fertilization by year (2014 and 2015)

Treatment ^{ab}	Occlusion layer number (OLN)		Cluster exposure layer (CEL)		Leaf exposure layer (LEL)		Cluster exposure flux availability (CEFA)		Leaf exposure flux availability (LEFA)		Photosynthetic photon flux (%PPF)	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
Control	2.46	2.56	0.40	0.40	0.24	0.20	0.44	0.32	0.48	0.44	18.21%	3.32%
15 N foliar	2.44	2.57	0.50	0.51	0.24	0.20	0.34	0.29	0.46	0.44	13.02%	2.84%
15 N foliar + 5 S foliar	2.42	2.95	0.53	0.60	0.19	0.28	0.36	0.25	0.50	0.40	17.32%	3.06%
Trt ^c	ns ^d	ns	ns	ns	ns	ns	Ns	ns	ns	ns	ns	ns

^aControl = no N fertilization; 15 N foliar = two 7.5 kg N/ha applications of urea separated by 7-10 days at véraison; 15 N foliar + 5 S foliar = two 7.5 kg N/ha applications of urea and 2.5 kg S/ha as micronized sulfur separated by 7-10 days at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Table 42. ISV: Select EPQA metrics in response to treatments (2014-2015)

Treatment^{ab}	Occlusion layer number (OLN)	Cluster exposure layer (CEL)	Leaf exposure layer (LEL)	Cluster exposure flux availability (CEFA)	Leaf exposure flux availability (LEFA)	Photosynthetic photon flux (%PPF)
15 N soil	2.27	0.45	0.16	0.35	0.48	2.40% ab
15 N soil + 10 N foliar	2.32	0.51	0.16	0.31	0.47	1.90% b
Crimson	2.61	0.49	0.25	0.31	0.41	2.18% ab
Crimson + 10 N foliar	2.27	0.53	0.24	0.28	0.43	2.65% a
White	2.34	0.42	0.18	0.34	0.47	2.51% ab
White + 10 N foliar	2.41	0.45	0.20	0.33	0.44	2.20% ab
Trt ^c	ns ^d	ns	ns	Ns	ns	0.0486
Yr	ns	ns	ns	Ns	ns	<0.0001
Trt × Yr	ns	ns	ns	Ns	ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using two-way ANOVA. Trt = significance of treatment effects; Yr = significance of year effects; Trt × Yr = significance of treatment by year interaction

^dns = not significant

Table 43. ISV: Select EPQA metrics in response to treatments by year (2014 and 2015)

Treatment ^{ab}	Occlusion layer number (OLN)		Cluster exposure layer (CEL)		Leaf exposure layer (LEL)		Cluster exposure flux availability (CEFA)		Leaf exposure flux availability (LEFA)		Photosynthetic photon flux (%PPF)	
	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015	2014	2015
15 N soil	2.36	2.19	0.43	0.47	0.18	0.15 b	0.34	0.37	0.46	0.51	2.74%	2.06%
15 N soil + 10 N foliar	2.36	2.29	0.51	0.51	0.18	0.15 ab	0.32	0.30	0.46	0.48	2.14%	1.67%
Crimson	2.51	2.72	0.44	0.54	0.25	0.25 ab	0.36	0.27	0.42	0.42	2.50%	1.86%
Crimson + 10 N foliar	2.20	2.36	0.56	0.51	0.18	0.30 a	0.28	0.29	0.48	0.40	3.32%	1.97%
White	2.15	2.53	0.42	0.42	0.13	0.25 ab	0.35	0.35	0.51	0.43	2.75%	2.27%
White + 10 N foliar	2.37	2.45	0.46	0.45	0.19	0.21 ab	0.33	0.34	0.45	0.44	2.56%	1.84%
Trt ^c	ns ^d	ns	ns	ns	ns	0.0307	Ns	ns	ns	ns	ns	ns

^a15 N soil = 15 kg N/ha applied to soil as calcium nitrate at bloom; 15 N soil + 10 N foliar = 15 kg N/ha applied to soil as calcium nitrate at bloom and two applications of 5 kg N/ha applied to foliage as urea at véraison; Crimson = under vine Crimson clover cover crop; Crimson + 10 N foliar = under vine Crimson clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison; White = under vine White clover cover crop; White + 10 N foliar = under vine White clover cover crop and two applications of 5 kg N/ha applied to foliage as urea at véraison

^bWithin columns, means with different letters indicate differences of means using Tukey's HSD ($\alpha=0.05$)

^cSignificance of effects using one-way ANOVA. Trt = significance of treatment effects

^dns = not significant

Cover crop stand density and biomass

White clover produced significantly more above ground biomass and a denser stand than Crimson clover in 2015 ($p < 0.05$) (Table 44). However, in both the White clover and Crimson clover plots at ISV, the majority of the biomass and plant species within the stand came from non-leguminous weeds.

Table 44. ISV: Average cover crop stand density and proportion of aboveground biomass from Crimson and White clover cover crops in 2015

Cover crop ^{ab}	Cover crop biomass (% of total)	Stand density rating ^c
White clover	40.38% a	3.08 a
Crimson clover	17.88% b	1.50 b
Trt ^d	0.0298	<0.0001

^aWhite clover and Crimson clover cover crops (both alone and with foliar N)

^bWithin column, means with different letters indicate differences in means using Student's T-test ($\alpha = 0.05$)

^cStand density rating 1 = 76-100% invasive species/bare ground; 2 = 51-75% invasive species/bare ground; 3 = 26-50% invasive species or bare ground; 4 = 10-25% invasive species/bare ground; 5 = <10% invasive species/bare ground; 6 = 100% ground cover by cover crop

^dSignificance of treatment effect using one-way ANOVA. Trt = Significance of treatment effect

Discussion

Excessive vegetative growth is a common characteristic of vineyards in the Eastern United States. Therefore, many viticulturists have opted to employ intensive cover cropping strategies in their vineyards, as cover crops have been demonstrated to effectively reduce vine vigor (Giese et al. 2014; Morlat and Jacquet 2003; Tesic et al. 2007; Wheeler et al. 2005). The vigor suppression of a cover crop is due to competition with the vine for both water and nutrients (Monteiro and Lopes 2007; Sweet and Schreiner 2010). The predominant soil N source for grapevines is in the form of nitrate (NO_3^-) (Keller et al. 2001b). Nitrates are dissolved in soil solution and reach the root via mass flow (Barber et al. 1963). Competition between grapevines and companion cover crops for nitrogen has been observed by others (Celette et al. 2009; Giese et al. 2014; Pérez-Álvarez et al. 2015; Sweet and Schreiner 2010; Tesic et al. 2007; Wheeler et al. 2005). Under perennial and complete floor cover cropping scenarios, vineyards can be subject to vine tissue and juice N deficiencies. The current study sought to evaluate several nitrogen management schemes with the aim of increasing vine tissue and juice N, while maintaining the agronomic benefits of the cover crop.

An increase in vine tissue N status in response to soil-applied N has been previously demonstrated (Bell and Robson 1999; Bell and Francis 2013; Conradie and Saayman 1989b; Ekbic et al. 2010; Keller et al. 2001b; Linsenmeier et al. 2008; Peuke 2009). However, the response of petiole N concentration to soil-applied N can be unreliable, resulting in only a marginal increase in tissue N concentration or an insignificant effect (Conradie and Saayman 1989b; Ekbic et al. 2010). While both soil- and foliar-applied N were evaluated in the current study, soil-applied nitrogen was more successful at raising petiole N status at véraison. In general, increasing the rate of N fertilization increased petiolar N concentration. However, the 45 N soil treatment at AREC 1 resulted in the highest petiole N concentration, not the 60 N soil treatment. The 60 N soil treatment was split into two equal applications of 30 kg N/ha, with the first being applied at bloom and the second application just prior to

véraison, whereas the 45 N soil treatment at AREC 1 was completely imposed at bloom. As N uptake is greatest at bloom, the 45 N soil treatment may have been more efficiently assimilated than the 60 N soil treatment, as more N was applied to the soil at the period of maximum N uptake (Schreiner et al. 2006). Petiole N concentration was not significantly impacted by the treatments at ISV. This strongly suggests that the application of 15 kg N/ha to the soil was insufficient for increasing vine N status, even in plots with a bare soil surface directly underneath the trellis.

A véraison petiolar N concentration of between 0.8-1.2% has been recommended as the optimum range for grapevines in the Eastern United States (Wolf 2008). No treatment in the current study was able to reach this “optimal” concentration. Perhaps this warrants further investigation as to the validity such a high “optimal” véraison petiolar N range. It should be noted that due to the great temporal variability of petiolar N status, leaf blade or whole leaf samples may be the most effective means of chemically assessing vine tissue N status (Davenport and Horneck 2011; Dominguez et al. 2015; Romero et al. 2014).

The clover cover crops at ISV did not affect vine N status. However, petiolar N status was maintained relative to the 15 N soil treatment. A similar maintenance of vine N status has been found by Pérez-Álvarez et al. (2015). The maintenance of vine N may have been due to rhizodeposition of nitrogen by the clover cover crops (Fustec et al. 2011). The potential N maintenance could be explored in the future by comparisons of white clover with non-leguminous cover crops in the same experiment.

The application of foliar urea alone was ineffective at significantly increasing petiole N concentrations, as has been reported by others (Hannam et al. 2014). Between 17 and 80% of N applied in the form of foliar urea can be assimilated by the berries (Lasa et al. 2012; Schreiber et al. 2002). Also, the foliar urea treatments at ISV, AREC 1 and 2 were imposed around véraison, which corresponds to the phenological stage at which point the grape cluster becomes the strongest N sink and is highly

competitive with the other organs of the grapevine for N nutrition (Conradie 1991). Therefore, while foliar urea applications may effectively elevate berry YAN, it should not be relied upon to improve vine tissue N and vine capacity when the vineyard is in a state of N deficiency.

As reported by others we too found a significant reduction in vine P concentration in response to N fertilization (Bell and Robson 1999; Bell and Francis 2013; Conradie and Saayman 1989b; D'Attilio 2014; Linsenmeier et al. 2008; Peuke 2009; Spayd et al. 1993). Soil-applied N has been shown to increase petiolar mass (Bell and Robson 1999). The depression of petiolar P concentration in the current study in response to soil-applied N may be due to a dilution caused by greater petiole dry weights in response to N fertilization. Others have suggested that the decreased P concentration in response to soil N fertilization is in response to soil acidification caused by the application of nitrogenous fertilizer (Bell and Robson 1999; Conradie and Saayman 1989b); however, N fertilizer was applied as calcium nitrate in the current study, which is known to increase soil pH over time (Pierre 1928; Riley and Barber 1971; Sarkar and Wynjones 1982), although soil pH was not monitored in the current study.

Although not always statistically significant, petiolar potassium (K) concentrations were diminished in response to soil-applied N at GMV. Diminished K nutrition due to soil-applied N has been found by others (Hilbert et al. 2003; Linsenmeier et al. 2008; Peuke 2009). However, in their long-term study, Conradie and Saayman (1989) found that nitrogenous fertilization did not impact tissue K concentrations. The concentration of K in the petioles at ISV was significantly lower in the White + 10 N foliar treatment in relation to the Crimson + 10 N foliar treatment. However, significance was only found in the first year of the study. Although stand density and above ground biomass was only measured in 2015, it was anecdotally observed that the cover crops behaved similarly in both years. The White clover established a much denser stand with a greater biomass than that of the Crimson clover treatments. Sweet and Schreiner (2010) found that clover/grass mixes can effectively compete with the vine for K. As there was quite a lot of bare ground within the plots sown with Crimson clover, which was

associated with their poor stand rating, the White clover plots may have been more competitive for K. Tesic et al. (2007) found that cover crops were able to effectively compete with the vine for K; however, this response was inconsistent from year to year, as we also observed at ISV.

Keller et al. (2001) found that N fertilization increased the translocation of Mg in the xylem. Others have also noticed an increase in tissue Mg in response to N fertilization (Bell and Francis 2013; Hilbert et al. 2003; Linsenmeier et al. 2008). These results are in agreement with the current study, in which increased Mg concentrations were found in the véraison petiole samples at GMV and AREC 1.

Calcium was mostly unaffected by fertilization treatments. However, the application of foliar urea resulted in a statistically significant increase in petiolar Ca concentration between the 45 N soil and 45 N soil + 15 N foliar treatments at AREC 1. However, when each year was evaluated individually, it was apparent that these results were not significant between treatments, but the similar trend did exist, with the highest Ca concentration coming from the 45 N soil + 15 N foliar treatments. This result is of little or no consequence, as foliar N had no effect upon petiole Ca in the same variety at AREC 2, nor with Sauvignon blanc at GMV.

The only apparent treatment effects on micronutrients were a slight depression of Mn and B in the case of soil-applied N. Upon evaluating each year individually, it seems that the Mn was not significantly affected by treatment within each year. However, boron was depressed by soil N in 2015. This result is in agreement with Peuke (2009), who found that boron concentration in leaves of vines fertilized with N decreased with increasing levels of N fertilization. Nitrate is taken up by roots through active proton cotransport via a membrane bound proton pump, H⁺-ATPase (Glass et al. 1992; Siddiqi et al. 1990). Boron is essential to the activity of the H⁺-ATPase pump (Camacho-Cristóbal and González-Fontes 2007; Ferrol et al. 1993). With increasing levels of NO₃⁻ fertilization and the subsequent increase in H⁺-ATPase activity, it may deplete plant boron concentrations. Like petiolar P, the response of manganese and boron to soil N application may also be a function of dilution due to increased dry

matter accumulation. Also, the depression of boron concentration in petioles at véraison in response to soil-applied N is not viticulturally significant in that B tissue values were still well within an acceptable range (Wolf, 2008).

Soil-applied N significantly increased season-long CCI values at GMV and AREC 1, similar to petiolar N, CCI readings were highest in the 60 N soil treatment at GMV and the 45 N soil treatment at AREC 1. Leaf chlorophyll content and CCI values have been found to be highly correlated in species as diverse as grapevines, sugar maple, birch and Asian pear (Cate and Perkins 2003; Filimon et al. 2014; Ghasemi et al. 2011; Richardson et al. 2002; Taskos et al. 2015). The relationship between N fertilization, petiolar N and CCI is not surprising, as leaf chlorophyll content has been found to increase with increasing concentrations of N (Bell and Francis 2013; Ghasemi et al. 2011; Keller 2005; Keller and Koblet 1995; Keller et al. 2001b; Taskos et al. 2015). A significant positive relationship existed between season-long CCI and petiole N% at véraison at AREC 1 and ISV. However, this significant relationship was not apparent at GMV. This may be due to the methodology by which leaf N was measured. In previous studies which evaluated the efficacy of leaf N prediction with the CCM-200, total leaf N% was measured, rather than just petiolar N (Ghasemi et al. 2011; Taskos et al. 2015). This difference in methodology may impact the strength of this correlation. The lack of a positive relationship between leaf petiolar N and CCI at GMV was also observed by D'Attilio (2014). Petiolar N is more variable than leaf blade N (Dominguez et al. 2015). Also, leaf blades have been found to contain nearly triple the concentration of N as petioles, which may improve the sensitivity of the analysis for N (Romero et al. 2010). However, Dominguez et al. (2015) also found that the stability period for N was longer during véraison within the petioles and Romero et al. (2010) found that the position of the leaf did not affect petiolar N concentrations as much as it did the leaf blade. Therefore, either methodology (leaf blade or petiole sampling) has sources of error which cannot be overcome. The use of whole leaf sampling, as a means of reducing these sources of error and variability, warrants further investigation. Treatment did not

have a significant effect upon season-long CCI at ISV in 2015. This is not surprising, as treatments also did not have a significant effect upon petiole N at ISV.

Yield and pruning weights followed a similar trend to CCI and petiolar N% at GMV and AREC 1, with higher yields and pruning weights associated with increasing levels of soil and foliar-applied N. It's interesting to note that although the soil-applied N had the greatest effect upon yield, the 30 N foliar treatment at GMV also significantly increased yield. Hannam et al. (2016) also found foliar urea increased vine yields in their study on Pinot gris and Merlot. In the current study, GMV was in the fourth and fifth year of treatments and a statistically significant yield differential was not found in previous years (D'Attilio 2014). In the previous study conducted by D'Attilio (2014), yield increased with soil-applied N treatments, but these yield increases were not statistically significant. A number of studies have also found an increase in fruit yield in response to soil-applied N (Conradie and Saayman 1989a; Keller et al. 1998; Keller et al. 2001a; Linsenmeier et al. 2008; Spayd et al. 1993). Conradie and Saayman (1989b) also found a delayed yield response with N fertilization treatments, with a N fertilization treatment increasing yield only after three years. The component of yield most affected by N fertilization at GMV was cluster weight. The increase in cluster weight with N fertilization was due to both more berries per cluster as well as greater individual berry weights. An increase of berry weight and the number of berries per cluster in response to increased N supply has also been demonstrated by others (Amiri and Fallahi 2007; Cheng et al. 2004; Duchene et al. 2001; Keller et al. 1998; Keller et al. 2001a; Spayd et al. 1993). Increased leaf chlorophyll and N content has been correlated with greater CO₂ assimilation (Chen and Cheng 2003b; Keller 2005; Keller et al. 2001b). Lower assimilation rates have been associated with limited berry growth (Dokoozlian and Kliewer 1996; Finger et al. 2002; Ollat and Gaudillere 1998). As N fertilization increased leaf chlorophyll content in N fertilized treatments, CO₂ assimilation may have also increased, thereby increasing berry size. The increase in fruit weight in response to N fertilization was likely due to increased cellular division and/or enlargement in the

pericarp brought on by increased assimilate supply, as has been found in cucumber, tomato and apple (Bohner and Bangerth 1988; Marcelis 1993; Xia et al. 2009).

It is commonly understood that limited N in the previous year can affect the vegetative and reproductive growth of the subsequent year, including bloom and fruit set (Cheng et al. 2004; Duchene et al. 2001; Guilpart et al. 2014; Zapata et al. 2004). Cheng and Xia (2004) found that berry size and the number of berries per cluster were affected by both reserve and current season N supply. As the Sauvignon blanc at GMV was in a state of N deficiency at the beginning of this long-term experiment in 2011, N reserves were likely increased over time. This may explain why neither berry number per cluster nor berry weight were significantly impacted until year 2014 and 2015. Berry number per cluster may have increased due to improved fruit set/ovule fertility brought on by improved N nutritional status (Ewart and Kliewer 1977). It is also possible that the improved N status of the grapevine increased the branching of the anlage in the previous season, which resulted in more flowers per inflorescence, as was speculated by others (Guilpart et al. 2014). Keller et al. (2001a) found that N fertilization did not increase the number of flowers per inflorescence, however the researchers only evaluated the effect of N fertilization in one year. Carbohydrate (CHO) reserves of the grapevine also play a crucial role in both inflorescence number per shoot and flower number per inflorescence (Bennett et al. 2005). Bennett et al. (2005) found a significant correlation between dormant CHO status of the grapevine and current season pruning weights. Therefore, pruning weights may be a good proxy for vine CHO status. Pruning weights significantly increased with both rates of soil-applied N at GMV. Therefore, it is likely that vine CHO at GMV was increased through N application and the increase in berries per cluster observed with increasing levels of soil N may have been due to improved fruit set and/or more flowers per inflorescence.

Soil-applied N increased vine vegetative growth at a faster rate relative to crop yield, as indicated by the significantly lowered yield to pruning weight ratio at GMV. Spayd et al. (1993) also

reported an increase in the crop weight to pruning weight ratio in response to N fertilization. A crop weight to pruning weight ratio of 5-10 has been proposed as being indicative of vines which are considered balanced (Kliewer and Dokoozlian 2005; Smart and Robinson 1991). If this ideal crop to pruning weight ratio is to be applied, the soil-applied N treatments at GMV brought the vines into balance. Kliewer and Dokoozlian (2005) found a significant negative correlation between the leaf area to crop weight to pruning weight ratio, it stands to reason that soil-applied N increased leaf area. Bell and Robson (1999) found that total vine leaf area, leaf area per leaf and the number of leaves per vine increased with increasing levels of nitrogen fertilization.

Canopy density was not affected by treatment at GMV, AREC 1 or at AREC 2. EPQA measurements were taken at véraison. It should be noted that these vineyards were commercially managed and that standard canopy management practices were undertaken prior to véraison (i.e. leaf removal, shoot positioning and hedging). These practices would have mitigated any potential increase in vine vigor caused by N fertilization. Others have found an increase in N supply can increase canopy density (Bell and Robson 1999; Bell and Francis 2013). Increasing N supply has been found to increase total vine leaf area, area per leaf and the number of leaves per vine (Bell and Robson 1999; Cheng et al. 2004; Keller et al. 1998). Others have found that the effect of N fertilization upon canopy density was inconsistent from year to year (Hannam et al. 2013). Bell and Francis (2013) found that although leaf layer number increased with N fertilization, the fruit zone light environment was unaltered.

The only significant treatment effect upon the canopy architecture metrics measured in the current study was at ISV. Significantly less light reached the fruit zone of the 15 N soil + 10 N foliar treatment than the Crimson + 10 N foliar treatment. However, this metric was not significant between treatments within each year of the study. Although the combined year model found statistical significance, it is unlikely that the treatment differences were biologically significant.

Infections of the grape cluster by the fungal pathogen *Botrytis cinerea* can lead to devastating crop losses and severe consequences for fruit and wine quality. *Botrytis* can cause juice/wine browning through the rapid enzymatic oxidation of phenolic compounds, diminish the aromatic impact of monoterpenes, and directly impart a mushroom-like aroma (Boidron 1978; La Guerche et al. 2006; Macheix et al. 1991; Miklosy et al. 2004). Others have found that increased N supply led to a greater incidence of *B. cinerea* in grapes (Keller et al. 2001a; R'Houma et al. 1998). In the current study, incidence of *B. cinerea* infection was recorded at each site in 2015 (data not presented) to investigate a potential disease response to N fertilization. Of the 4 sites, only two clusters were harvested with evidence of *B. cinerea* (both at GMV in a 30 N soil and 60 N soil experimental unit). Lacroux et al. (2008) also found that soil and foliar N applications did not significantly affect incidence of *B. cinerea*. Canopy density, weather, inoculum levels and physical damage to the grape berry are all known to affect the severity of *B. cinerea* infections (Broome et al. 1995; English et al. 1993). Keller et al. (2001a) hypothesized that the increased incidence of *B. cinerea* associated with N fertilization was due to increased cluster compactness caused by increased fruit set. Others have also found that more compact clusters are more susceptible to *B. cinerea* infection (Hed et al. 2009; Vail and Marois 1991). Cluster compactness was not measured in the current study; however, N fertilization was found to significantly increase berry weights and the number of berries per cluster at GMV and AREC 1. The increase in berry and berries per cluster may have resulted in an increase in cluster compactness. Therefore, although a significant relationship between N fertilization and *B. cinerea* incidence was not observed in the current study, it is still recommended that growers be aware of the potential risk and manage their vineyard accordingly, should they opt to utilize an N fertilization scheme.

Conclusion

The application of nitrogenous fertilizers to the soil increased vine nitrogen status and leaf chlorophyll content. After four years of fertilization in a N deficient vineyard, soil-applied N resulted in increased yields by increasing vine size and capacity, without negatively impacting canopy architecture. The application of nitrogenous fertilizers to the soil at bloom (~30-60 Kg N/ha) and a small application of urea to the foliage around véraison (~10-15 kg N/ha) has the possibility to both increase vine N status, capacity and juice N while maintaining the agronomic benefits of the cover crop. The use of white clover as a perennial cover crop under the vine may assist in the maintenance vine N status, but this hypothesis was not confirmed over the course of this study and warrants further investigation. Future studies could include an evaluation of vineyard N fertilization schemes upon varietal aromas and their precursors. Foliar-applied N provides the researcher with an opportunity to decouple the relationship between methoxypyrazines, canopy density and N fertilization, as foliar-applied N appeared to have no impact upon canopy density.

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