

# Influence of Ambient Ozone on Grape Cultivars ‘Chambourcin’ and ‘Vidal’ in Pennsylvania, USA

## Research Article

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### Abstract

The French-American hybrid wine-grape cultivars ‘Chambourcin’ and ‘Vidal’ are commonly grown in Pennsylvania USA and are sensitive and tolerant, respectively, to the air pollutant ozone. In 2003, the two grape cultivars were planted within an agricultural field in south-central Pennsylvania. Open-top chambers were placed over the grapevines in June 2004. During 2004-2008, grapevines were exposed to either non-filtered air containing ambient ozone concentrations or to charcoal-filtered air that contained 63% of ambient ozone, annually from the beginning of June through the end of September. Ozone exposures and monitoring were conducted 24 h/day. Annual ozone-induced injury to grape leaf tissue was recorded in mid-September; annual berry and juice data were determined at harvest. The ozone-susceptible cultivar Chambourcin exposed to ambient ozone averaged 5.28% leaf tissue injury over the five growing seasons, whereas Chambourcin foliage in the charcoal-filtered treatment exhibited a mean of 0.48% injury during the same time period. In contrast, foliage of the ozone-tolerant Vidal cultivar exposed to either ozone treatment averaged only 0.02% means tissue injury over the five seasons. For both cultivars, berry harvests at the end of each growing season revealed that mean berry weight, juice pH, total acidity, and total sugars significantly differed among years but not in response to ambient ozone. These results should be useful in the development of future government legislation on standards to protect public health and public welfare, including agricultural ecosystems. Also since tropospheric ozone is a component of climate change, its influence on wine berry quality could impact grape cultivar effectiveness in specific sites and/or future vineyard locations.

**Keywords:** Air pollution; Ozone; Grapes; Grape-berries

## Introduction

### Pennsylvania grape industry

Pennsylvania (PA) is the fifth largest grape producer in the U.S. behind California (CA), Washington (WA), New York (NY), and Oregon (OR) [1]. PA has 350 commercial vineyards on 5,666 ha that annually produce more than 57,700 metric tonnes of grapes. Although the grape industry in PA is dominated by grape cultivars grown for juice, (*i.e.*, ‘Concord’), the state also has a diversity of soils and climates that are ideal for growing grapes for wine production

and has approximately 728 ha of wine-grapes and more than 160 wineries.

Two of the more popular wine-grapes (*Vitis vinifera*) in PA are the American-French hybrid (French-hybrid) cultivars ‘Chambourcin’ and ‘Vidal’. Chambourcin is widely planted in the mid-Atlantic region of the U.S., especially in New Jersey, NY, and PA. This high-yielding cultivar is productive and offers a fine taste and pleasant aromas related to the berry’s thick skin, high tannins, and acidity. Vidal is also a widely planted in PA as well as in Canada, where this

cultivar is regarded as a producer of excellent ice-wine. The high, natural acidity of Vidal makes it suitable for a range of wine styles. In addition, Vidal grows well in cold climates such as northern PA, where it can produce good crops even from secondary buds on grape plants injured by late spring frosts.

### Tropospheric ozone in the U.S.

The U.S. Environmental Protection Agency (EPA) has designated tropospheric (from ground level to ~10 km) ozone as one of six criteria air pollutants to be federally regulated in the U.S. to reduce the risk of harmful effects of ozone to people, agricultural crops, native ecosystems, and other resources [2]. Tropospheric ozone is a secondary air pollutant formed during chemical reactions involving the primary air pollutants nitrogen oxides (NO<sub>x</sub>) and hydrocarbons in the presence of sunlight and warm temperatures [3,4]. Ozone is of regional-scale importance in PA, especially during long-range transport across the state within slow-moving, stagnant, high-pressure systems. Significant concentrations of ambient ozone have been recorded across agricultural and forested regions of PA that vary both daily and seasonally [5,6]. Daily ozone variations in the U.S. typically follow diurnal day-night patterns, wherein concentrations are low in early morning when NO<sub>x</sub> is present to react with and consume ozone. Concentrations are greatest in the late afternoon, when ozone production exceeds the continuous destruction via the reverse reaction [4]. Ozone levels are generally minimal at night, but night time concentrations vary with elevation. Seasonally, ambient ozone concentrations downwind from urban areas often peak late in the summer season and may exceed 60 ppb in northeastern U.S. [3], a concentration capable of injuring ozone-sensitive plant species if duration of exposure time is sufficient [7].

### Effect of ozone on grapes

Ambient ozone is responsible for significant economic damage to agricultural crops worldwide, including grapes [3,4]. In 1958, Richards, et al. presented the first detailed description of ozone-induced visible foliar injury on broad leaved plants, using grape as a model [8]. They reported that putative ozone injury on CA table and wine grapes had been observed as early as 1954 from exposure to the “Los Angeles smog,” which contained ozone. Visible symptoms on ozone-injured grape leaves were small spots termed “stipples” on the upper leaf surface. Stipples were comprised of brown-to-black groups of palisade mesophyll cells, which were visible through the adaxial epidermis and overlying cuticle. Wild grapes are also sensitive to ozone, exhibiting similar adaxial stipple symptoms, and have been used as bioindicators of phytotoxic levels of ambient ozone in the U.S. [9-12].

Visible ozone-induced stipple on the foliage of French-hybrid grape cultivars has been observed in NY since 1966 [13]. Some grape vineyards in NY and PA are located downwind from sources of ozone precursors [6]. Kender and Carpenter reported Chambourcin to be among the most sensitive grape cultivars grown in NY and Ontario (ON), Canada [14]. They also stated that Vidal was the most ozone-tolerant of 39 grape selections from NY and ON grape breeding programs. Likewise, we have observed ozone injury on Chambourcin, but not Vidal, within PA grape-growing regions. Further, our

unpublished field observations reveal that Chambourcin grapes may be even more sensitive to ambient ozone in PA than was indicated by early studies in NY.

While ozone effects on grape visual injury have been reported, the quality components of yields are not as well understood for wine grapes as they are for many of the larger acreage [15], more economically important agricultural crops including rice, wheat, and soybeans [16]. In a field study over four growing seasons with ‘Thompson Seedless’ grapes in CA, ambient oxidants (including, but not restricted to ozone) reduced berry set and yield and had greater deleterious effects in years of low yield [17]. Thompson Seedless berry size and percent acid were not affected by exposure to ambient oxidants, but the oxidants slightly reduced berry sugar concentration.

In addition, ozone is a powerful greenhouse gas influencing climate change and tropospheric ozone is projected to increase in many of the world’s most productive agricultural growing regions as a result of climate change [16]. Wine grapes typically have narrow climate ranges for optimum quality [18], so a better understanding of changing climatic variables on wine berry juice quality may be important to understand and better forecast cultivar effectiveness in specific locations in the future [19].

The objectives of this study are to determine the effects of the two ozone treatments on foliar injury and fruit characteristics (berry size and juice quality) of field-grown Chambourcin (ozone-sensitive) and Vidal (ozone-tolerant) French-hybrid grape cultivars during five growing seasons (2004-2008) in south-central PA.

## Materials and Methods

### Study site

This study was conducted within an agricultural field in south-central PA at The Pennsylvania State University Fruit Research and Extension Center (39.934550° N, 77.255259° W) located near the city of Biglerville in Adams County. The region is characterized by cold winters and hot summers, with a winter average low of -5 °C and an average summer high of 23 °C. Total annual precipitation is ca.1100 mm, with approximately 680 mm falling during the growing season of April through October. The research site is 220 m above mean sea level.

Chambourcin and Vidal grapevines were planted during spring 2003 in a fine-loamy soil with 1.5-m plant spacing within rows and 3-m spacing between rows. A mix of tall fescue (*Festuca arundinacea*) and orchard grass (*Dactylis glomerata*) was established between grapevine rows prior to planting. Vines were trained to a bilateral flat-cane system and spur-pruned during dormancy to leave approximately 20 buds/plant. Vines were thinned to 12 shoots/m canopy and selective leaf removals were conducted within the fruiting-zone to achieve a desirable canopy density and fruit exposure to sunlight [20]. Grapevines were managed during the 5 years of ozone treatment using conventional vine practices for the Mid-Atlantic region.

### Ozone treatment design

Ozone exposures began in spring 2004, at which time four open-top chambers were placed over the grapevines in the field (Figure 1)



**Figure 1:** Open-top chambers placed over Chambourcin and Vidal grapevines allowing exposure of vines to ambient and 63% ambient ozone concentrations.

[21]. Each chamber contained eight Chambourcin (ozone-sensitive) and two Vidal (ozone-tolerant) grapevines. The field plot design consisted of two replications of two ozone treatments assigned to each replication. Ozone treatments consisted of 1) a non-filtered air chamber containing ambient concentrations of ozone ("ambient ozone or non-filtered air") and 2) a carbon-filtered air chamber containing reduced concentrations of ambient concentrations of ozone ("charcoal-filtered or filtered" air). Ozone reductions were accomplished by passing ambient air through activated charcoal-filters [22]. Annual ozone treatments were conducted 24 h/day from May 1 until mid-September to early October from 2004-2008. The plastic sides of the chambers were removed in mid-October, leaving plants exposed to the outside environment until the following May, at which time the plastic sides were re-attached to the chambers and ozone treatments resumed for the following growing season. The experiment was terminated after the 2008 berry harvest.

### Ozone concentrations

Ozone concentrations (ppb) were monitored within each open-top chamber for 24 h/day during June, July, August and September of 2004-2008. Measurements were made at 5-minute intervals using calibrated TECO Model-49 ozone analyzers (Thermo Environmental Instrumentals Franklin, MA, USA) connected to a computerized monitoring/exposure system. Mean 12-h/day (0800-2000 hours) ozone concentrations (ppb) from June to September of each year (2004-2008) were calculated from the 24-h monitoring data to estimate the level of ozone within carbon-filtered and non-filtered ozone treatments.

### Percentage of ozone-induced leaf tissue injury

Ozone-induced foliar injury, expressed as percentage of upper leaf surface stippled/plant, was evaluated annually during early September. In 2004 and 2005, the percentage injured leaf tissue in each treatment was rated from 0-100% in 10% increments for all leaves on each plant, and a mean leaf injury rating/plant calculated. To improve rating efficiency during 2006 to 2008, five leaves/plant were tagged, and each tagged leaf rated from 0-100% ozone-induced injury in 10% increments. A mean leaf injury rating/plant was calculated for 2006, 2007, and 2008 based on the average percentage tissue injury on the tagged leaves.

### Berry fruit characteristics

Grape berries were harvested annually from mid-September to early October. Replicate samples from the cumulative harvest of each cultivar within each ozone treatment were taken and weighed (g/100 berries). After weighing, berries were dissected using a razor blade and the pulp separated from the skin and seeds. The pulp was crushed, and the resulting juice analyzed for sugars as Total Soluble Solids (TSS), Titratable Acidity (TA), and pH. TSS was measured using a digital refractometer for food (Hanna Instruments, Smithfield, RI) and expressed as °Brix. TA was determined by neutralization of juice to pH 8.10 with 0.10 N NaOH and expressed as g/l of tartaric acid [23]. Both TA and pH were measured using a Titratable Acidity Mini Titrator (Hanna Instruments, Smithfield, RI).

### Statistical analyses

The experiment utilized a completely randomized design with 2 replications, 5 years (2004-2008), 2 grape cultivars (Chambourcin and Vidal), and 2 ozone treatments: 1) non-filtered air = ambient ozone and 2) filtered air. As described in the Methods, four open-top chambers were used to deliver the two ozone treatments, with each chamber containing eight Chambourcin and two Vidal grapevines. The average leaf tissue injury-rating/plant was analyzed using analysis of variance (ANOVA) with SAS PROC MIXED [24]. Initially the data were analyzed as a 3-way factorial (5 years x 2 cultivars x 2 ozone treatments). However, the 3-way interaction was significant, so data were reanalyzed by cultivar as a 2 x 5 factorial, where plant nested in the chamber was considered a random effect. Grape berry fresh weight and juice data (pH, total acidity, and soluble solids concentration) were also statistically analyzed by ozone treatment and year. Since year x cultivar x treatment interactions were significant, data were reanalyzed as a 2 x 5 factorial by cultivar using ANOVA.

## Results and Discussion

### Ambient ozone concentrations

Mean 12-h/day (0800-2000 hours) ozone concentrations from June to September for all 5 years (2004-2008) in the filtered-air treatment averaged 21.4 ppb, whereas ozone concentrations in the non-filtered treatment averaged 34.0 ppb. These concentrations indicate that grapevines in the filtered-air chambers were exposed to 63% of the non-filtered ambient ozone over the 5 years.

### Foliar ozone injury

The most common ozone-induced foliar symptom was a dark stipple on the upper leaf surface (Figure 2). Other visual symptoms, but not recorded, included late-season yellowing and premature defoliation (accelerated senescence), similar to symptoms previously reported on grape [9]. For the grape cultivar Chambourcin, the ANOVA P-values for year, treatment, and the interaction of year x treatment were significant (Table 1). Leaves exposed to the non-filtered treatment (ambient ozone) exhibited 5.28% tissue injury averaged over 5 years. In contrast, leaves in the filtered-air treatment exhibited a mean of only 0.48% tissue injury over the 5 years. For Vidal, an average of only 0.02% ozone-induced leaf tissue injury was exhibited in both ozone treatments.



**Figure 2:** Severe adaxial leaf surface stipple induced by ambient ozone on cultivar Chambourcin.

Researchers had evaluated ozone injury on 59 American, 40 French-hybrid, and six *V. vinifera* grape cultivars growing in NY vineyards during 1977-1980 [25]. Chambourcin was reported to be among the most ozone-sensitive of the French-hybrid wine-grape cultivars. Others reported that Vidal was the most ozone-tolerant of 39 grape selections from NY and ON grape breeding programs [14]. Our results confirm the above findings with regard to ozone-sensitivity of Chambourcin and ozone-tolerance of Vidal [14,25].

### Berry fruit characteristics

Table 1 illustrates the impact of non-filtered air (ambient ozone) vs filtered air (approx. 63% of ambient ozone) on berry weight, as well as juice pH, juice sugar, and juice total acid, as determined

annually following berry harvest in late September-early October of 2004-2008. Ambient ozone did not significantly affect berry weight or any juice characteristics. It has been reported that ozone reduced chlorophyll and associated photosynthesis, berry fresh weight, berry sugar concentration, and overall yield of 'Zinfandel' grapes in CA [26,27]. Others also reported that berry size and percent acid were not affected by exposure to ambient oxidants, but that oxidants slightly reduced berry sugar concentration [17].

The variable "Year" (but not the "Year x Treatment" interaction) was significantly related to all berry and juice characteristics. The Year variable likely reflected variation in growing season weather parameters (i.e., rainfall and temperature) known to affect grape berry yield and juice parameters. For example, 1.0 °C increase in average daily bud temperature during the 3 weeks before bloom caused significant reductions in fruit set, berry weight, and titratable acids and increases in pH and berry color, but soluble solids were not appreciably affected by temperature [28]. In our study, acidity was high in 2008 and may be related to low recorded temperatures in April, but we have no explanation for year-to-year variation in the other juice parameters. Berry weight and most juice parameters can also be influenced by crop load and number of berries per cluster [29]. While those variables were not recorded in this study, they should be considered for inclusion in future ozone wine grape quality investigations.

### Conclusion

The objective of this study was to evaluate effects of ambient ozone on foliar injury (% leaf tissue stippled) and berry fruit characteristics (size and juice quality) of Chambourcin (ozone-sensitive) and Vidal (ozone-tolerant) French-hybrid grape cultivars. Exposures were

**Table 1:** Influence of two ozone treatments on mean leaf injury, berry fresh weight, juice pH, juice acidity, and juice sugar content for Chambourcin and Vidal grape cultivars. Measurement techniques are given in the Methods section.

| Year             | Ozone treatment <sup>1</sup> | Ozone conc (ppb) | Chambourcin   |                           |          |                            |                          | Vidal         |                           |          |                            |                          |
|------------------|------------------------------|------------------|---------------|---------------------------|----------|----------------------------|--------------------------|---------------|---------------------------|----------|----------------------------|--------------------------|
|                  |                              |                  | % Leaf injury | Berry weight <sup>2</sup> | Juice pH | Juice acidity <sup>3</sup> | Juice sugar <sup>4</sup> | % Leaf injury | Berry weight <sup>2</sup> | Juice pH | Juice acidity <sup>3</sup> | Juice sugar <sup>4</sup> |
| 2004             | Non-Filtered                 | 33.59            | 5.8           | 232.2                     | 3.38     | 8.8                        | 20.1                     | 0.0           | 171.7                     | 3.49     | 6.4                        | 22.6                     |
|                  | Filtered                     | 18.27            | 0.8           | 249.9                     | 3.37     | 8.7                        | 20.7                     | 0.0           | 177.3                     | 3.48     | 7.3                        | 22.6                     |
| 2005             | Non-Filtered                 | 37.25            | 3.1           | 216.2                     | 3.08     | 8.9                        | 21.2                     | 0.0           | 164.6                     | 3.45     | 6.3                        | 23.4                     |
|                  | Filtered                     | 20.26            | 0.3           | 219.9                     | 3.07     | 9.6                        | 20.6                     | 0.0           | 162.6                     | 3.31     | 6.5                        | 22.9                     |
| 2006             | Non-Filtered                 | 36.34            | 10.1          | 230.3                     | 3.29     | 6.4                        | 21.9                     | 0.1           | 158.5                     | 3.38     | 5.3                        | 24.9                     |
|                  | Filtered                     | 19.77            | 0.6           | 216.7                     | 3.28     | 7.0                        | 21.7                     | 0.1           | 168.2                     | 3.26     | 6.3                        | 24.0                     |
| 2007             | Non-Filtered                 | 41.42            | 6.7           | 244.3                     | 3.42     | 7.7                        | 11.6                     | 0.0           | 186.1                     | 3.65     | 5.4                        | 12.5                     |
|                  | Filtered                     | 22.37            | 0.2           | 231.8                     | 3.44     | 7.8                        | 10.8                     | 0.0           | 177.6                     | 3.69     | 5.2                        | 13.0                     |
| 2008             | Non-Filtered                 | 37.36            | 0.7           | 233.4                     | 3.15     | 11.9                       | 19.1                     | 0.0           | 202.4                     | 3.38     | 8.7                        | 22.6                     |
|                  | Filtered                     | 20.32            | 0.5           | 233.6                     | 3.18     | 11.7                       | 19.2                     | 0.0           | 192.8                     | 3.39     | 9.1                        | 19.2                     |
| ANOVA P-values   |                              |                  |               |                           |          |                            |                          |               |                           |          |                            |                          |
| Year             |                              |                  | 0.0190        | 0.0093                    | 0.0001   | < 0.0001                   | < 0.0001                 | 0.5772        | 0.0001                    | < 0.0001 | < 0.0001                   | 0.0002                   |
| Treatment        |                              |                  | < 0.0001      | 0.9282                    | 0.9028   | 0.1084                     | 0.8546                   | 0.3524        | 0.8869                    | 0.4474   | 0.5939                     | 0.5659                   |
| Year x Treatment |                              |                  | < 0.0001      | 0.2970                    | 0.9040   | 0.0916                     | 0.9963                   | 0.4800        | 0.5719                    | 0.3741   | 0.2971                     | 0.9330                   |

<sup>1</sup>Non-Filtered = filtered treatment consisted of ambient ozone concentrations monitored within open top chambers; filtered treatments contained ambient ozone concentrations reduced by passing ambient air through charcoal filters (resulting treatment contained ambient ozone concentrations reduced by passing ambient air through charcoal filters (resulting in ca. 63% of ambient ozone))

<sup>2</sup>Berry Weight = per 100 berries (g)

<sup>3</sup>Juice acidity = total acidity (g/l tartaric acid)

<sup>4</sup>Juice sugar = total soluble solids (°Brix)



conducted during five growing seasons (2004-2008) using open-top chambers in south-central PA. Chambourcin grapevines that were exposed to ambient ozone averaged significantly more mean leaf tissue injury over the five growing seasons than did those vines in the filtered-air treatment, confirming the cultivar's susceptibility to ambient phytotoxic levels of ozone. In contrast, Vidal grapevines that were exposed to ambient ozone exhibited minimal ozone-induced leaf tissue injury averaged over the five seasons, confirming its tolerance to ambient ozone. Mean berry weight and juice pH, juice total acidity, and juice total sugars were significantly related to the variable "Year," but not to exposure to ambient ozone for either cultivar, suggesting that ozone-induced foliar injury did not affect fruit juice characteristics important in wine making.

On a greater scale, tropospheric ozone is the major air pollutant capable of adversely affecting the health and productivity of native and agricultural ecosystems in the U.S. On 1 October 2015, EPA reduced the U.S. National Ambient Air Quality Standard (NAAQS) for ozone from 75 to 70 ppb, making it more stringent in order to help protect public health and public welfare, including agricultural and ecological ecosystems [30,31]. Hopefully, this ruling will reduce levels of ambient ozone, preventing future ozone-induced injuries in the U.S. to ozone-sensitive plant species or cultivars such as grapes in PA. Also more knowledge on the influence of tropospheric ozone on wine berry quality should lead to better understandings of the influence of future climate change (with projected increases in ozone concentrations) on wine grape cultivar effectiveness in specific sites and/or industry (vineyard) locations.

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