

***Response of Cabernet Sauvignon and Chardonnay to Fungicide  
Applications and Deficit Irrigation***

A Senior Project  
Presented to  
the Faculty of the Horticulture and Crop Science Department  
California Polytechnic State University, San Luis Obispo

In Partial Fulfillment  
of the Requirements for the Degree  
Bachelor of Science

By

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December, 2012

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## Introduction

Fungicides have been widely used on nearly every crop imaginable and are continually being developed and reformulated as more information on their efficacy on specific crops and pathogens is discovered. It is well known that fungicides are developed and used in order to have a direct effect on one or more pathogens, usually to prevent their establishment on a crop. By spraying fungicides, the occurrence of pathogens can be eliminated or minimized which reduces plant stress and allows for improved yields and crop quality. However, relatively little research has been done on the effects of fungicides on the crops they are applied to. The effects of fungicide use on plants have the potential to affect genetic expression, physiological responses, and aspects of plant development not fully understood yet. One purpose of this experiment is to discover the effects of using the fungicides trifloxystrobin (Flint®), quinoxyfen (Quintec®), myclobutanil (Rally®), and sulfur on Cabernet Sauvignon and Chardonnay vines, grapes, and wine.

The second purpose of this experiment was to test the effects of deficit irrigation on the genetic expression, physiological responses, and plant development- similar to the first purpose mentioned. In arid regions such as California, rainfall is not adequate enough to supply all the water needs of wine grape vines and drip irrigation is often used to supplement. Previous research has indicated that plants can tolerate a certain level drought stress without significant detrimental effects and that in a few circumstances the drought stress can be used as a management technique to reduce costs while improving specific aspects of plant or fruit development. In this experiment, deficit irrigated vines received twenty-five percent of the water usually applied on the vines at our field site from June 7, 2012 until September 17, 2012 in the Chardonnay and October 14, 2012 in the Cabernet Sauvignon.

The information gained from this experiment could be potentially useful for wine grape breeders, vineyard managers, and wine makers. By testing the genetic responses, specifically which genes are being turned on, off, upregulated, or downregulated, wine grape breeders will have better knowledge of the cultivars and genotypes they should or shouldn't use under specific conditions. If specific genes can be identified as responsive to fungicides and deficit irrigation, plant breeders and geneticists may better understand the mechanisms plants use to

resist fungal infection and tolerate drought conditions. These genes could then be used to develop new wine grape cultivars. Expanded knowledge on the genetic responses to fungicide use and deficit irrigation could possibly shorten a breeder's time-line to creating a new cultivar by using varieties known to show positive or neutral responses to fungicide applications and deficit irrigation.

Vineyard managers could use the information from this experiment to better choose specific fungicides to apply to certain wine grape varieties based their beneficial or detrimental responses. The information could also be used to find specific irrigation rates that maximize the water use efficiency of the vines while minimizing any negatives effects on plant and fruit development. Wine makers could use the information to request that vines be sprayed with specific fungicides and that vines be fully or deficit irrigated based on the characteristics they want or don't want in their wines.

## **Literature Review**

Previous research had established that the use of fungicides has a direct effect on the severity and establishment of pathogens on crops and can therefore reduce crop stress. Crops under reduced stress conditions will typically have an enhanced ability to grow and develop to their maximum potential which can increase yields, crop quality, and the economic value of the crop. Wine grapes in most production areas are susceptible to a number of stresses, one of the most significant in terms of economic losses are fungi such as *Botrytis cinerea* and *Erisphe necator*.

More recent research has started to examine the effects of fungicide application on the responses by the plant to these chemicals. Plants have the potential to respond to fungicide applications in many ways that include changes in photosynthetic rates, photosynthate distribution, production of phytochemicals, pigment concentrations, and many other aspects of plant growth and regulation. It is important to understand and account for these plant responses while managing a crops production because the responses can be beneficial as well as detrimental to the crops health.

Saladin et al. (2003) sought to explain some of the physiological responses of grapes to two fungicides, fludioxonil and pyrimethanil, that are used against *Botrytis cinerea*. The

researchers found that the two fungicides stimulated an increase in photosynthesis, photosynthetic pigments, and monosaccharide concentrations up to five months after treatment applications. They also cited that based on their results, they believed the two fungicides to improve nutrient availability and plant vigor which helps the plants fight other forms of infection from pathogens. There were notable differences in the level of response between the three varieties of grapes tested. Chardonnay appeared to be more sensitive to the fungicide treatment and benefited longer than the Pinot noir and Pinot Meunier vines.

Another aspect of fungicide applications to wine grapes is the effects of the residues on the character and profile of the wine produced from the treated grapes. Angeles Garcia et al. (2004) and R.M. Gonzalez-Rodriguez et al. (2010) noted significant changes in the aromatic characteristics of wines from fungicide treated grapes versus untreated grapes. Gonzalez-Rodriguez et al. (2010) cited that the fungicide residues may induce modifications in yeast metabolism that promotes a fruity note to the aroma. They also found that there was potential of the smell of banana which is a sign of a poor quality wine. This result is supported by Alvarez et al. (2011) which stated that fungicide residues had the potential to induce changes in yeast metabolism that could result in the promotion of a fruity aroma, a sweeter balance with a ripe fruit taste, and higher viscosity and cloudiness. Angeles Garcia et al. (2004) noted significant changes in the aromatic compounds in the wines studied but also said that the differences in the aromatic compounds were below the perception threshold.

The response of wine grapes to deficit irrigation was examined by Shellie (2006). In that article she noted previous research that seemed to indicate that deficit irrigation could enhance fruit quality for wine production but could also reduce berry size and yield. The results of her experiment, which used the cultivar Merlot, showed that deficit irrigation could lead to a decline in main shoot growth, reduced yields, reduced titratable acidity, and increased soluble solids. The reduction in shoot growth also increased canopy light transmission.

Based the results of all the experiments discussed in the articles it is clear that it is important to better our understanding of crop responses to fungicide applications and deficit irrigation to further improve crop quality and yields, particularly when it comes to grapes for wine production.

## Materials and Methods

This experiment was conducted at Scheid vineyards near San Lucas, California between June 7, 2012 and October 17, 2012. The wine grape varieties tested were Cabernet Sauvignon, on a 7 x 10 foot spacing and trained to a quadrilateral cordon and Chardonnay, on a 5 x10 foot spacing and trained to a bilateral cordon. All grape-to-wine processing and testing was conducted at California Polytechnic State University. The experiment consisted of fifteen plots for each Cabernet Sauvignon and Chardonnay varieties and three blocks or repetitions per treatment, arranged in a randomized complete block design. Plot size was three rows by six vines in the Chardonnay, and one row by 8 vines in the Cabernet Sauvignon. The five treatments included four fungicides and a deficit irrigation treatment. The fungicides used were trifloxystrobin (Flint<sup>®</sup>), quinoxyfen (Quintec<sup>®</sup>), myclobutanil (Rally<sup>®</sup>), and sulfur which are all commercially used on wine grapes and relatively common in the industry. The deficit irrigation treatment was sprayed with Rally<sup>®</sup> to prevent fungal establishment and ensure consistent data. The first application of fungicides and deficit irrigation began on June 7, 2012. Vines under the sulfur treatment were sprayed once a week while the trifloxystrobin (Flint<sup>®</sup>), quinoxyfen (Quintec<sup>®</sup>), and myclobutanil (Rally<sup>®</sup>) treatments were sprayed every other week.

All fungicide applications were achieved using backpack sprayers and fungicide rates were consistent with the labeling for wine grapes. The Chardonnay vines were sprayed every 14 days with synthetic fungicides and once a week with sulfur between June 7, 2012 and September 3, 2012. Fungicide application continued until 21 days prior to harvest for the sulfur treatment and 14 days prior to harvest for synthetic treatments. Deficit irrigation was maintained until harvest which occurred on September 17, 2012. The Cabernet Sauvignon vines were sprayed every 14 days with synthetic fungicides and once a week with sulfur from June 7, 2012 to September 27, 2012. Fungicide application continued until 21 days prior to harvest for the sulfur treatment and 14 days prior to harvest for synthetic treatments. Deficit irrigation was maintained until harvest which occurred on Oct 14, 2012. All plots were hand harvested. Both the Chardonnay and Cabernet Sauvignon plots were sampled for average berry weight and yield just prior to harvest.

The Chardonnay samples were processed using a de-stemmer/crusher which de-stems and crushes the grapes. The crushed grapes were then pressed using a hand-turned press and racked into one and three gallon carboys per treatment. The juice from the Chardonnay grapes was tested for brix, pH, and titratable acid content between September 25 and 26, 2012. The Chardonnay was then re-racked twice to remove sediment and increase clarity. Argon was added to the carboys to displace any oxygen that was remaining in the carboys due to insufficient volumes of wine.

The Cabernet Sauvignon samples were de-stemmed and crushed using the same equipment as the Chardonnay plots. The juice from the Cabernet Sauvignon plots was also tested for brix, pH, and titratable acid content which occurred between October 17 and 19, 2012. The crushed grapes of the Cabernet Sauvignon plots were then placed in multiple ten gallon plastic buckets for primary fermentation and punched twice a day from October 19, 2012 until their brix level reached zero. After primary fermentation was complete, the grapes were pressed using a hydraulic press and the wine was placed in 5 gallon, 3 gallon, and 1 gallon carboys depending on the volume of wine from each plot.

Both Chardonnay and Cabernet Sauvignon varieties were tested using a hydrometer for brix, a digital pH meter for pH, and titration in combination with a digital pH meter for titratable acid content. The first step of the procedure for the titration using the digital pH meter involves adding 5ml of grape juice to 50ml of deionized water and inserting the pH probe. The second step requires titration using sodium hydroxide to obtain a pH of  $8.20 \pm .03$ . All carboys used for both Chardonnay and Cabernet Sauvignon were sterilized using potassium metabisulphite with citric acid and trisodium phosphate.

The Cabernet Sauvignon was re-racked once between November 13 and 15, 2012 to remove sediment from the wine. In the Cabernet Sauvignon plots the brix, pH, and titratable acid levels were tested after primary fermentation between October 17 and 19, 2012. On November 20, 2012 the Cabernet Sauvignon was tested for color using a color spectrophotometer. The samples were allowed to settle for several days simulating a second racking then diluted by 1:9. Results were produced by multiplying the raw data by 10. The wavelengths used to test the wine were 420nm, 520nm, and 620nm. Before using the color

spectrophotometer all the samples from the plots of Cabernet Sauvignon were adjusted to a pH of as close to 3.4 as possible using sodium hydroxide.

## Results

In the Chardonnay plots the brix, pH, and titratable acid levels were tested prior to fermentation between September 25 and 26, 2012. The data collected are shown in table 1 and figures 1 and 2.

Table 1.

| Chardonnay |              |           |          |
|------------|--------------|-----------|----------|
| Treatment  | Average Brix | pH        | T.A.     |
| Sulfur     | 23           | 3.4533333 | 5.816667 |
| Rally      | 23.833333    | 3.4433333 | 5.966667 |
| Flint      | 24.16667     | 3.45      | 5.316667 |
| Drought    | 23.5         | 3.4233333 | 5.833333 |
| Quintec    | 23.833333    | 3.7133333 | 5.766667 |

Figure 1.

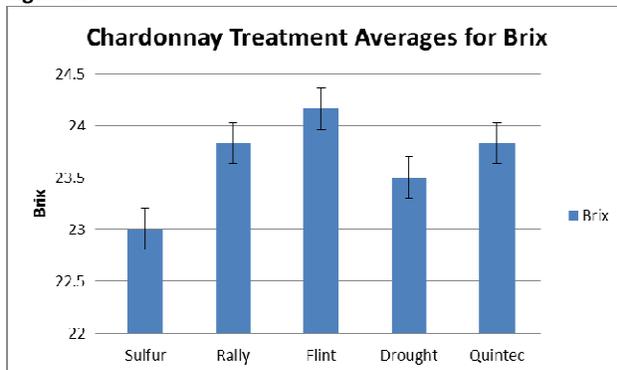
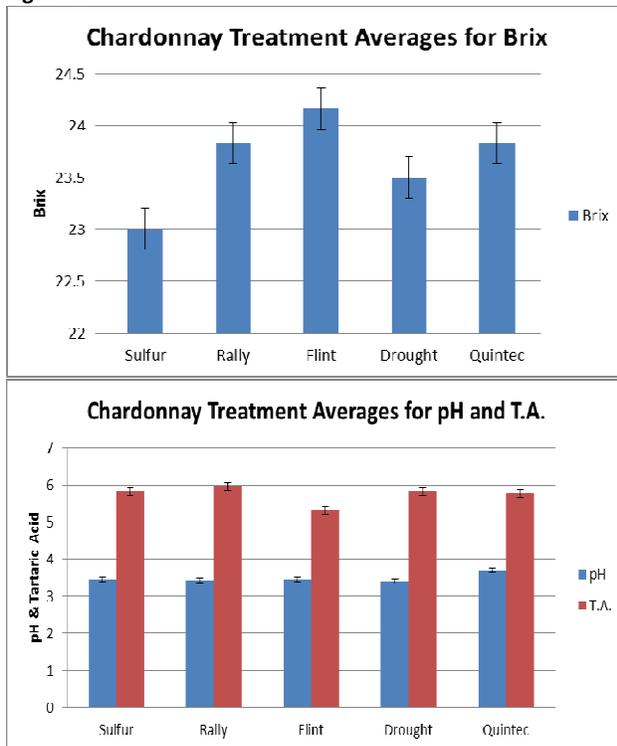


Figure 2.



The average brix of the Chardonnay treatments varied between 23.0 and 24.16. The sulfur treatment had the lowest brix level while the Flint® treatment produced the highest brix level (Figure 1). The average pH ranged between 3.42 and 3.71 while the average titratable acid content fluctuated between 5.31 and 5.96. The drought treatment had the lowest average pH

while Quintec® had the highest pH (Figure 2). The lowest average titratable acid reading came from the Flint® treatment while the highest came from the Rally® treatment (Figure 2). A completely randomized block ANOVA was run and found no significant difference between treatments.

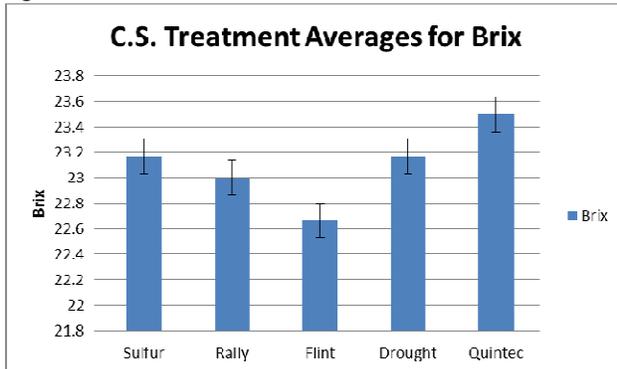
In the Cabernet Sauvignon plots the brix, pH, and titratable acid levels were tested after primary fermentation between October 17 and 19, 2012. The data collected are shown in table 2 and figures 3 and 4.

**Table 2.**

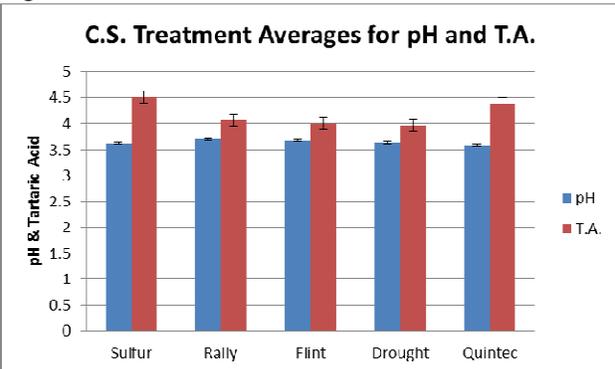
| <b>Cabernet Sauvignon</b> |          |           |          |
|---------------------------|----------|-----------|----------|
| Treatment                 | Ave Brix | pH        | T.A.     |
| Sulfur                    | 23.16667 | 3.62      | 4.516667 |
| Rally                     | 23       | 3.6966667 | 4.066667 |
| Flint                     | 22.66667 | 3.6666667 | 4        |
| Drought                   | 23.16667 | 3.6266667 | 3.966667 |
| Quintec                   | 23.5     | 3.58      | 4.383333 |

The average brix for the treatments varied between 22.66 and 23.5. Flint® had the lowest average brix reading while Quintec® had the highest. The average pH of the treatments fluctuated between 3.58 and 3.69 (Figure 3). Quintec® had the lowest average pH reading while Rally® had the highest. The average titratable acid content varied between 3.96 and 4.51 (Figure 4). The drought treatment had the lowest average titratable acid content while the sulfur treatment had the highest. A completely randomized block ANOVA was run and found no significant difference between treatments.

**Figure 3.**



**Figure 4.**



| Cabernet Sauvignon | Wavelengths           |          |          |           |               |
|--------------------|-----------------------|----------|----------|-----------|---------------|
|                    | Treatment Aver: 420nm | 520nm    | 620nm    | 420+520nm | 420+520+620nm |
| Sulfur             | 1.045                 | 1.425    | 0.4      | 2.47      | 2.87          |
| Rally              | 2.16                  | 3.666667 | 0.646667 | 5.826667  | 6.473333      |
| Flint              | 2.443333              | 4.323333 | 0.72     | 6.766667  | 7.486667      |
| Drought            | 2.266667              | 3.726667 | 0.713333 | 5.993333  | 6.706667      |
| Quintec            | 2.19                  | 3.596667 | 0.703333 | 5.786667  | 6.49          |

The results of the color analysis for the Cabernet Sauvignon treatment averages are represented in table 3 and figures 5 through 8. The

Table 3.

treatment averages for color ranged between 1.045 and 2.26 at 420nm, 1.425 and 4.32 at 520nm, and .4 and .71 at 620nm. At 420nm+520nm the treatment averages ranged from 2.47 and 6.76 while the treatment averages at 420nm+520nm+620nm ranged from 2.87 and 7.48. The lowest average color reading for all treatments at all wavelengths was sulfur. The highest average color reading for all treatments at all wavelengths was the Flint®. A completely randomized block ANOVA was run and found no significant difference between treatments.

Figure 5.

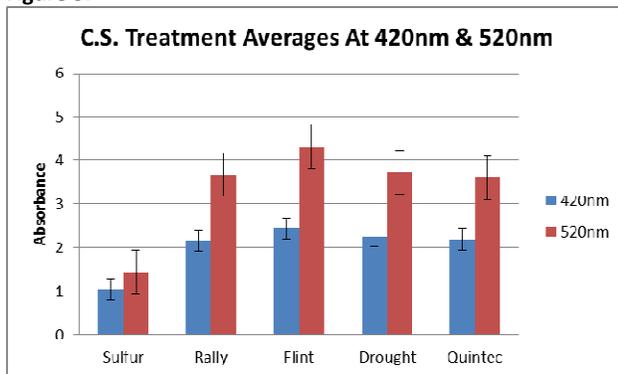


Figure 6.

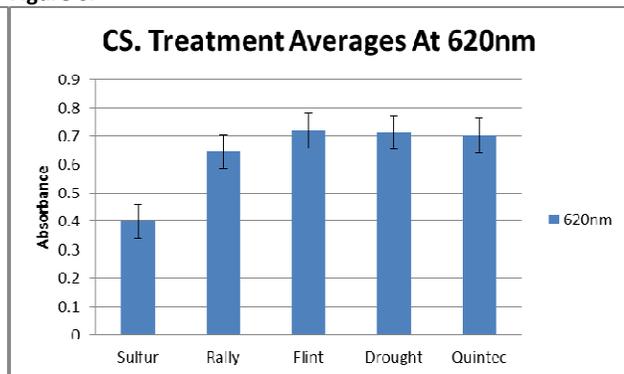


Figure 7.

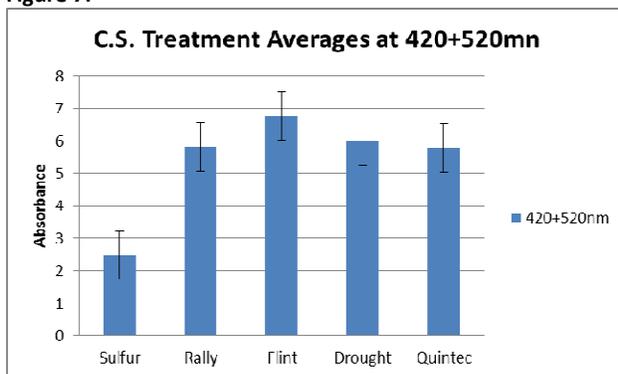
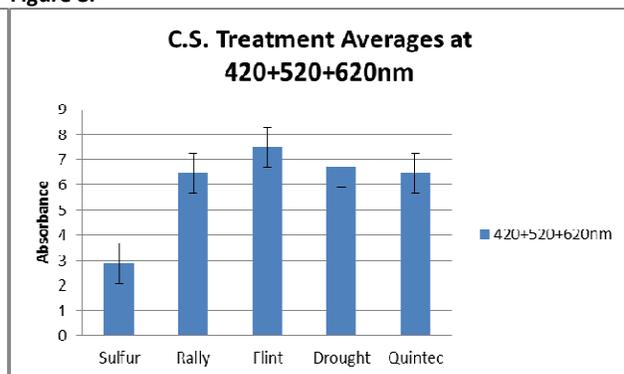


Figure 8.



Both the Chardonnay and Cabernet Sauvignon plots were sampled for average berry weight and average yield per vine just prior to harvest. The results for average yield per vine and average berry weight in the Chardonnay plots are shown in table 4 and figures 9 and 10.

The Cabernet Sauvignon average yield per vine and average berry weight results are displayed in table 5 and figures 11 and 12.

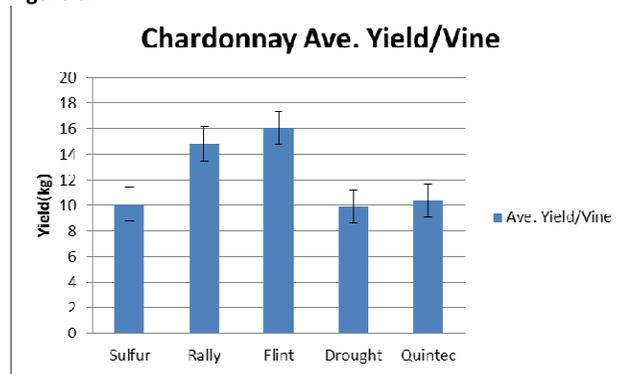
**Table 4.**

| Average Yield Per Vine(kg)     |                 | Average Berry Weight(g)        |                |
|--------------------------------|-----------------|--------------------------------|----------------|
| Treatment Averages(Chardonnay) |                 | Treatment Averages(Chardonnay) |                |
|                                | Ave. Yield/Vine |                                | Ave. Berry Wt. |
| Sulfur                         | 10.12644        | Sulfur                         | 1.316667       |
| Rally                          | 14.81833        | Rally                          | 1.40625        |
| Flint                          | 16.02667        | Flint                          | 1.416667       |
| Drought                        | 9.913939        | Drought                        | 1.297917       |
| Quintec                        | 10.36778        | Quintec                        | 1.308333       |

**Table 5.**

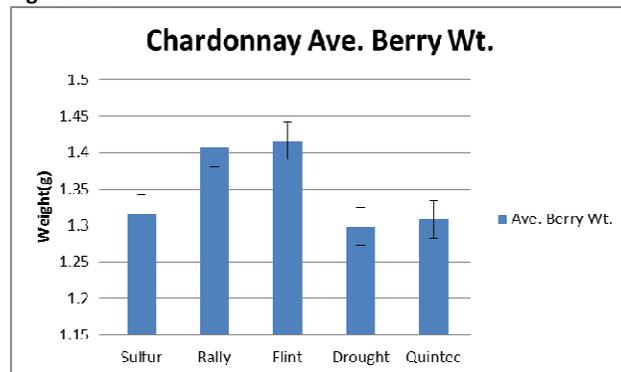
| Average Yield Per Vine(g) |                 | Average Berry Weight(g)  |                |
|---------------------------|-----------------|--------------------------|----------------|
| Treatment Averages(C.S.)  |                 | Treatment Averages(C.S.) |                |
|                           | Ave. Yield/Vine |                          | Ave. Berry Wt. |
| Sulfur                    | 11.4375         | Sulfur                   | 1.139095       |
| Rally                     | 13.16667        | Rally                    | 1.293254       |
| Flint                     | 12.66667        | Flint                    | 1.246825       |
| Drought                   | 11.45833        | Drought                  | 1.113068       |
| Quintec                   | 12.66667        | Quintec                  | 1.189626       |

**Figure 9.**



The lowest average yield per vine in the Chardonnay plots was the drought treatment while the greatest was Flint®. The average yields per vine from the sulfur and Quintec® treatments were nearly as low as the drought treatments. The lowest average berry weight in the Chardonnay was recorded in the drought treatments while the greatest average berry weight was from Flint®. The average berry weights from the sulfur and Quintec® treatments were also nearly as low as the drought treatments.

**Figure 10.**



The average yield per vine was also calculated in the Cabernet Sauvignon plots. The lowest average yield per vine came from the sulfur treatments while the greatest average vine yield was recorded in Rally®. The lowest average berry weight in the Cabernet Sauvignon plots came from the drought treatment while the greatest average berry weight was from Rally®. A completely randomized block ANOVA was run and found no significant difference between treatments.

Figure 11.

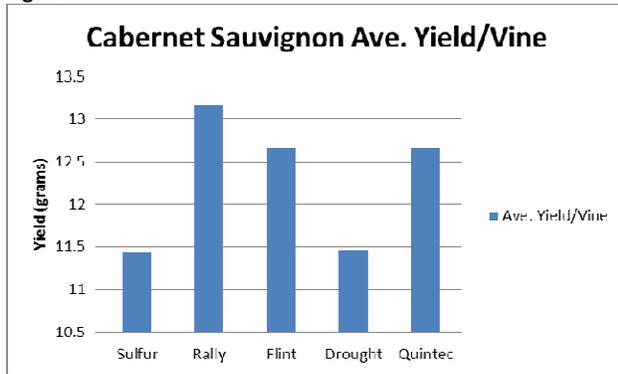
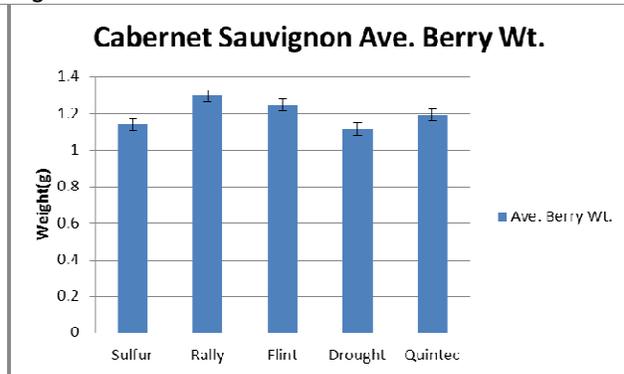


Figure 12.



## Discussion

In the Chardonnay blocks, the averaged sulfur treatments tended to produce lower brix levels than any other treatment average. pH and titratable acid content were minimally affected by treatment and there were no significant differences between the treatments. Average yield per vine (kg) and average berry weight(g) showed a similar pattern to each other in response to fungicide and drought irrigation application. Sulfur, drought, and Quintec® treatments results saw reduced yields, at about 5-6 kg per vine, compared to the average treatment results for Rally® and Flint®, which produced about 15kg per vine. However, these results were not found to be significantly different. Average berry weight followed a similar pattern, though the difference in results is not significant at an individual berry level.

For the Cabernet Sauvignon blocks, Flint® resulted in the lowest treatment averages for brix. The differences for brix between all other treatment averages were marginal. There was almost no difference between all treatment average results when it came to pH and titratable acid content. The treatment averages resulting from the color spectrophotometer indicated that sulfur had the greatest effect on the color and absorbance of the wine. The averaged sulfur treatments produced about half the absorbance as all other treatment averages at all wavelengths tested. The difference between sulfur and the other treatments could be noticed visually, as it caused a “miliness” or white haze in the wine color. There was relatively little

difference between the averaged color absorbance of Rally®, Flint®, and drought treatments at all wavelengths.

The average yield per vine and average berry weights of the averaged treatments in the Cabernet Sauvignon both followed similar patterns to fungicide and drought irrigation application. Sulfur and drought treatments both produced the lowest average yield per vine at around 11.45 grams each. The Rally®, Flint®, and Quintec® produced greater yields that exceeded 12.67 grams. The average berry weights followed a similar pattern in response to the treatments although the differences between the results are not significant for the average yield per vine or at the individual berry level.

Based on the results of this experiment, Rally® and Flint® are the optimum fungicide choices for producing the greatest yields and berry weights. In the Cabernet Sauvignon blocks, Flint® also produced the greatest absorbance readings at all wavelengths tested. In addition, Flint® produced the highest brix levels in the Chardonnay blocks. The effects of Flint® on the brix in Cabernet Sauvignon blocks seemed to have the opposite reaction than the Chardonnay and produced to lowest brix result when compared to all other treatments. For obtaining the highest brix level in Cabernet Sauvignon, it appears that Quintec® is the best choice.

The worst fungicide to use on either Chardonnay or Cabernet Sauvignon appears to be elemental sulfur. The sulfur produced the lowest brix result in the Chardonnay blocks, reduced color absorbance in Cabernet Sauvignon, reduced yields compared to Rally® and Quintec®. The sulfur also tended to burn the leaves of the vines and produce grape clusters that were visually inferior to all other treatments. The berries of the sulfur plots were often shriveled or wrinkled.

The drought treatment, which was sprayed with Rally®, produced reduced yields compared the regularly irrigated Rally® treatment averages. It also produced lower brix level in the Chardonnay while increasing the brix level in Cabernet Sauvignon, when compared to its Rally® counterpart. The differences recorded in the two treatments for color absorbance, pH, and titratable acid were relatively minimal.

In conclusion, this experiment suggests that fungicides and deficit irrigation do have effects on the growth and characteristics of the vines, berries, and wine of Cabernet Sauvignon and Chardonnay varieties. It also shows that more research needs to be conducted to better

understand the causes and effects resulting from fungicide use and deficit irrigation on grape vines and other valuable crops. More research on other grape varieties and in different climates would also benefit the grape and wine industries by providing more information on the optimal fungicides to use based on the characteristics they desire. Further studies on deficit irrigation would also help vineyard managers better manage their water use where water is a rare and expensive resource.

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