An investigation into the maturity and ripening characteristics of Syrah grapes when sprayed with HML32 foliar spray.

By

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A report submitted in partial fulfilment of the requirements for the programme

CONCURRENT BACHELOR OF VITICULTURE AND WINE SCIENCE

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ABSTRACT

The title of this investigation is an investigation into the maturity and ripening characteristics of Syrah grapes when sprayed with HML32 bunch line spray. This will be investigated on two different vineyards sites within Hawkes Bay, One in the Bridge Pa triangle and the other in the Gimblett gravels. The sites were divided into control and treatment areas. This will be examined and investigated by Leigh O'Connor, a third year degree student in the Concurrent wine science and viticulture degree at EIT in Hawkes Bay. The methods employed throughout this experiment include marking out the respected bays from each vineyard and carry out berry sampling. Berries were then taken back to the EIT laboratory to be analysed and measured for berry weight, brix, pH, TA and on the final harvest date berries were kept inside a freezer to preserve them (Due to Covid-19) until they were ready to be analysed for phenolics and anthocyanin content later in the year. Results showed there wasn't any significant differences between berries sprayed with HML32 and berries not sprayed in the controlled areas of the vineyards except for one TA result which showed there was a significant difference on the very last harvest date. It was concluded that implications due to Coronavirus were quite prevalent and in turn led to some inconclusive results. The investigation should be repeated again with more sampling carried out.

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EIT HAWKES BAY

CERTIFICIATE OF ORIGINALITY

AND

AGREEMENT FOR THE RETENTION AND USE

I, Leigh O'Connor, certify that I am the author of the project titled:

An investigation into the maturity and ripening attributes of Syrah grapes when sprayed with HML32 foliar spray.

Which is submitted to EIT for assessment on this day.

I certify that this project has not been previously submitted by me or any other person to EIT or any other tertiary institution.

Should the project be favorably assessed, I agree that if requested I will provide a copy of the project as specified to be lodged in the EIT library. I also agree that the project be accessible for the purposes of publication, study and research.

Signed:_____ Date:_____

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

The subsequent pages include:

- Introduction
- Literature review
- Material and methods
- Results
- General discussion
- Conclusions and recommendations
- Reflection
- References
- Appendices (should contain all raw data)

CHAPTER ONE - INTRODUCTION

Syrah is up and coming in Hawkes Bay and there is a big market and desire to promote it. However, as with anything, there are issues with this particular variety. One of these issues is what this trial will focus on. Syrah tends to be prone to botrytis near end of growing stage so viticulturalists will choose to pick the fruit earlier than they might want to so the disease can be avoided. A way to combat this would be make Syrah grapes more resilient to botrytis so it can hang out on the vine for longer to allow further development of aroma and phenolic compounds that make Hawkes Bay Syrah so unique.

With an industry aim to Increase resistance to botrytis on Syrah berries, more research and investigating must be carried out to find a way of supporting this variety and creating a growing program that produces high quality New Zealand Syrah.

In this investigation, a well -known spray called HML32 was elected for this particular trial. This aim of this investigation was to find ways to improve ripening and maturity characteristics of Syrah using late season HML32 foliar sprays and how the impacts of HML32 bunch spray application affects the accumulation of sugar and phenolic compounds in Syrah grape berries.

HML32 was applied at veraison, and again 10 days after the first application. Sampling was carried out every two weeks from the first date of 13.02.2020 up until 24.03.2020. However, samplings were not able to be taken at harvest date because of implications with Covid-19 lockdown restrictions and the results were inconclusive.

CHAPTER TWO - LITERATURE REVIEW

The idea of this trial is to explore what prospective benefits an HML32 foliar spray might have on Syrah grape ripening, maturity and overall quality. The main focus of the following literature review is to find and evaluate any appropriate research that could link ideas between HML32 spray and how it can enhance maturity and ripening as well as potentially outlining any possible areas where further research may be required in this subject. All literature written in this section will be comprised of the product description, journal articles and previous thesis work completed on this topic to retain that all information sourced for this research is reliable.

The wine industry in New Zealand has been cultivating and producing grapes for well over 100 years. However, it has only been in the last 30 years that New Zealand has grown a worldwide acclamation for its wine. in the space of 10 years from 1997 to 2007, New Zealand wineries have climbed from 262 to 543 wineries and from 7410 hectares to 24,660ha (NZ wine, 2020). Hawkes Bay is New Zealand's second largest grape growing region with over 5000ha of vineyards planted with 339Ha of that being Syrah. This accounts for over 60% of New Zealand's total Syrah (NZ wine, 2020). These vineyards run across historic landscapes and sub regions which comprise of coastal areas, hillsides, alluvial plains and river valleys (NZ wine, 2020). As backed up by (Chappell, P. 2013), Hawkes bay is one of the country's warmest regions with an average GDD of 1476, annual sunshine hours in the vicinity of 2200 and rainfall of less than 1000mm.

Syrah is believed to be traced back to Dureza and Mondeuse blanc varieties that hail from the Rhone valley in France. However, previous beliefs entailed that Syrah originated from Shiraz, a town in ancient Persia (Robinson, 1999).

Wine quality can often be defined as a wine when the alcoholic strength, acidity, residual sugar and the tannins all flatter eachother so every component blend in together and no one overpowers another on the palate (Robinson,1999).

Grapevines produce non climacteric berry fruit that show a double sigmoidal growth curve on which there are three stages of growth. Stage one and three display rapid growth whereas stage two is referred to as the lag phase (Coombe, 1992). The transition stage from stage two to stage three is known as the onset of ripening where the berries will begin to soften and grow, decrease in acidity, increase in sugar accumulation and in red varieties such as Syrah, colour will begin developing (Coombe, 1992) as can be seen in figure 1 below.

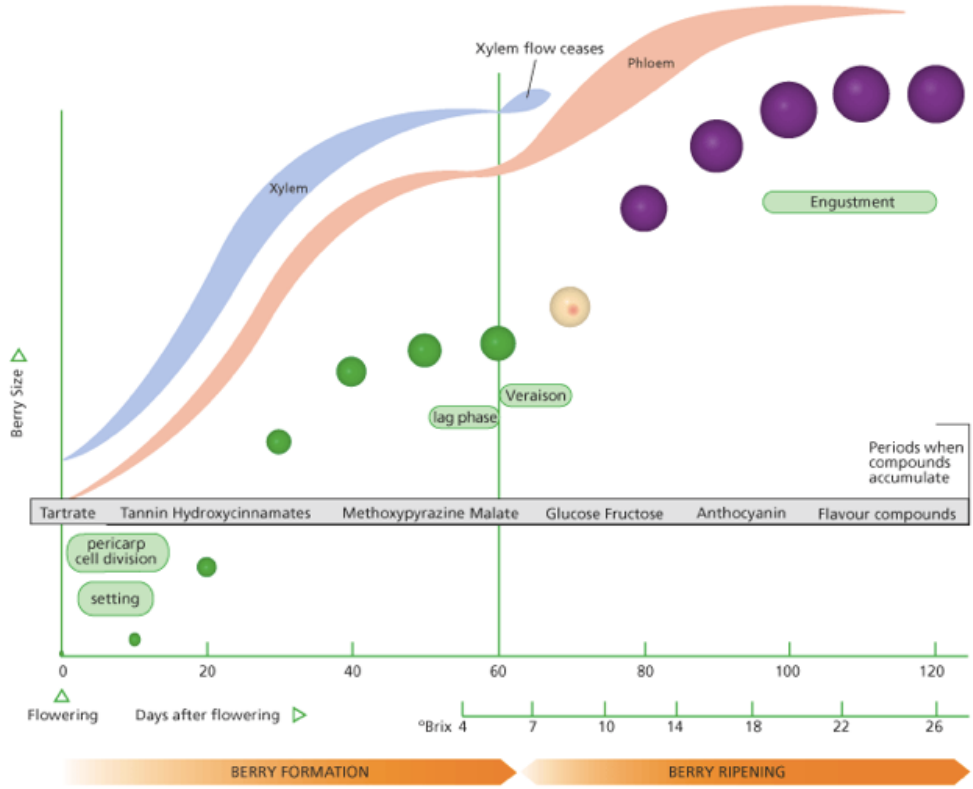

Figure 1: Figure displaying the approximate size and colour of berries throughout different stages of the growing season. Furthermore, showing approximate time of various compound accumulation (Urska, 2017).

An important element of climate is temperature which can ultimately affect a grapevines entire processes such as respiration, transpiration and photosynthesis. Higher temperatures can create an increased rate of photorespiration and can also denature enzymes (Rogiers, 2004).

Higher temperatures have also been found to prevent the accumulation of anthocyanins in the skins of grape berries. As stated by Yamane, et all (2006), ABA hormones can increase the anthocyanin enzyme genes therefore directly affecting anthocyanin accumulation. Temperatures exceeding 30 degrees or more during the ripening period can influence to creation of ABA in the berry skins. It can also be said that solar radiation has the ability to influence flavour and composition of fruit as backed up by (Robinson & Smart. 1998). Brix levels,

anthocyanins, phenolics and tartaric acids can decrease due to a decrease in sunlight. Sunlight is an essential aspect in anthocyanin production because anthocyanin creation relies on light regulated enzymes (Rogiers, 2004). On the other hand, anthocyanins are found within the vacuoles of a grape berry and if exposed to too much sunlight, the skin cells can effectively be sunburnt resulting in discolouring and therefore a reduction in the overall grape quality (Rogiers, 2004).

Anthocyanins are responsible for giving the colour of fruit such as cherries, berries, black currents and grapes (Jayaprakasam, et al. 2006) and can also help to attract pollinating insects and animals, can discourage predators and give protection from harmful UV radiation (Holton & Cornish, 1995). Phenolics are largely found in the stems, skins and seeds of red grape varieties and encompass a large number of compounds that can be classed as flavonoid or non-flavonoid (Robinson, 1999). Interestingly, anthocyanins are antioxidant polyphenol compounds which have been linked to reducing the risk of cancer (Jayaprakasam, et al. 2006). Because red wine has antioxidant flavonoids within it, there has been a great deal of recognition in the beverage as palatable source of antioxidants for consumers (Kitson & Stanley, 2001).

What is HML32?

HML32 foliar spray is an active fungicide that is used for the control of diseases such as powdery mildew and botrytis in grapevines (Henry, C. 2018). It is an adjuvant which helps to improve the coverage of fungicide over grapes. The formulation is certified as a permitted fungicide by BioGro which can be used on organic properties. Ingredients within HML32 include 15 – 18% fatty acids as Potassium salts, 24-28% Potassium bicarbonate, and water is used to balance the remainder (Henry, C. 2018).

Henry Manufacturing have conducted numerous amounts of studies using HML32 as a resistance treatment for powdery mildew and botrytis. The trials started off as mere hand spraying methods and have transitioned over time with machinery after displaying outstanding resistance to botrytis, sour rot and an enhanced maturity (Henry, C. 2018). Eight seasons of trials have been conducted and recorded on the henry manufacturing website (Henry, C. 2018) and have used different combinations of HML32 with other compounds such as copper,

Sulphur and HML Silco and have also found that HML32 with HML Silco can offer a good replacement to the use of Sulphur.

HML32 is currently only available in New Zealand but there are current movements towards it being made available in Australia and America.

CHAPTER THREE - MATERIALS AND METHODS

For this research two different sites were chosen. One was within the Bridge Pa triangle in Hawkes Bay and the other was situated on the Gimblett gravels also in Hawkes Bay (Figure 2).

Figure 2: Aerial photograph of TK and Te Awa vineyards

Both sites have relatively similar soils, vine health and had even grounding so as to get the most accurate samples as possible by minimising potential variable errors. On the Te Awa vineyard block, the Syrah sampled was planted in 2013 on Schwartzman rootstock with the Syrah clone 470. The TK vineyard block was planted in 2001 on Schwartzman rootstock and clone MS Unison.

Site set up

For both sampling sites, 4 control rows and 4 trial rows were chosen with a spacing of one row to minimise any spray drift or cross contamination (Figure 3). These rows were specifically chosen as they are the most uniform and are a good representation of the whole vineyard. Sampling areas were within the 2 middle

rows to ensure the trial product had adequate coverage over the berries and that the control row had no cross contamination. Besides the different treatment of the vines, the treated and controlled areas in both vineyards were treated the same way viticulturally pre and post spray treatments.

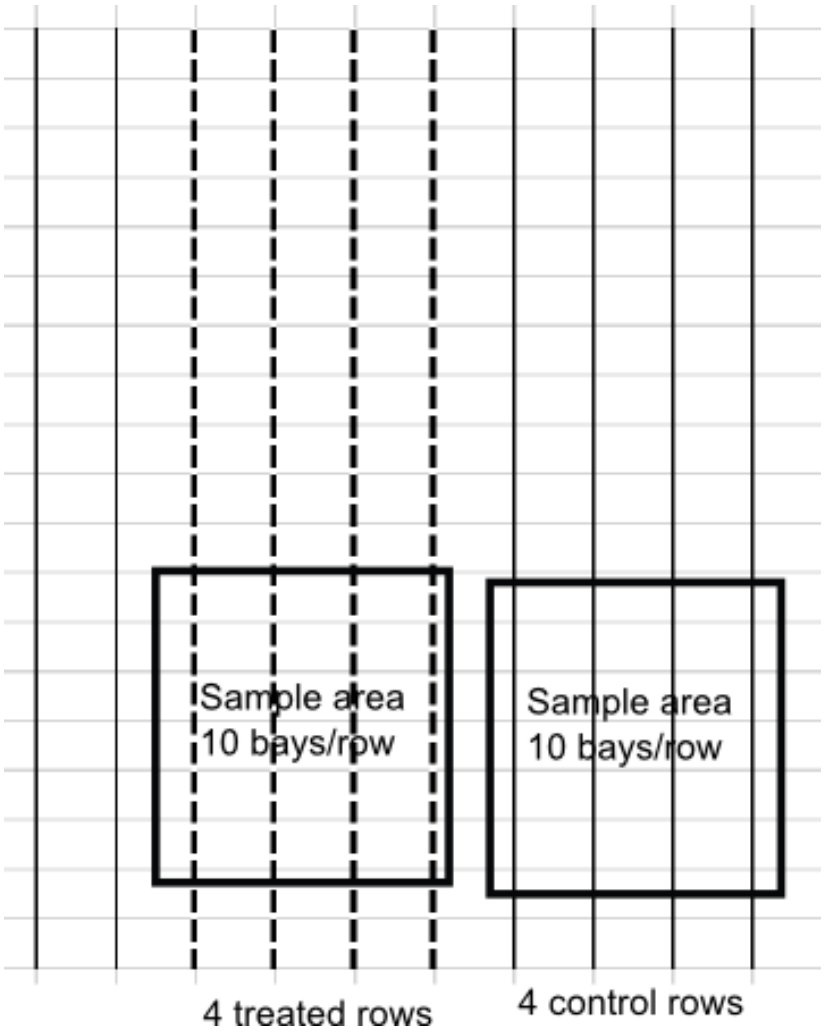

Figure 3: Diagram displaying the layout of chosen bays on each vineyard. One control area and one treatment area.

Site spraying methods

HML32 spray was applied to the vines at a rate of 1.25L HML32 per 100L of water. Both sites were 2 cane VSP which required the spray mix to be applied at a rate of 9L per 100 metres. The spray was applied in both directions down the rows of each vineyard to maximise coverage until runoff. Spraying was done on days where the sun didn't dry out the sprayed areas too much and it was applied on the bunch line. The spray was not mixed or sprayed with any other kind of product, it was not applied on water stressed vines, nor were any potassium-based fertilizers present.

The first application of HML32 spray was applied approximately 7 days after 50% veraison and sugar levels were roughly around 8.5 brix. The second application was applied 10 days after the first application was completed on both vineyards.

Berry Sampling

Bays were individually chosen in each row for both vineyard sites. This helped to ensure vines were healthy and looked to have similar yield levels in order to give a comprehensive representation of the vineyards. Berry samples were picked at random from 5 bays that were previously tagged down the rows.

From each of the 5 bays in the specified row, 50 berries were taken per bay and kept in separate bags for accuracy. In total, 1000 berries were taken. 500 per vineyard and 250 each per control and treatment areas.

The first samples were picked off both vineyards on the morning of February 13th, 2020 and approximately every 2 weeks following the date until March 24th, 2020 (pre coronavirus lockdown).

Once samples from both vineyards were collected, they were immediately taken to the EIT lab for analysis. This analysis included recording the weight of each bag collected (40 in total) to find the average berry weights as well as the brix, TA, PH and sensory features of each sample. These analysis methods took place roughly every two weeks until harvest.

Table 1: Sampling dates for each vineyard.

Sample 1	13.02.2020
Sample 2	25.02.2020
Sample 3	11.03.2020
Sample 4	24.03.2020

On March 24th, an extra 50 berry sample for each bay was collected which was then placed in a -80°C freezer to be later used after lockdown for phenolic analysis.

At harvest time, another 100 berry sample from each bay was meant to be collected and tested for Brix, TA,PH, Phenolics, total anthocyanins, YAN, organic acids and yield. Unfortunately, due to Coronavirus, these samples were not able to be taken and therefore the experiment was semi inconclusive in that all the

data could not be officially recorded, only phenolics could be tested at a later date.

Chemical analysis

Immediately after picking, the samples were taken to the EIT laboratory for analysis.

Table 2: Chemical analysis methods used for vineyard samples

Berry weight	Taking the weight of the empty bag into consideration, the full sample bags were then counted to ensure there was exactly 50 berries and then the bag was weighed on a set of scales. This weight was then divided by 50 to find the average berry weight of each sample.
Brix	All berries were then crushed within their sperate bags to extract as much juice as possible. The juice was then poured through a strainer and into a clean beaker to remove solids. Some of this juice was then placed onto a clean and calibrated electronic refractometer so the brix levels could then be recorded.
PH & TA	Remaining juice from each bag was then transferred into a pre labelled centrifugal tube and placed into a centrifuge for 5 minutes on the highest setting. Once centrifuged, the juice was then transferred into another clean set of bigger beakers and placed onto an auto titrator machine which measures the PH and TA for you. These results were then recorded as needed.
Phenolics	The extra frozen berries collected on the last sampling date were taken out of the -80°C freezer a few months later 24 hours prior to analysis. The phenolics and anthocyanins of the berries were analysed as per the method from (Iland et al. 2013)

Statistical analysis

All data collected over the sampling dates was transferred into an excel spreadsheet were the technology could help to determine deviations and trends

within the data collected. Results from this can all be viewed within the results section.

Box and whisker graphs were created using the phenolic data to plot the mean values of phenolic compounds per treatment in both vineyards. Line graphs were created using the raw data of berry weights, PH, TA and brix with error bars included to displays differences and similarities within the two vineyards control and treatment areas. All graphs made were produced in an easy to follow and logical way.

T testing was carried out on excel and is also displayed in the results section.

The program Jasp was used to conduct significance, post-hoc and ANOVA testing on the berry weight, brix, PH and TA. This program was used for its simplicity and efficiency displaying data that is easy to follow. All outcomes for this are displayed in the results section.

CHAPTER FOUR - RESULTS

Key:

Vineyard A – Te Awa

Vineyard B – TK

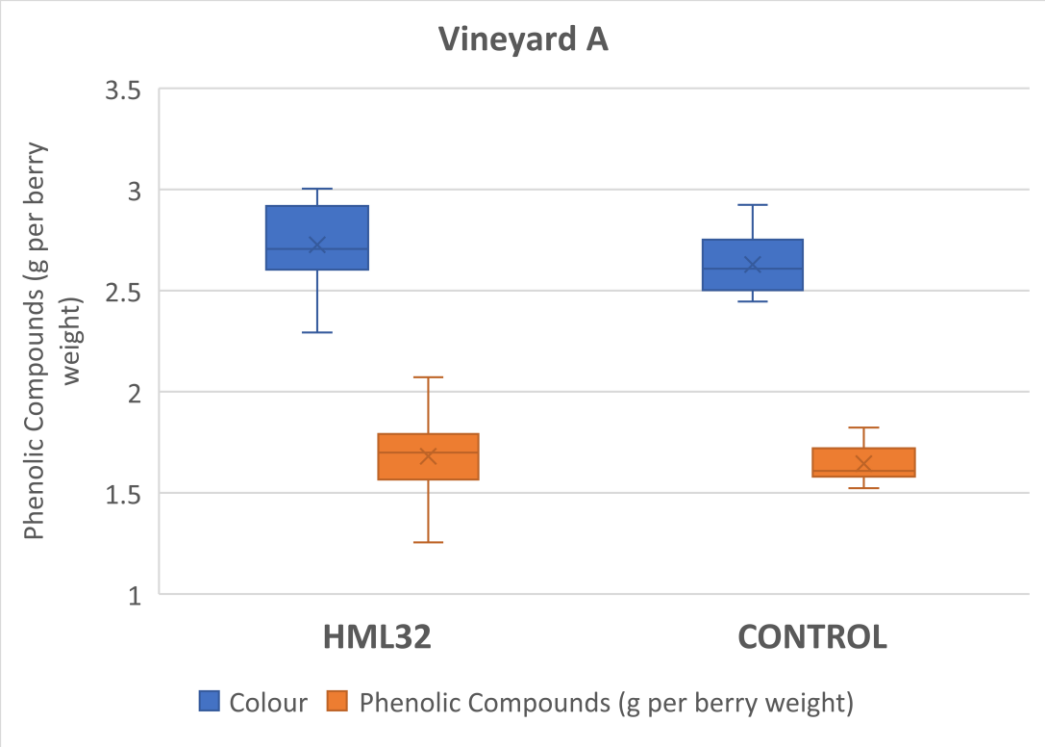

Figure 4: Plot of the mean values of phenolic compounds per treatment in Vineyard A.

Figure 4 above demonstrates the total average phenolic compounds measured within the berry skins picked on 24.03.2020 at Vineyard A. No significant differences were found between the control and treatment based on a t-test conducted (Table 3).

The berries that were collected were analysed for total colour and phenolics. As seen in figure 3, the variation between Colour for the treated vines and the control vines was quite different. Vines applied with HML32 tended to have higher levels of colour however there also a lot of spread within the data.

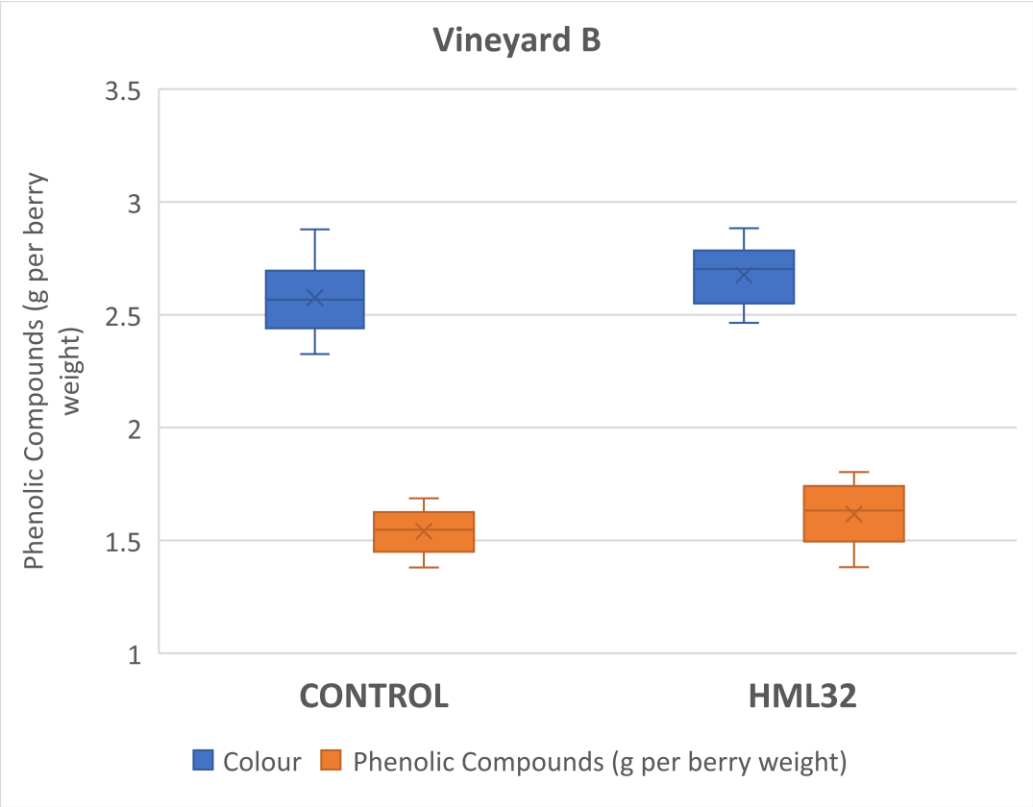

Figure 5: Anthocyanin and phenolic data for vineyard B which compares treated and controlled vines. (Appendix B)

Figure 5 above demonstrates the total average phenolic compounds measured within the berry skins picked on 24.03.2020 at Vineyard B. No significant differences were found between the control and treatment based on a t-test conducted (Table 3).

Table 3: T-test values for Vineyard A and B phenolic and anthocyanin contents

Vineyard A	Colour	0.26131998
Vineyard A	Phenolics	0.6294377
Vineyard B	Colour	0.09291993
Vineyard B	Phenolics	0.10160538

Anthocyanin and Phenolic content of berries were assessed from both vineyard A and B. The data was plotted and then significance tested using a t-test. Both vineyards concluded similar results in that there appeared to be no significant difference between the phenolic or anthocyanin content of the different berries.

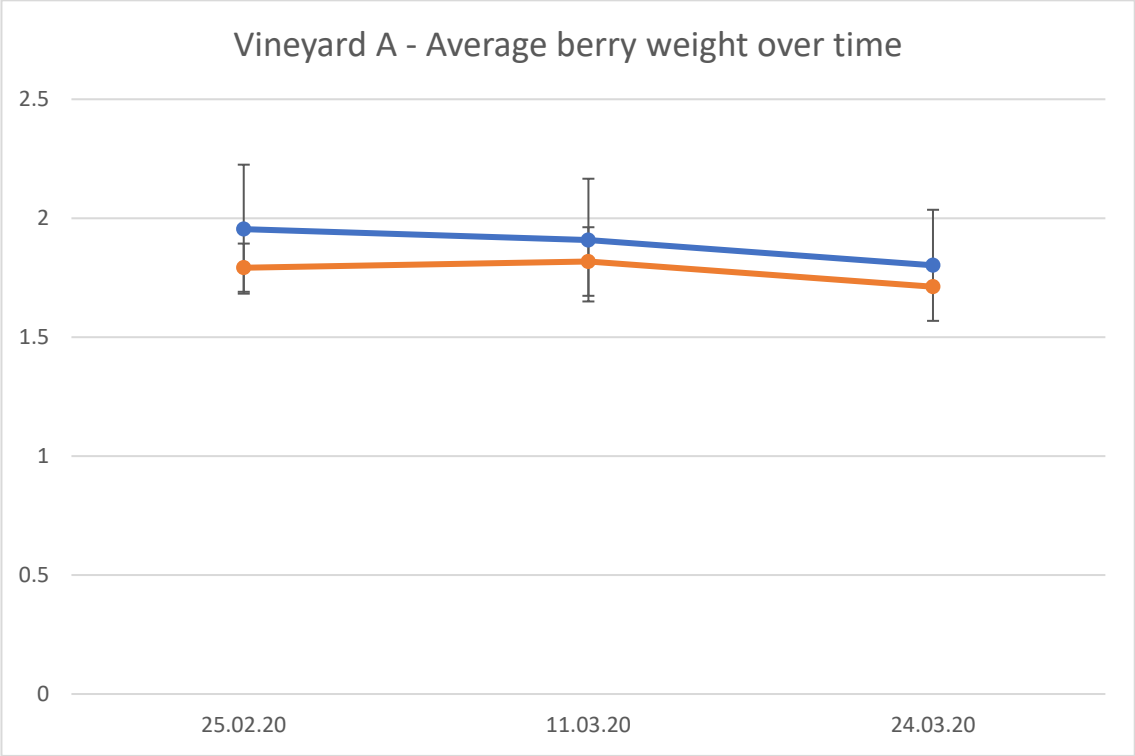

Figure 6: The average berry weights of vineyard A treated and controlled vines over time. (Appendix A)

Figure 6 above displays the average berry weights for both the control (blue) and HML32 treated (orange) vines. The very first sampling date failed to acquire any results for average berry weight as seen, however the other three dates were collected. The overall trend shows there seems to be a decline in average berry weight over time. This could be due to the fact that as sugar accumulates inside the berry over the growing season, water concentration decreases which therefore might explain the decline in weight.

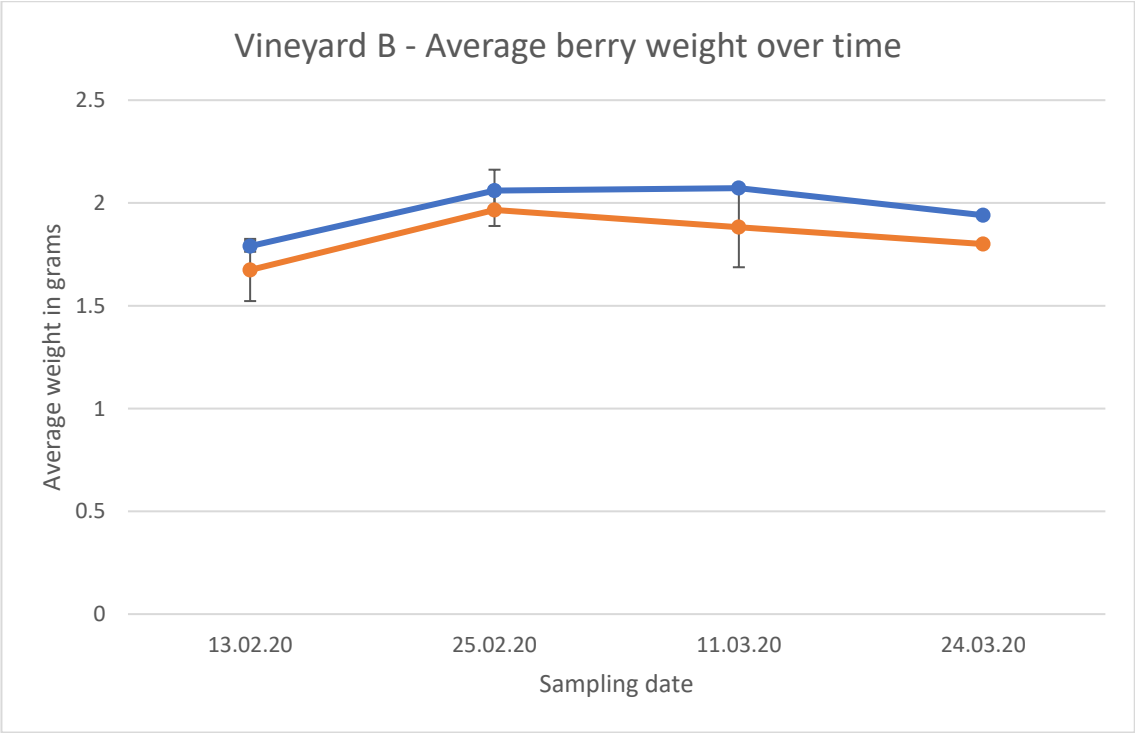

Figure 7: The average berry weights of vineyard B treated and controlled vines over time. (Appendix A)

Figure 7 above displays the average berry weights for both the control (blue) and HML32 treated (orange) vines. Trendlines show there is an initial significant increase within the 2 weeks of sampling 1 and sampling 2. This could be due to rapid growth of the berry at that time. From sample 2 however, figure 6 shows there is a steady decline in average berry weights which again is similar to that of vineyard A (figure 6 and may be explained by sugar concentration overtaking water in the berry resulting in a weight decrease.

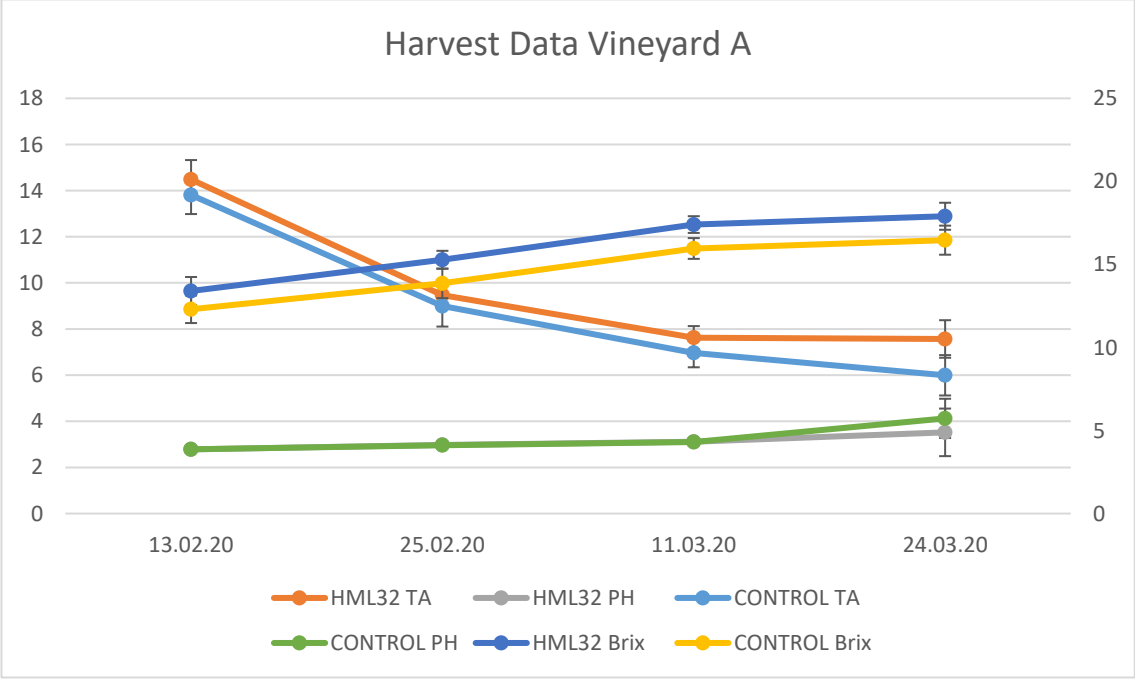

Figure 8: Brix, PH and TA results of vineyard A comparing controlled and treated berries.

All of the data collected over the four sampling days during the growing season for vineyard A are displayed above in figure 8. After performing an ANOVA post hoc test (appendix 4), no statistical differences were shown between the control and treated berries except for on the very last sampling day 24.03.2020. That particular day showed that after performing an ANOVA post hoc test, there was a significant difference between Vineyard A titratable acidity data of controlled and treated berries.

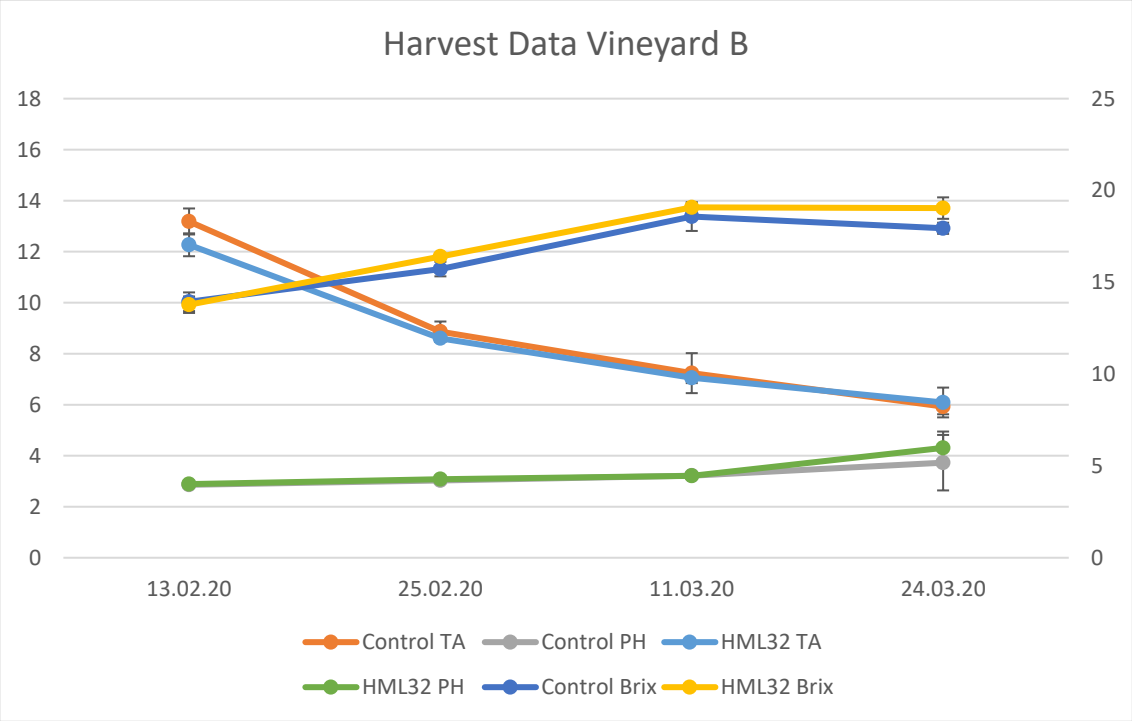

Figure 9: Brix, PH and TA results of vineyard B comparing controlled and treated berries.

All of the data collected over the four sampling days during the growing season for vineyard A are displayed above in figure 9. After performing an ANOVA post hoc test (appendix 5), no statistical differences were shown between the control and treated berries.

CHAPTER FIVE - GENERAL DISCUSSION

Overall, the investigation was very interesting although the results were somewhat inconclusive because of Covid-19.

If the experiment were to be repeated some changes could be made to the methods so as to gain more accurate results. 2020 however, proved to be a very challenging year and so it is understandable that all information gathered for this investigation might not have been fully completed or done to the best of its ability. One aspect that would have been beneficial to do would be to have done a rot assessment prior to harvest. Being able to do this would have been noteworthy to observe if the HML32 spray had actually done anything in terms of botrytis prevention or not. This season would not have been a good time anyway as there was very minimal rot within the vineyards and obviously as already mentioned, Coronavirus brought everything to a standstill.

Results showed that berry weights underwent a stark increase between the first and second sampling dates and then tended to slowly decrease from there until the last sampling date. This could be due to an initial influx of water into the berry at the beginning of the sampling dates and then decreasing towards the end as water levels decrease and sugars increase. There was no significant difference between berry weights of treated and treated berries.

Brix levels display an obvious incline on the graphs above showing that as time goes on, sugars accumulate in the berry and will eventually plateau. Both vineyards project a similar curve in sugar accumulation with no significant differences between treated and non-treated berries.

PH levels across both vineyards don’t show any change until the final sampling date where they display a slight increase which will be due to acidity levels in the berries declining as the berry becomes riper.

Titratable acidity is the only aspect where a significant difference was apparent after ANOVA and T-testing. There is an obvious different in both vineyards when looking at the respective graphs. Vineyard A starts off with a higher TA reading than vineyard B and drops further than vineyard A at the second sampling date. The final sampling date is where the significant difference occurs with a p value of less than 0.05. In contrast, vineyard B shows that the treated and non-treated start off different but as the growing season goes on, they become almost the same and there is no significant difference.

Phenolics data from both vineyards again displayed no significant differences between treated and non-treated vines after performing ANOVA and T-tests. Vineyard A, however, did show a greater spread of results compared to vineyard B.

CHAPTER SIX – CONCULSIONS AND RECOMMENDATIONS

To sum up this investigation, it can be stated that there were no significant differences between controlled vines and vines that had been sprayed with HML32. There was however one anomaly where on the final sampling date, the titratable acidity showed a significant difference between treated and non-treated vines. This could have been due to human or machine error or could have been accurate results from the spray itself.

In future, completing a rot assessment prior to harvest would be very beneficial to examine if there were any effects on the berries from the HML32 spray. Completing additional sampling analysis would also be recommended as more data can be used to find trend and averages over the growing period. In particular, carrying out a sample collection prior to the first application of HML32 would have interesting to see where the berries naturally started off in both blocks, then there would be a standard result to compare the first sampling to after the first round of treatment.

CHAPTER SEVEN - REFLECTION

Looking back on the investigation, I have thoroughly enjoyed the process although it was somewhat stressful at times, especially with a global pandemic putting a damper on things, I found the whole process a great learning curve and it turned out to be an experience I will keep with me for my career and it will be a reminder of where I first started. I have learnt (although am still not great at) to read through a lot of scientific material and pull out parts of journal articles beneficial to the topic. Additionally, it gave me a small insight into what it might be like to be in the industry once I have completed my degree in terms of what lab work I might do and how to conduct berry sampling in a vineyard.

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APPENDICES

Appendix A

Raw data gathered over growing season on various dates for both vineyards A (Te Awa) and B (TK) including brix, titratable acidity, PH and average berry weight measurements.

DATE	VINEYARD	TREATMENT	ROW	BAY	BRIX	TA	PH	WEIGHT (g)
13.02.20	A	Control	488	3	12.4	14.24	2.79	
13.02.20	A	Control	489	4	12	13.37	2.8	
13.02.20	A	Control	488	5	12.9	14.67	2.79	
13.02.20	A	Control	489	6	11.9	12.59	2.77	
13.02.20	A	Control	488	7	12.3	14.19	2.78	
13.02.20	A	Treatment	484	3	12.9	15.73	2.77	
13.02.20	A	Treatment	483	4	13.4	13.97	2.80	
13.02.20	A	Treatment	483	6	13	14.25	2.74	
13.02.20	A	Treatment	484	7	13.4	14.38	2.78	
13.02.20	A	Treatment	483	8	14.3	13.98	2.82	
13.02.20	B	Control	A22	3	13.9	17.6	2.81	92.1
13.02.20	B	Control	A23	4	15.3	17.4	2.89	92.5
13.02.20	B	Control	A22	5	13.7	17.6	2.82	89.8
13.02.20	B	Control	A23	6	13.5	16.7	2.92	91.2
13.02.20	B	Control	A22	7	13.3	18.6	2.86	83
13.02.20	B	Treatment	A22	12	14.4	15.7	2.89	79.9
13.02.20	B	Treatment	A23	14	13.6	16.7	2.89	73.1
13.02.20	B	Treatment	A22	15	13.5	16.5	2.90	83.9
13.02.20	B	Treatment	A22	20	14.4	15.8	2.89	91.7

13.02.20	B	Treatment	A22	22	13.0	17.1	2.87	89.6

25.02.20	A	Control	488	3	13.8	9.85	2.93	109.2
25.02.20	A	Control	489	4	12.7	8.38	2.99	84.3
25.02.20	A	Control	488	5	15.5	9.95	2.95	105.1
25.02.20	A	Control	489	6	12.8	7.94	2.94	82.5
25.02.20	A	Control	488	7	14.5	8.85	3.00	110.3
25.02.20	A	Treatment	484	3	14.5	10.33	2.95	88.5
25.02.20	A	Treatment	483	4	15.1	8.88	2.98	83
25.02.20	A	Treatment	483	6	15.4	9.35	2.96	96
25.02.20	A	Treatment	484	7	15.2	9.24	2.97	89.3
25.02.20	A	Treatment	483	8	16.2	9.56	3.03	94.8
25.02.20	B	Control	A22	3	16.2	9.57	2.99	101.7
25.02.20	B	Control	A23	4	16.5	8.73	3.04	105
25.02.20	B	Control	A22	5	15	8.65	3.03	103.8
25.02.20	B	Control	A23	6	15.8	8.64	3.05	104.4
25.02.20	B	Control	A22	7	15.1	8.74	3.04	103.3
25.02.20	B	Treatment	A22	12	15.9	8.72	3.07	103.7
25.02.20	B	Treatment	A23	14	16.6	8.65	3.06	89.8
25.02.20	B	Treatment	A22	15	15.9	8.69	3.11	102.5
25.02.20	B	Treatment	A22	20	17.0	8.43	3.09	95.4
25.02.20	B	Treatment	A22	22	16.6	8.52	3.07	96.9

11.03.20	A	Control	488	3	15.7	7.30	3.13	2.15
11.03.20	A	Control	489	4	15.3	6.66	3.11	1.73
11.03.20	A	Control	488	5	18.1	7.88	3.12	2.19
11.03.20	A	Control	489	6	15.3	6.30	3.06	1.60
11.03.20	A	Control	488	7	15,4	6.69	3.08	1.87
11.03.20	A	Treatment	484	3	16.6	8.22	3.11	1.91
11.03.20	A	Treatment	483	4	17.5	6.89	3.11	1.61
11.03.20	A	Treatment	483	6	16.1	7.84	3.03	1.75
11.03.20	A	Treatment	484	7	17.5	7.40	3.13	1.84
11.03.20	A	Treatment	483	8	19.3	7.77	3.18	1.98
11.03.20	B	Control	A22	3	18.8	8.52	3.12	2.01
11.03.20	B	Control	A23	4	19.9	6.53	3.30	2.23
11.03.20	B	Control	A22	5	18.3	7.37	3.17	2.12
11.03.20	B	Control	A23	6	18.1	6.73	3.7	2.0
11.03.20	B	Control	A22	7	17.8	7.04	3.23	2.0
11.03.20	B	Treatment	A22	12	19.5	7.24	3.25	1.71
11.03.20	B	Treatment	A23	14	19.3	7.26	3.26	1.74
11.03.20	B	Treatment	A22	15	18.5	7.09	3.03	2.2
11.03.20	B	Treatment	A22	20	19.3	6.91	3.26	1.90
11.03.20	B	Treatment	A22	22	18.8	6.78	3.26	1.86

24.03.20	A	Control	488	3	17.1	6.55	4.13	2.0
24.03.20	A	Control	489	4	14.8	4.85	3.12	1.6
24.03.20	A	Control	488	5	18.0	7.12	5.11	1.95
24.03.20	A	Control	489	6	16.6	5.60	3.07	1.50
24.03.20	A	Control	488	7	15.8	5.85	5.20	1.96
24.03.20	A	Treatment	484	3	15.7	8.45	3.14	1.70
24.03.20	A	Treatment	483	4	19.0	6.50	3.13	1.53
24.03.20	A	Treatment	483	6	17.1	6.98	5.04	1.60
24.03.20	A	Treatment	484	7	17.2	7.75	3.10	1.73
24.03.20	A	Treatment	483	8	20.5	8.16	3.19	2.0
24.03.20	B	Control	A22	3	18.6	6.43	3.15	1.90
24.03.20	B	Control	A23	4	19.1	5.88	3.28	2.03
24.03.20	B	Control	A22	5	17.2	5.65	5.67	1.92
24.03.20	B	Control	A23	6	17.2	5.74	3.33	1.97
24.03.20	B	Control	A22	7	17.1	5.94	3.21	1.88
24.03.20	B	Treatment	A22	12	21.1	6.96	3.97	1.84
24.03.20	B	Treatment	A23	14	19.9	6.38	4.79	1.64
24.03.20	B	Treatment	A22	15	18.0	5.49	3.35	1.01
24.03.20	B	Treatment	A22	20	19.0	5.82	4.57	1.78
24.03.20	B	Treatment	A22	22	17.2	5.79	4.86	1.83

Appendix B

Anthocyanin and Phenolics results measured from absorbance data of both Vineyard A (Te Awa) and B (TK).

Vineyard	Control or Treatment	Row number	Bay number	Colour per g berry weight (mg of anthocyanins per g berry weight)	Total phenolics per mg berry weight. (absorbance units per berry)
A	HML32	484	3	2.906303094	1.769773185
A	HML32	484	3	2.94057586	1.810882153
A	HML32	483	4	2.910380047	1.761668951
A	HML32	483	4	3.004100381	2.07191198
A	HML32	483	6	2.538906371	1.636537679
A	HML32	483	6	2.702560139	1.784280664
A	HML32	484	7	2.625651999	1.55370103
A	HML32	484	7	2.292442658	1.254739325
A	HML32	483	8	2.631963813	1.569720749
A	HML32	483	8	2.709809679	1.598391607
A	CONTROL	488	3	2.750783156	1.617317133
A	CONTROL	488	3	2.924089992	1.802247093
A	CONTROL	489	4	2.755487778	1.822555
A	CONTROL	489	4	2.551835	1.6330325
A	CONTROL	488	5	2.614044977	1.600366357
A	CONTROL	488	5	2.602238462	1.573012308
A	CONTROL	489	6	2.446193543	1.582696384
A	CONTROL	489	6	2.504553106	1.693701087

A	CONTROL	488	7	2.493228198	1.52313986
A	CONTROL	488	7	2.649071822	1.5943059
B	HML32	A22	3	2.464	1.467529412
B	HML32	A22	3	2.482117647	1.381470588
B	HML32	A23	4	2.770298723	1.746743815
B	HML32	A23	4	2.8835136	1.803351254
B	HML32	A22	5	2.686534435	1.645390028
B	HML32	A22	5	2.720857383	1.620714793
B	HML32	A23	6	2.572524219	1.504406738
B	HML32	A23	6	2.718990996	1.658561678
B	HML32	A22	7	2.640977323	1.595351869
B	HML32	A22	7	2.827777561	1.739566101
B	CONTROL	A22	12	2.717042533	1.6433725
B	CONTROL	A22	12	2.878402531	1.68632495
B	CONTROL	A22	14	2.688659177	1.619911365
B	CONTROL	A22	14	2.660869419	1.610648112
B	CONTROL	A22	15	2.443153295	1.45887814
B	CONTROL	A22	15	2.474981764	1.501030607
B	CONTROL	A22	20	2.32559207	1.380097878
B	CONTROL	A22	20	2.430741076	1.486597297
B	CONTROL	A22	22	2.485415842	1.422843564
B	CONTROL	A22	22	2.410169307	1.411442574

Appendix C

Raw calculated data of spectrometry absorbance results of both vineyard A (Te Awa) and B (TK).

Vineyard	Treatment	Row number	Bay number	Absorbance 280 nm	Absorbance 520 nm
A	HML32	484	3	1.50	1.23
A	HML32	484	3	1.61	1.31
A	HML32	483	4	1.51	1.24
A	HML32	483	4	1.81	1.31
A	HML32	483	6	1.44	1.12
A	HML32	483	6	1.57	1.19
A	HML32	484	7	1.36	1.15
A	HML32	484	7	1.11	1.01
A	HML32	483	8	1.36	1.14
A	HML32	483	8	1.37	1.16
A	CONTROL	488	3	1.44	1.23
A	CONTROL	488	3	1.53	1.24
A	CONTROL	489	4	1.56	1.18
A	CONTROL	489	4	1.38	1.08
A	CONTROL	488	5	1.41	1.15
A	CONTROL	488	5	1.35	1.12
A	CONTROL	489	6	1.38	1.07
A	CONTROL	489	6	1.44	1.06
A	CONTROL	488	7	1.36	1.11
A	CONTROL	488	7	1.38	1.14
B	HML32	A22	3	1.30	1.09

B	HML32	A22	3	1.22	1.10
B	HML32	A23	4	1.51	1.20
B	HML32	A23	4	1.56	1.25
B	HML32	A22	5	1.47	1.20
B	HML32	A22	5	1.40	1.18
B	HML32	A23	6	1.33	1.14
B	HML32	A23	6	1.45	1.19
B	HML32	A22	7	1.39	1.15
B	HML32	A22	7	1.55	1.26
B	CONTROL	A22	12	1.43	1.18
B	CONTROL	A22	12	1.43	1.22
B	CONTROL	A22	14	1.40	1.16
B	CONTROL	A22	14	1.39	1.15
B	CONTROL	A22	15	1.25	1.05
B	CONTROL	A22	15	1.31	1.08
B	CONTROL	A22	20	1.19	1.01
B	CONTROL	A22	20	1.30	1.06
B	CONTROL	A22	22	1.25	1.09
B	CONTROL	A22	22	1.24	1.06

Appendix 4

VINEYARD A – TE AWA

ANOVA

ANOVA - BRIX

Cases	Sum of Squares	df	Mean Square	F	p
DATE	119.726	3	39.909	31.421	< .001
TREATMENT	18.225	1	18.225	14.349	< .001
DATE ✻ TREATMENT	0.209	3	0.070	0.055	0.983
Residuals	40.644	32	1.270		

Note. Type III Sum of Squares

Post Hoc Tests

Standard

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
11.03.20, CONTROL	13.02.20, CONTROL	3.660	0.713	5.135	< .001	***
	24.03.20, CONTROL	-0.500	0.713	-0.701	0.996	
	25.02.20, CONTROL	2.100	0.713	2.946	0.096	
	11.03.20, HML32	-1.440	0.713	-2.020	0.485	
	13.02.20, HML32	2.560	0.713	3.592	0.021	*
	24.03.20, HML32	-1.940	0.713	-2.722	0.153	

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
	25.02.20, HML32	0.680	0.713	0.954	0.978	
13.02.20, CONTROL	24.03.20, CONTROL	-4.160	0.713	-5.836	< .001	***
	25.02.20, CONTROL	-1.560	0.713	-2.189	0.385	
	11.03.20, HML32	-5.100	0.713	-7.155	< .001	***
	13.02.20, HML32	-1.100	0.713	-1.543	0.779	
	24.03.20, HML32	-5.600	0.713	-7.857	< .001	***
	25.02.20, HML32	-2.980	0.713	-4.181	0.005	**
24.03.20, CONTROL	25.02.20, CONTROL	2.600	0.713	3.648	0.019	*
	11.03.20, HML32	-0.940	0.713	-1.319	0.885	
	13.02.20, HML32	3.060	0.713	4.293	0.003	**
	24.03.20, HML32	-1.440	0.713	-2.020	0.485	
	25.02.20, HML32	1.180	0.713	1.655	0.714	
25.02.20, CONTROL	11.03.20, HML32	-3.540	0.713	-4.966	< .001	***
	13.02.20, HML32	0.460	0.713	0.645	0.998	
	24.03.20, HML32	-4.040	0.713	-5.668	< .001	***
	25.02.20, HML32	-1.420	0.713	-1.992	0.502	
11.03.20, HML32	13.02.20, HML32	4.000	0.713	5.612	< .001	***
	24.03.20, HML32	-0.500	0.713	-0.701	0.996	
	25.02.20, HML32	2.120	0.713	2.974	0.091	
13.02.20, HML32	24.03.20, HML32	-4.500	0.713	-6.313	< .001	***
	25.02.20, HML32	-1.880	0.713	-2.638	0.180	
24.03.20, HML32	25.02.20, HML32	2.620	0.713	3.676	0.017	*

Note. P-value adjusted for comparing a family of 8
* p < .05, ** p < .01, *** p < .001

- No significant difference between the brix of Control and HML32 treatments over time.

ANOVA

ANOVA - TA

Cases	Sum of Squares	df	Mean Square	F	p
DATE	337.513	3	112.504	205.896	< .001
TREATMENT	7.056	1	7.056	12.913	0.001
DATE ∗ TREATMENT	1.848	3	0.616	1.127	0.353
Residuals	17.485	32	0.546		

Note. Type III Sum of Squares

Post Hoc Tests

Standard

Post Hoc Comparisons - DATE ∗ TREATMENT

		Mean Difference	SE	t	p tukey	
11.03.20, CONTROL	13.02.20, CONTROL	-6.846	0.468	-14.644	< .001	***
	24.03.20, CONTROL	0.972	0.468	2.079	0.449	
	25.02.20, CONTROL	-2.028	0.468	-4.338	0.003	**
	11.03.20, HML32	-0.658	0.468	-1.407	0.847	
	13.02.20, HML32	-7.496	0.468	-16.034	< .001	***
	24.03.20, HML32	-0.602	0.468	-1.288	0.897	
	25.02.20, HML32	-2.506	0.468	-5.360	< .001	***
13.02.20, CONTROL	24.03.20, CONTROL	7.818	0.468	16.723	< .001	***
	25.02.20, CONTROL	4.818	0.468	10.306	< .001	***
	11.03.20, HML32	6.188	0.468	13.236	< .001	***
	13.02.20, HML32	-0.650	0.468	-1.390	0.855	
	24.03.20, HML32	6.244	0.468	13.356	< .001	***
	25.02.20, HML32	4.340	0.468	9.283	< .001	***
24.03.20, CONTROL	25.02.20, CONTROL	-3.000	0.468	-6.417	< .001	***

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
	11.03.20, HML32	-1.630	0.468	-3.487	0.028	*
	13.02.20, HML32	-8.468	0.468	-18.113	< .001	***
	24.03.20, HML32	-1.574	0.468	-3.367	0.037	*
	25.02.20, HML32	-3.478	0.468	-7.439	< .001	***
25.02.20, CONTROL	11.03.20, HML32	1.370	0.468	2.930	0.100	
	13.02.20, HML32	-5.468	0.468	-11.696	< .001	***
	24.03.20, HML32	1.426	0.468	3.050	0.077	
	25.02.20, HML32	-0.478	0.468	-1.022	0.967	
11.03.20, HML32	13.02.20, HML32	-6.838	0.468	-14.626	< .001	***
	24.03.20, HML32	0.056	0.468	0.120	1.000	
	25.02.20, HML32	-1.848	0.468	-3.953	0.008	**
13.02.20, HML32	24.03.20, HML32	6.894	0.468	14.746	< .001	***
	25.02.20, HML32	4.990	0.468	10.674	< .001	***
24.03.20, HML32	25.02.20, HML32	-1.904	0.468	-4.073	0.006	**

Note. P-value adjusted for comparing a family of 8
* p < .05, ** p < .01, *** p < .001

Harvest data collected

- There was no significant difference in data collected from 13.02.20 to 11.03.2020. However, on 24.03.2020 there was a significant difference between control TA and HML32 TA data

ANOVA

ANOVA - PH

Cases	Sum of Squares	df	Mean Square	F	p
DATE	6.195	3	2.065	9.219	< .001
TREATMENT	0.212	1	0.212	0.945	0.338
DATE ∗ TREATMENT	0.707	3	0.236	1.053	0.383
Residuals	7.168	32	0.224		

Note. Type III Sum of Squares

Post Hoc Tests

Standard

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
11.03.20, CONTROL	13.02.20, CONTROL	0.314	0.299	1.049	0.963	
	24.03.20, CONTROL	-1.026	0.299	-3.428	0.032	*
	25.02.20, CONTROL	0.138	0.299	0.461	1.000	
	11.03.20, HML32	-0.012	0.299	-0.040	1.000	
	13.02.20, HML32	0.318	0.299	1.062	0.960	
	24.03.20, HML32	-0.420	0.299	-1.403	0.849	
	25.02.20, HML32	0.122	0.299	0.408	1.000	
13.02.20, CONTROL	24.03.20, CONTROL	-1.340	0.299	-4.477	0.002	**
	25.02.20, CONTROL	-0.176	0.299	-0.588	0.999	
	11.03.20, HML32	-0.326	0.299	-1.089	0.955	
	13.02.20, HML32	0.004	0.299	0.013	1.000	
	24.03.20, HML32	-0.734	0.299	-2.452	0.252	
	25.02.20, HML32	-0.192	0.299	-0.641	0.998	
24.03.20, CONTROL	25.02.20, CONTROL	1.164	0.299	3.889	0.010	*
	11.03.20, HML32	1.014	0.299	3.387	0.035	*
	13.02.20, HML32	1.344	0.299	4.490	0.002	**
	24.03.20, HML32	0.606	0.299	2.024	0.482	
	25.02.20, HML32	1.148	0.299	3.835	0.011	*
25.02.20, CONTROL	11.03.20, HML32	-0.150	0.299	-0.501	1.000	
	13.02.20, HML32	0.180	0.299	0.601	0.999	
	24.03.20, HML32	-0.558	0.299	-1.864	0.584	
	25.02.20, HML32	-0.016	0.299	-0.053	1.000	
11.03.20, HML32	13.02.20, HML32	0.330	0.299	1.102	0.952	
	24.03.20, HML32	-0.408	0.299	-1.363	0.867	
	25.02.20, HML32	0.134	0.299	0.448	1.000	
13.02.20, HML32	24.03.20, HML32	-0.738	0.299	-2.465	0.246	
	25.02.20, HML32	-0.196	0.299	-0.655	0.998	
24.03.20, HML32	25.02.20, HML32	0.542	0.299	1.811	0.618	

Note. P-value adjusted for comparing a family of 8

* p < .05, ** p < .01

Appendix 5

VINEYARD B - TK

ANOVA

ANOVA - BRIX

Cases	Sum of Squares	df	Mean Square	F	p
DATE	161.678	3	53.893	76.214	< .001
TREATMENT	2.809	1	2.809	3.972	0.055
DATE ✻ TREATMENT	2.061	3	0.687	0.972	0.418
Residuals	22.628	32	0.707		

Note. Type III Sum of Squares

Post Hoc Tests

Standard

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
11.03.20, CONTROL	13.02.20, CONTROL	4.640	0.532	8.724	< .001	***
	24.03.20, CONTROL	0.640	0.532	1.203	0.925	
	25.02.20, CONTROL	2.860	0.532	5.378	< .001	***
	11.03.20, HML32	-0.500	0.532	-0.940	0.979	
	13.02.20, HML32	4.800	0.532	9.025	< .001	***
	24.03.20, HML32	-0.460	0.532	-0.865	0.987	
	25.02.20, HML32	2.180	0.532	4.099	0.006	**
13.02.20, CONTROL	24.03.20, CONTROL	-4.000	0.532	-7.521	< .001	***
	25.02.20, CONTROL	-1.780	0.532	-3.347	0.039	*
	11.03.20, HML32	-5.140	0.532	-9.665	< .001	***
	13.02.20, HML32	0.160	0.532	0.301	1.000	
	24.03.20, HML32	-5.100	0.532	-9.589	< .001	***
	25.02.20, HML32	-2.460	0.532	-4.625	0.001	**
24.03.20, CONTROL	25.02.20, CONTROL	2.220	0.532	4.174	0.005	**
	11.03.20, HML32	-1.140	0.532	-2.144	0.411	
	13.02.20, HML32	4.160	0.532	7.822	< .001	***
	24.03.20, HML32	-1.100	0.532	-2.068	0.455	
	25.02.20, HML32	1.540	0.532	2.896	0.107	
25.02.20, CONTROL	11.03.20, HML32	-3.360	0.532	-6.318	< .001	***
	13.02.20, HML32	1.940	0.532	3.648	0.019	*
	24.03.20, HML32	-3.320	0.532	-6.243	< .001	***
	25.02.20, HML32	-0.680	0.532	-1.279	0.900	
11.03.20, HML32	13.02.20, HML32	5.300	0.532	9.965	< .001	***
	24.03.20, HML32	0.040	0.532	0.075	1.000	
	25.02.20, HML32	2.680	0.532	5.039	< .001	***

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
13.02.20, HML32	24.03.20, HML32	-5.260	0.532	-9.890	< .001	***
	25.02.20, HML32	-2.620	0.532	-4.926	< .001	***
24.03.20, HML32	25.02.20, HML32	2.640	0.532	4.964	< .001	***

Note. P-value adjusted for comparing a family of 8

* p < .05, ** p < .01, *** p < .001

ANOVA

ANOVA - TA

Cases	Sum of Squares	df	Mean Square	F	p
DATE	258.722	3	86.241	400.098	< .001
TREATMENT	0.902	1	0.902	4.182	0.049
DATE ∗ TREATMENT	1.513	3	0.504	2.339	0.092
Residuals	6.898	32	0.216		

Note. Type III Sum of Squares

Post Hoc Tests

Standard

Post Hoc Comparisons - DATE ∗ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
11.03.20, CONTROL	13.02.20, CONTROL	-5.947	0.294	-20.253	< .001	***
	24.03.20, CONTROL	1.310	0.294	4.461	0.002	**
	25.02.20, CONTROL	-1.628	0.294	-5.544	< .001	***
	11.03.20, HML32	0.182	0.294	0.620	0.998	
	13.02.20, HML32	-5.032	0.294	-17.137	< .001	***
	24.03.20, HML32	1.150	0.294	3.916	0.009	**
	25.02.20, HML32	-1.364	0.294	-4.645	0.001	**
13.02.20, CONTROL	24.03.20, CONTROL	7.257	0.294	24.715	< .001	***
	25.02.20, CONTROL	4.319	0.294	14.709	< .001	***
	11.03.20, HML32	6.129	0.294	20.873	< .001	***
	13.02.20, HML32	0.915	0.294	3.116	0.066	
	24.03.20, HML32	7.097	0.294	24.170	< .001	***
	25.02.20, HML32	4.583	0.294	15.608	< .001	***
24.03.20, CONTROL	25.02.20, CONTROL	-2.938	0.294	-10.006	< .001	***
	11.03.20, HML32	-1.128	0.294	-3.842	0.011	*
	13.02.20, HML32	-6.342	0.294	-21.598	< .001	***
	24.03.20, HML32	-0.160	0.294	-0.545	0.999	
	25.02.20, HML32	-2.674	0.294	-9.107	< .001	***
25.02.20, CONTROL	11.03.20, HML32	1.810	0.294	6.164	< .001	***
	13.02.20, HML32	-3.404	0.294	-11.593	< .001	***
	24.03.20, HML32	2.778	0.294	9.461	< .001	***

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
	25.02.20, HML32	0.264	0.294	0.899	0.984	
11.03.20, HML32	13.02.20, HML32	-5.214	0.294	-17.757	< .001	***
	24.03.20, HML32	0.968	0.294	3.297	0.044	*
	25.02.20, HML32	-1.546	0.294	-5.265	< .001	***
13.02.20, HML32	24.03.20, HML32	6.182	0.294	21.054	< .001	***
	25.02.20, HML32	3.668	0.294	12.492	< .001	***
24.03.20, HML32	25.02.20, HML32	-2.514	0.294	-8.562	< .001	***

Note. P-value adjusted for comparing a family of 8

* p < .05, ** p < .01, *** p < .001

ANOVA

ANOVA - PH

Cases	Sum of Squares	df	Mean Square	F	p
DATE	7.639	3	2.546	12.640	< .001
TREATMENT	0.266	1	0.266	1.319	0.259
DATE ✻ TREATMENT	0.584	3	0.195	0.966	0.421
Residuals	6.446	32	0.201		

Note. Type III Sum of Squares

Post Hoc Tests

Standard

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}	
11.03.20, CONTROL	13.02.20, CONTROL	0.358	0.284	1.261	0.906	
	24.03.20, CONTROL	-0.510	0.284	-1.797	0.627	
	25.02.20, CONTROL	0.188	0.284	0.662	0.997	
	11.03.20, HML32	0.006	0.284	0.021	1.000	
	13.02.20, HML32	0.330	0.284	1.163	0.937	
	24.03.20, HML32	-1.090	0.284	-3.840	0.011	*
	25.02.20, HML32	0.138	0.284	0.486	1.000	
13.02.20, CONTROL	24.03.20, CONTROL	-0.868	0.284	-3.058	0.075	
	25.02.20, CONTROL	-0.170	0.284	-0.599	0.999	
	11.03.20, HML32	-0.352	0.284	-1.240	0.913	
	13.02.20, HML32	-0.028	0.284	-0.099	1.000	
	24.03.20, HML32	-1.448	0.284	-5.101	< .001	***
	25.02.20, HML32	-0.220	0.284	-0.775	0.993	
24.03.20, CONTROL	25.02.20, CONTROL	0.698	0.284	2.459	0.249	
	11.03.20, HML32	0.516	0.284	1.818	0.613	
	13.02.20, HML32	0.840	0.284	2.959	0.094	
	24.03.20, HML32	-0.580	0.284	-2.043	0.471	
	25.02.20, HML32	0.648	0.284	2.283	0.333	
25.02.20, CONTROL	11.03.20, HML32	-0.182	0.284	-0.641	0.998	
	13.02.20, HML32	0.142	0.284	0.500	1.000	
	24.03.20, HML32	-1.278	0.284	-4.502	0.002	**
	25.02.20, HML32	-0.050	0.284	-0.176	1.000	
11.03.20, HML32	13.02.20, HML32	0.324	0.284	1.141	0.942	
	24.03.20, HML32	-1.096	0.284	-3.861	0.011	*
	25.02.20, HML32	0.132	0.284	0.465	1.000	
13.02.20, HML32	24.03.20, HML32	-1.420	0.284	-5.002	< .001	***
	25.02.20, HML32	-0.192	0.284	-0.676	0.997	

Post Hoc Comparisons - DATE ✻ TREATMENT

		Mean Difference	SE	t	p_{tukey}
24.03.20, HML32	25.02.20, HML32	1.228	0.284	4.326	0.003**

Note. P-value adjusted for comparing a family of 8

* p < .05, ** p < .01, *** p < .001