



Technical report including return bloom measurements, for “Changing bunch architecture for sustainable botrytis control” SFF project

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Executive summary

Technical report for “Changing bunch architecture for sustainable botrytis control update” SFF project

Dion Mundy D, Agnew R, Raw V & Jia Y, January 2011, SPTS No. 5004

The SFF project aim was to investigate compounds that could be applied at flowering and change bunch shape at harvest and hence reduce the potential botrytis bunch rot risk without producing a residue in the wine. Gibberellic acids (GA) were selected for the experiment as they have been shown to change the shape of bunches. However, they are also known to produce undesired effects such as reduced return bloom. Treatments were applied to plots at flowering and assessed at harvest. Assessment included measurements of yield, bunch openness, berry volume and juice composition.

The GA3 treatment in the experiment resulted in a number of significant differences in the bunches and berries compared with the control. These changes included decreased yield, bunch weight, bunch openness and shaded area. Significant increases in soluble solids were also observed for the GA3 treatment and changes in berry volume were observed with an increase in the number of small volume berries. When return bloom was measured in November 2010, the GA3 treatment had significantly lower return bloom than the control. None of the other treatments reduced return bloom.

When photographs of GA3-treated bunches were shown at wine industry presentations, questions were asked about the number of ‘shot’ (not fully formed) berries in bunches. A high number of shot berries would not be desirable to the industry. Only the GA3 and GA4+GA7 treatments generated significant numbers of berries of this type.

None of the six treatments used reduced the disease incidence compared with the control in the experiment, and disease severity in the control was well below the penalty level normally used by wine companies, indicating that this was a ‘low disease’ year.

We would recommend that GA3 is **not** used for changing bunch architecture in New Zealand vineyards because of the negative effects on return bloom observed. In terms of experimental method for similar projects, it is recommended that flowers of similar length in the various treatments be tagged so that differences in final bunch size can be measured with greater accuracy and valid comparisons made between treatments.

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1 Introduction

Botrytis bunch rot caused by *Botrytis cinerea* is the main seasonal disease risk for grapes grown in Marlborough and many of the other wine production regions in New Zealand. Residues often occur in wine if fungicides are used late in the season for disease control.

One of the factors influencing the potential for grape bunches to develop botrytis bunch rot is bunch compactness. Hence a project was conducted to investigate methods of changing bunch compactness using products acceptable to both conventional and organic wine grape growers.

Gibberellic acids (GA) are naturally occurring plant hormones that can also be produced by fermentation from selected fungi. GA is used in table grapes to change the bunch architecture early in the season and lower the risk of late season disease, allowing fruit to be produced that meets high consumer expectations and postharvest storage requirements. Recently, wine grape producers in Germany have investigated using GA on wine grapes. Seaweed products have also been used in organic wine producing vineyards to alter bunch structure.

The aim of this project was to investigate if bunch architecture could be changed in the vineyard by the application of GA and if differences to bunch architecture increase or decrease the botrytis bunch rot disease risk under Marlborough conditions.

2 Methods and Materials

2.1 Site plan and experimental treatments

This Trial block was located on the vineyard of the Marlborough Research Centre Trust situated in Rowley Crescent, Grovetown, Marlborough. It consists of seven-year-old Sauvignon blanc (mass selected), grafted onto 101-14 rootstock. Vineyard rows were spaced 2.7 m apart with vines 1.8 m apart within the row. The vines were winter cane-pruned to carry three canes or approximately 36 nodes per vine. Canopy management consisted of vertical shoot positioning followed by summer pruning with a machine trimmer on two occasions on 08 January 2010 and 19 February 2010. One machine leaf pluck was carried out on 20 January 2010. As no shoot thinning or leaf plucking was carried out by hand, the canopy was very dense, with only 28% fruit exposure.

The experiment utilised a randomised plot design with six replicates of each of six treatments. Each treatment plot consisted of a vineyard bay (7.2 metres long) of four vines. The first half of the vine at each end of every plot was used as a buffer zone. There were six replicate plots for each treatment, laid out in a randomised complete block design. Each replicate was in a separate vineyard row.

The chemicals used are shown in Table 1 and treatments with dates of application are shown in Table 2. Treatments 2-9 had 100 ml/100 L Li700 added to the spray tank to acidify the solution. The untreated control received no growth regulators but was subjected to the same fungicide programme as other treatments.

Table 1. Chemicals used

Product	Active ingredient	Concentration of active ingredient applied	Rate
Falgro®	GA3	40 ppm	To run off
Novagib®	GA4 + GA7	40 ppm	To run off
Acadian®	Seaweed		10.1 g/10 L or 20.2 g per 10 L
Product X			15 g product/10 L
Li700	Acidifier		10 ml/10 L

For all treatments, 10 L was applied to 6 plots to provide application to run off.

Product X was supplied by a commercial company who paid fully for the inclusion of this treatment in the experiment and the other chemicals were purchased from commercial suppliers.

Table 2. Treatments, rates and timing of application

	Treatment	Growth stage	date of application
1	Untreated control		
2	Gibberellin (GA4+GA7) 40 ppm ¹ with 100 ml/100 L Li700	50% cap-fall	19/12/2009
3	Gibberellin (GA3) 40 ppm ¹ with 100 ml/100 L Li700	50% cap-fall	19/12/2009
4	Acadian® label rate	50% cap-fall	19/12/2009
5	Acadian 2 x Label rate ²	50% cap-fall	19/12/2009
6 ³	Product X	50% cap-fall	19/12/2009

¹ Higher than label rate (10 ppm, which may result in decreased flowering in apples the next season) were used as work by Trevor Lupton in Gisborne in 2009 suggested the label rate is too low to change bunch architecture under New Zealand conditions (pers. comm. Trevor Lupton Lewis Wright Valuation and Consultancy Ltd).

² Double the recommended rate per application (750 g per ha) was included, as the label suggests more than one application.

³ Product X was supplied by a commercial company who paid fully for the inclusion of this treatment.

In the early and mid season, all vines received a standard fungicide programme. For botrytis bunch rot, this comprised an application of Shirlan® on 16 December 2009 at approximately 30% capfall and an application of Switch® on 25 January at pre-bunch closure.

Treatments before and after flowering were applied using a 15-litre knapsack sprayer with a water rate of approximately 500 L/ha.

Fruit maturity was assessed on 15 April as this was the date of commercial harvest but fruit yield was measured on 28 April to allow additional botrytis bunch rot assessments because of low severity of disease at the commercial harvest date.

Botrytis

Assessments were carried out on 13, 16 and 26 April on 30 bunches per plot by assessing botrytis bunch rot incidence (% of bunches infected) and severity (% of individual bunch area infected). Severity is calculated as the average of individual % infection on each sampled bunch across the 30 of bunches sampled per plot. For that reason, it can be used as an estimate of the 'total yield loss'.

Fruit maturity at harvest

Grape berry soluble solids were measured with an Atago® temperature compensating refractometer. Titratable acidity (TA) was measured by the titration of 5 ml juice with 0.1 N NaOH to pH 8.2. Juice pH was measured by a temperature compensating pH meter.

Fruit yields

At harvest, total yields per plot and individual yield components (bunch number, average bunch weight calculated from total yield and number of bunches, berry number per bunch, berry composition) were measured by harvesting all bunches on the four treatment vines per plot.

Berry Volume

The final berry sample was collected on 27 April 2010. Five-bunch samples from each plot had individual berries removed from the rachis and the berry samples processed using a Dyostem.

The Dyostem uses a non-destructive photographic technique to count the number of berries in the sample, measure individual berry volume and measure berry colour (hue).

Meteorological details and disease risk

Environmental data (surface wetness, temperature and rainfall) were collected from the nearby Blenheim weather station. Botrytis bunch rot infection periods were identified using the Broome Botrytis bunch rot risk infection model parameters originally developed for botrytis bunch rot in grapes (Broome et al. 1995) and the software package Metwatch™ (Figure 1).

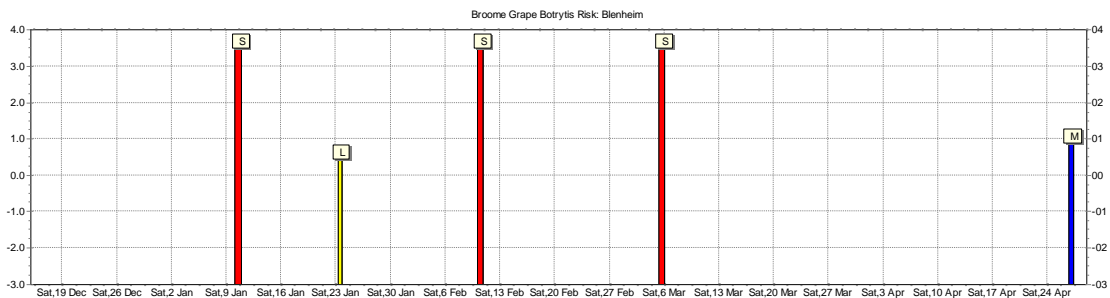


Figure 1. *Botrytis cinerea* infection periods during the 2009-2010 season for the Sauvignon blanc research site in Marlborough.

The 2009-2010 season had very low *B. cinerea* infection risk because of the lack of wetness periods during key growth stages (flowering, pre-bunch closure and from véraison to harvest). At the end of March 2010 there was almost no botrytis bunch rot in the trial area. In order to try to stimulate infection, overhead misters were set up over the whole trial area. Three artificial wetness periods were induced by the application of overhead water in early to mid-April (Figure 2).

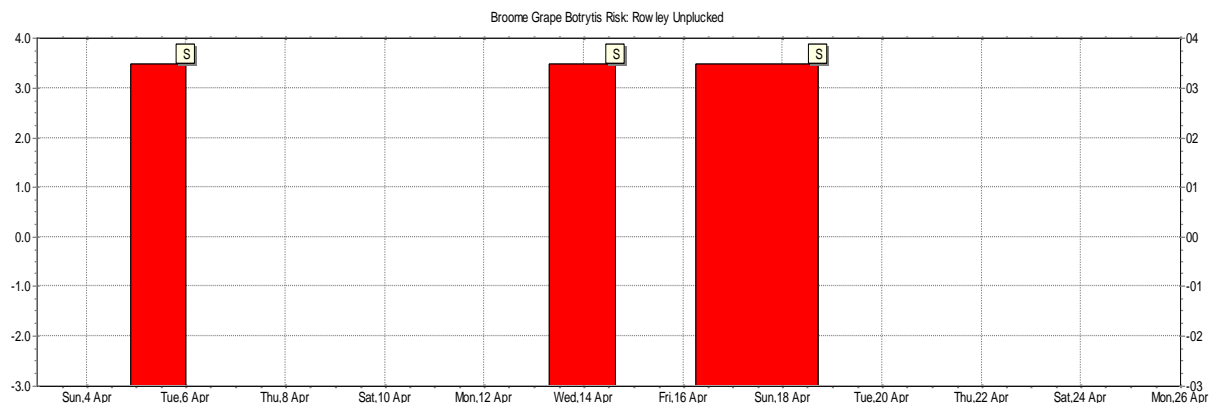


Figure 2. *Botrytis cinerea* infection periods induced by the application of overhead water to the research trial area over a period of three weeks before harvest during April 2010.

Actual and calculated volume of bunches

In order to compare the openness of bunches, the maximum width and length of the bunch was measured for five sample bunches from each plot. These values were then used to calculate a theoretical volume of the bunch based on a cone of these dimensions. Each bunch was also weighed before it was suspended in water to determine the actual volume, by displacement. The ratio of actual volume to calculated volume was determined to give a ratio that allows the comparison of bunch openness. Values that approach 1.0 are tight bunches with less space between berries. Bunches with a ratio closer to zero are more open, with more space in them.

Shaded area of the bunch

To provide another measurement of openness, five bunches from each plot were collected at harvest and photographed on the same scale. Photoshop® was used to manipulate the image to black and white, and ImageJ ®was used to calculate the area to provide an estimated shaded area of the bunch to be compared between treatments. This method does not allow comparisons of measurement between seasons or varieties, as it does not account for different length bunches, but does provide a rapid method of comparison within an experiment.

Return bloom measurement

In November 2010 the number of inflorescences (flower clusters) on each of the middle two vines of each plot was counted.

The numbers of inflorescences on the treated vines were then compared with those on the control vines. The number of canes was also recorded, as vines in the block had been pruned to three canes but individual vines may have had fewer canes, resulting in lower inflorescence numbers independently of treatment.

Statistics

Unless noted otherwise, data were not transformed before ANOVA tests were used to test the treatment effects on the response. When the ANOVA test is significant, the pairwise comparison, with significance at 5% level, using Dunnett's correction, was used to compare different treatments with the control treatment. The analyses were done using general linear model (GLM) procedure in the SAS 9.2 system.

The means of calculated cone volume were log transformed (base 10) before the analysis.

The means of volume ratio were multiplied by 100 and then log transformed (base 10) before the analysis.

For disease incidence, plot means were calculated and the data were angular transformed using $\text{asin}(\sqrt{X})$ before doing one-way ANOVA analysis.

For disease severity, plot mean percentages were calculated and then the plot mean percentages were taken, logit transformation $\text{LN}(X/(100-X))$ was taken before the one-way ANOVA analysis.

3 Results

3.1 Plot-based measurements of composition and yield

Average total yield per plot, average bunch weigh per plot, average berry weight and berry composition are shown in Table 3. The untreated control (treatment 1) had the highest average plot yield of any of the treatments at 25.4 kg/plot. Treatment 3 had significantly lower average yields at 14.8 kg/plot. While yields in none of the other treatments (2, 4, 5, 6) were significantly different from the control, all were lower than in treatment 1.

Bunches on control vines (treatment) had the highest average weight of 124.4 g. The average weights of bunches in treatment 3 (83 g) was significantly lower than the control. All other treatments had intermediate bunch weights.

Average berry weight for the 50-berry samples was not significantly different between treatments, but treatment 3 had the lowest average berry weight.

Soluble sugars (as °Brix), for berries in treatment 3 were significantly higher than the control. While soluble sugars in none of the other treatments were significantly different from the control, all were equal to or higher than the average for treatment 1.

The titratable acidity and pH of the juice were not significantly different between the treatments.

Table 3. Average total Sauvignon blanc yield per plot, average bunch weigh per plot, average berry weight and berry composition following different treatments

	Treatment	Average plot weight (kg)	Calculated bunch weight per plot (g)	Average berry weight (g)	°Brix	pH	TA
1	Untreated control	25.4	124.4	1.22	22.1	2.9	10.5
2	Gibberellin (GA4+GA7) 40 ppm ¹ with 100 ml/100 L Li700	20.0	107.5	1.24	22.6	3.0	10.4
3	Gibberellin (GA3) 40 ppm ¹ with 100 ml/100 L Li700	14.8	83.0	1.14	23.6	3.0	10.2
4	Acadian® label rate (750 g/ha) with 100 ml/100 L Li700	20.4	111.9	1.25	22.4	3.0	10.6
5	Acadian 2 x Label rate ² (1500 g/ha) with 100 ml/100 L Li700	20.8	119.8	1.21	22.4	3.0	10.6
6	Product X 150 g prod/100 L mixed with 100 ml/100 L Li700	21.2	114.9	1.23	22.1	3.0	10.5

Values in bold are significantly different at the 5% level when compared with treatment 1 using Dunnett's correction following a two-way ANOVA test.

3.2 Measurements of bunch shape

The average bunch weight, bunch dimensions, calculated volumes, volume ratios and % shaded area of a five-bunch sample from the different treatments are shown in Table 4. Bunch weight of the five-bunch sample was significantly lower for treatment 3 (GA3) than for treatment 1 (control). All other treatments had average bunch weight values that were between those of the control (148.8 g) and treatment 3 (98.4 g). The displaced volume of the bunch was also significantly lower for treatment 3 than the control. The ratio of actual volume to calculated volume was significantly lower for treatment 3, showing that these bunches were significantly more open than the control.

No significant differences were detected for the length or width of bunches, or the calculated cone volume.

Table 4. Average Sauvignon blanc bunch weight, bunch dimensions, calculated volumes, volume ratios and % shaded area of a five bunch sample for nine different treatments.

Treatment	Treatment	length in (mm)	width (mm)	bunch wt (g)	displacement wt (g)	Calculated cone	ratio actual:calc	% shaded
1	Untreated control	125.9	84.6	148.8	135.0	269.7	0.632	25.5
2	Gibberellin (GA4+GA7) 40 ppm ¹ with 100 ml/100 L Li700	124.4	82.1	124.7	110.5	251.5	0.548	24.1
3	Gibberellin (GA3) 40 ppm ¹ with 100 ml/100 L Li700	117.3	79.6	98.4	85.5	213.8	0.470	21.1
4	Acadian® label rate (750 g/ha) with 100 ml/100 L Li700	125.3	83.5	140.5	128.2	259.1	0.612	25.0
5	Acadian 2 x Label rate ² (1500 g/ha) with 100 ml/100 L Li700	127.1	84.3	146.7	133.5	264.1	0.590	25.9
6	Product X 150 g prod/100 L mixed with 100 ml/100 L Li700	144.3	81.5	135.2	121.5	268.4	0.607	24.3

Values in bold are significantly different at 5% level when compared with the treatment 1 using Dunnett's correction following a two-way ANOVA test.

3.3 Dyostem analysis of bunches

The average berry number per bunch and proportion of berries in each size class for six different treatments are shown in Table 5. Overall, treatment 3 had the lowest average number of berries per bunch and the untreated control had the highest. However, none of the differences was statistically different.

Berries were divided into 13 volume (size) classes and the percentage of berries in each class determined. Treatment 3 had significantly more berries per bunch in the smallest four volume classes (0.1, 0.35, 0.6, 0.85 ml/berry) than the control. Treatment 2 also had significantly more berries in the 0.1 and 0.35 classes than the control. Graphing of the distribution of proportions of berries in each size class (Figure 3) indicates that treatment 3 and to a lesser extent treatment 2 had a higher proportion of small berries than the other treatments.

Visual observations at véraison indicated that treatment effects had occurred (Figure 4) and that a range of bunch openness and berry sizes might be observed at harvest. Treatment 3 (Figure 5) appeared to have more small, poorly formed (or “shot”) berries than the control (Figure 6). However, at harvest bunches from these, the two most extreme treatments, were not as easily distinguished with regard to visual openness, and measurements of volume were required.

Table 5. Average Sauvignon blanc berry number per bunch and proportion of berries in each size class for six different treatments. Treatments are described in the text.

treatment	Average # berries per bunch sampled	proportion (%) of berries in each bunch with the average volume (ml) of each size class												
		0.1	0.35	0.6	0.85	1.1	1.35	1.6	1.85	2.1	2.35	2.6	2.85	3.1
1	82.5	1.1	2.4	2.4	2.7	4.7	11.8	18.8	17.0	10.3	4.4	1.9	0.7	0.2
2	73.1	3.5	5.2	3.3	3.4	4.5	7.2	11.0	11.6	9.1	4.4	2.4	0.9	0.5
3	67.8	5.2	5.4	5.9	5.5	4.4	7.1	9.4	8.6	6.5	3.9	1.3	0.9	0.2
4	82.4	0.7	2.3	2.4	3.4	6.6	12.3	17.5	15.7	8.8	4.5	2.1	0.6	0.4
5	78.3	1.2	2.8	4.0	4.4	6.2	11.2	15.6	12.4	7.8	4.0	2.0	0.7	0.2
6	75.4	1.1	2.6	3.4	4.2	4.6	8.8	13.7	13.9	10.4	5.2	2.3	1.1	0.3

Values in bold are significantly different at 5% level when compared with the treatment 1 using Dunnett's correction following a two-way ANOVA test.

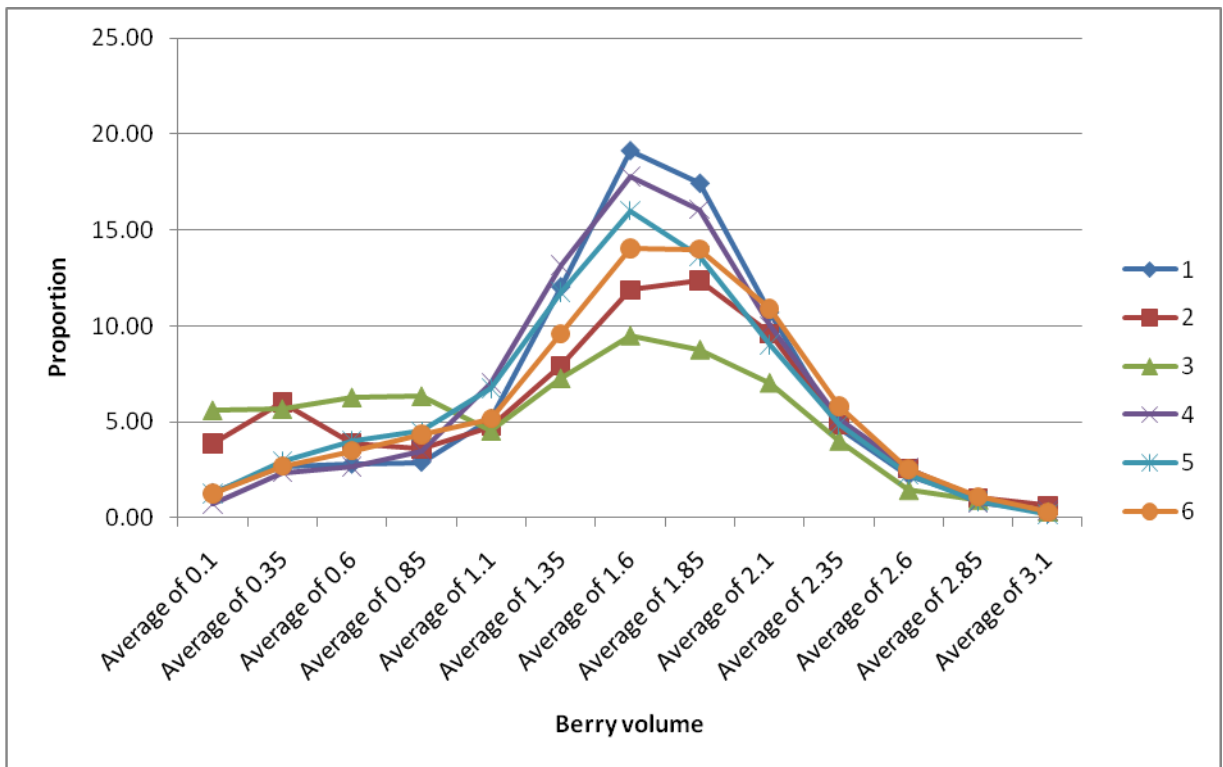


Figure 3. Distribution of proportions of Sauvignon blanc berries in each volume class for six different treatments. Treatments are described in the text.



Figure 4. Single Sauvignon blanc bunches collected from the six treatments at véraison. Treatments are described in the text.



Figure 5. Five Sauvignon blanc bunches collected at harvest from treatment 3 (GA3) showing some with very small berries.



Figure 6. Five Sauvignon blanc bunches collected at harvest from the control treatment.

3.4 Disease incidence and severity

The percentage incidence indicates the proportion of bunches that have botrytis bunch rot present, but it does not provide any information of the extent to which each bunch was affected. The highest incidence of botrytis bunch rot was observed in treatments 1 and 5 (Table 6). The lowest incidence was observed in treatments 2 and 3. The remaining treatments had intermediate incidence scores (Figure 7).

The average percentage severity (average severity/incidence (also known as crop loss) is the proportion of the crop affected by disease. This figure is used by wineries to determine whether a crop meets acceptable disease thresholds. Treatments 1, 5 and 8 had the highest severity and treatments 2 and 9 had the lowest percentages; however, no significant difference was detected (Figure 7). All severity scores were lower than the five percent threshold used by many wine companies.

3.5 Return bloom measurements

Only treatment 3, the GA3 treatment (gibberellin (GA3) 40 ppm¹ with 100 ml/100 L Li700), had significantly fewer inflorescences per vine than the control (Table 7).

Table 6. Percentage incidence and severity of botrytis bunch rot in Sauvignon blanc at harvest 2010. Treatments are described in the text.

Treatment	% incidence	% severity
1	19.4	1.6
2	10.0	0.6
3	11.1	1.0
4	16.7	1.4
5	21.1	1.7
6	16.1	1.3

No significance differences between treatments in either disease incidence or severity were detected.

Table 7. Mean number of Sauvignon blanc inflorescences per vine the season following the application of treatments. Treatments are described in the text.

Treatment	Inflorescences per vine
1	47.5
2	51.9
3	28.4
4	43.5
5	52.8
6	47.5

Values in bold are significantly different at the 5% level when compared with treatment 1 using Dunnett's correction following an ANOVA test.

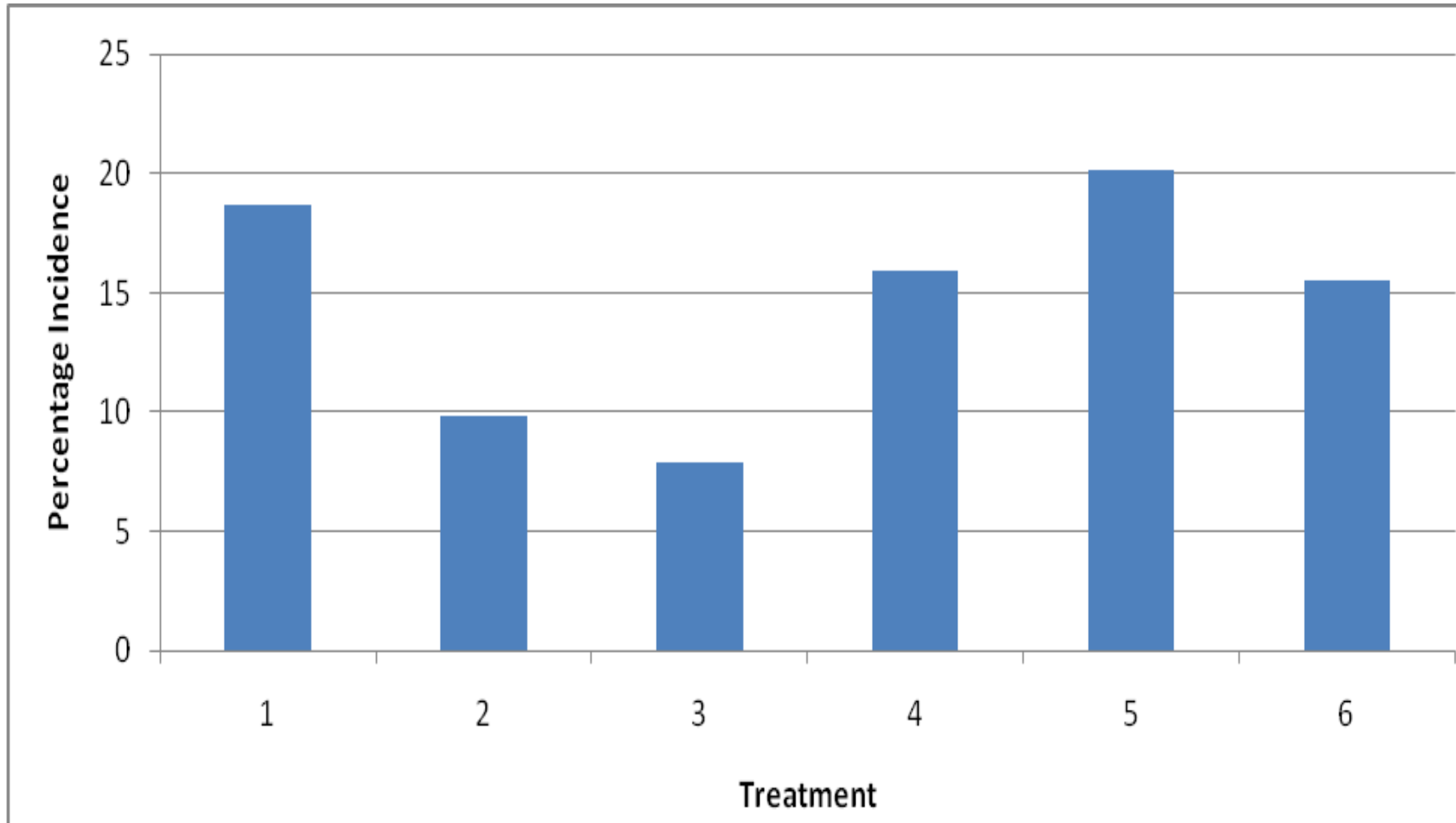


Figure 7. Percentage incidence of botrytis bunch rot in Sauvignon blanc on 26 April 2010. Treatments are described in the text.

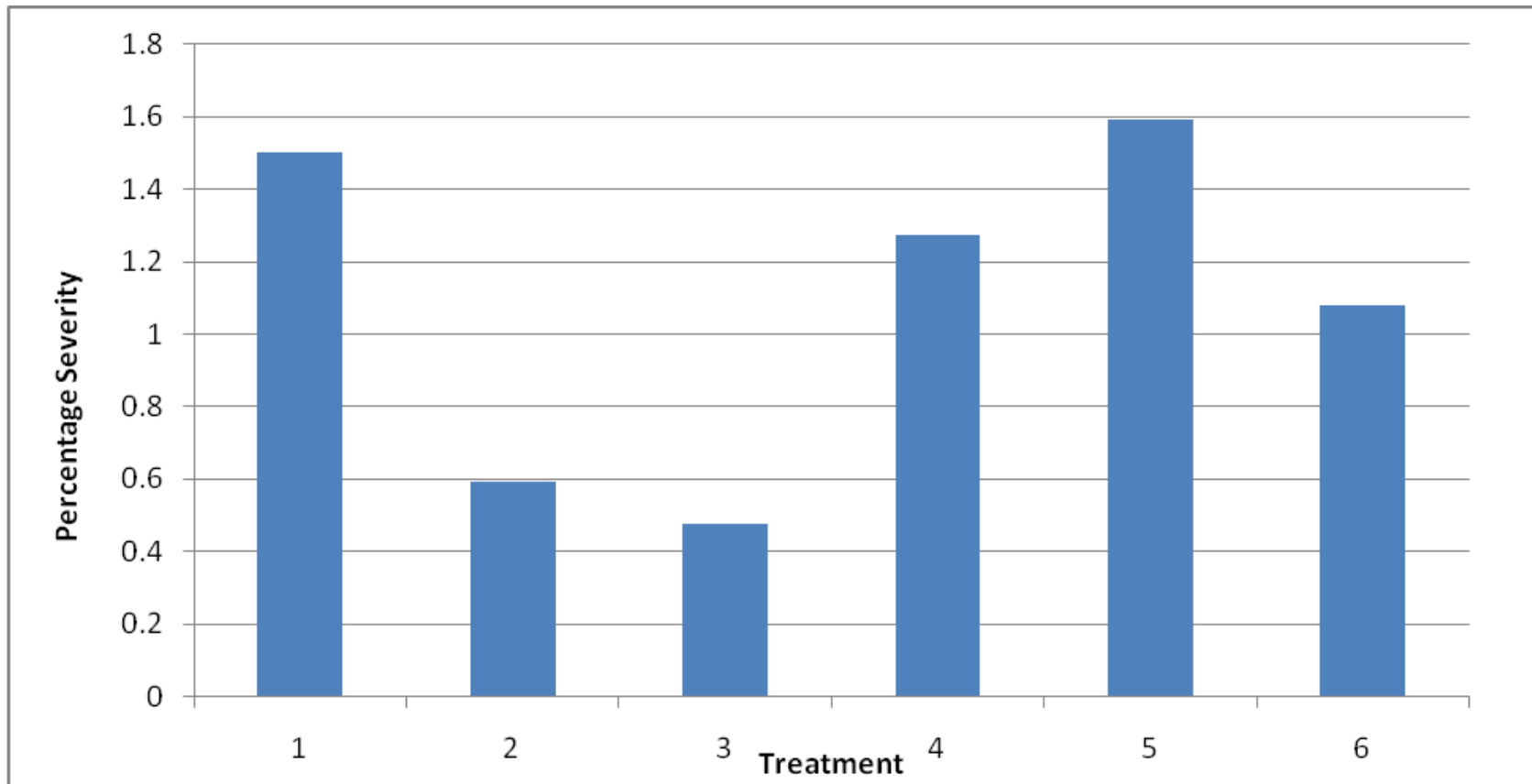


Figure 7. Average percentage severity of botrytis bunch rot in Sauvignon blanc on 26 April 2010. Treatments are described in the text.

4 Discussion

1. This was a preliminary study to determine whether it was possible to change bunch shape by the application of hormones or growth-altering compounds.
2. Some of the rates used were significantly higher than label rates to try to demonstrate an experimental effect.
3. The high GA3 rate used inhibited flowering in the following season; therefore, the use of this compound at these rates is not recommended.
4. In this experiment, variability in bunch architecture of the untreated controls made it difficult to detect clear effects of the experimental treatments. Nevertheless, some consistent treatment effects were observed.

Treatment 3 (Gibberellin (GA3) 40 ppm^l with 100 ml/100 L Li700) was the treatment that consistently had significantly different bunches (bunch weight, openness, proportion of small volume berries) from the untreated control. However, at the rates used, this treatment reduced return bloom in the 2010/2011. The degree of reduction in yield in the current season (equivalent to 5.1 tonnes per hectare) resulting from that treatment may not be acceptable to growers.

The higher total soluble solids of fruit following this treatment are likely to be the result of the lower total yield per vine. While treatment 3 had the lowest bunch weight and a significantly more open bunch than the control, the size distribution of berries indicated that these bunches contained a large number of very small berries. The treatment had the lowest incidence and severity of disease of all the treatments, which was consistent with our theory that bunches that are more open will have less disease pressure. However, the low disease incidence and severity of disease in this experiment meant that no significant differences between treatment effects could be detected. While GA3 produced the most marked changes to bunch architecture, many of the other treatments, while not statistically significant, produced results intermediate between those of control and treatment 3 without return bloom problems. These treatments may offer some possibility of altering bunch architecture and associated disease without the crop loss and return bloom issues associated with the GA3 treatment.

In terms of experimental method, it is recommended that flowers of similar length in the various treatments be tagged so that differences in final bunch size can be measured with greater accuracy and valid comparisons made between treatments.

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Within the project, the assistance of the other staff at the Marlborough Wine Research Centre during harvest is also acknowledged.

We would also like to thank Robert Lamberts, Plant & Food Research for his work on the shaded-area photographs.