# Options for Growth Control of Male Vines for GREEN Orchards

# Final Report on Zespri Innovation Project VI 1390

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## Executive summary

The scope of this project was to evaluate a range of novel vine management techniques deemed to have potential to reduce male vine vigour and thereby reduce the need for pruning cuts. Conventional male vine management requires significant post-flowering pruning. This vine wounding is likely to be an important factor contributing to male vine susceptibility to infection by Psa-v during the growing season.

A commercial male-only pollen orchard comprising mature vines of the cultivar 'Chieftain', was used to evaluate several alternative growth control strategies. Thirteen treatment options were considered based on input from PlusGroup, Zespri, and Plant & Food Research. Several approaches were tested, including injection of plant growth regulators (PGRs), root pruning following flowering, extended trunk girdling, and soil application of the elicitor Actigard™ with or without root pruning.

Plant growth regulators (PGRs), with a mode of action involving modification of natural gibberellic acid activity, have been used in many fruit crops (e.g. apples, pears, avocado) to significantly control shoot extension growth. The commercially available PGRs B-Nine®SG (daminozide), Regalis® (prohexadione-calcium), and Sunny® (Uniconazole-P) were evaluated in a fully replicated trial. The PGR Moddus® (trinexapac-ethyl) was also evaluated in a pilot study on vines growing adjacent to the main trial. PGRs were applied to vines by "drip feed" thereby eliminating the need to spray vines.

The PGR Actigard™ is a plant activator or elicitor that contains the active component acibenzolar-S-methyl. This compound stimulates the natural defence system in plants via induced systemic resistance (ISR), and has shown promise in the control of Psa. Actigard™ has also been observed to have growth control properties on kiwifruit vines.

Uptake of B-Nine®SG, Regalis®, and Moddus® via injection into the trunk xylem was an effective way of introduction to Chieftain vines. Uptake of Sunny® by this method was much less effective and is not recommended. Moddus® at the concentration used in the associated pilot study resulted in some leaf toxicity.

Growth control treatments including B-Nine®SG, Sunny® (medium conc.), extended trunk girdling, root pruning and Actigard™ plus root pruning significantly reduced vine vigour. The growth retardation effects translated into large reductions in winter pruning requirements. For the B-Nine®SG, extended trunk girdling, and Actigard™ plus root pruning treatments, reductions in winter pruning weights per vine were 83, 58, and 53% respectively. Reductions in vine vigour were principally from reduced proportions of vigorous non-terminating shoots (> 1 m long).

Flower numbers per unit canopy area were more than doubled by the application of B-Nine®SG. However, while several treatments had beneficial effects on vine vigour there was evidence that normal flower development leading to high pollen viability was impaired. Reductions in pollen viability of 46% for vines treated with B-Nine®SG and 30% for vines treated with Moddus® were apparent. Even for the vines treated with an extended trunk girdle, pollen viability dropped by 15%. Positive effects of vigour control therefore need to be balanced against reductions in pollen viability.

Actigard™ application via soil drench at a high rate (equivalent to 400g product per canopy ha. x 4 applications) did not reduce vine vigour whereas vines treated at the lower rate (200 g x 4) did show some evidence of reduced vigour, a result consistent with grower observations. Incidence of secondary symptoms of Psa-v within the trial area was restricted to one vine only. Analysis of effects of Actigard™ on vine health was not therefore possible as infection was very low. Disease expression across all treatments was low, most likely because climatic conditions across the duration of the trial were mostly not favourable for infection.

Both treatments involving root pruning resulted in significant reductions in vine vigour without loss of pollen viability. This orchard management technique is relatively simple to apply and is now commonly used on female vines to enhance fruit dry matter content. It is recommended that further research into the technique for male vines be considered, particularly timing (pre- and post-flowering) and opportunities to severe higher proportions of root biomass.

It should be noted that the use of synthetic PGR's such as those tested here is not permitted in female vines. The use of injection and soil treatments were used here to minimise the risk of drift onto female vines, if practices were to be used on a commercial producing block. Application of any agrichemical to female vines via trunk injection is not recommended due to the risk of residues.

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## 1. Introduction

The scope of this project was to evaluate a range of novel vine management techniques deemed to have potential to reduce male vine vigour and thereby reduce the need for pruning cuts. Conventional male vine management commonly requires significant post-flowering pruning. This vine wounding may contribute to male vine susceptibility to infection by Psa-v during the growing season, either by increasing the number of wound surfaces or by increasing the quantity of new shoot growth following pruning after flowering.

A commercial male-only pollen orchard comprising mature vines of the cultivar Chieftain, was used to evaluate several alternative growth control strategies. Thirteen treatment options were considered based on input from PlusGroup, Zespri, and Plant & Food Research. Several approaches were tested, including injection of plant growth regulators (PGRs), root pruning following flowering, extended trunk girdling, and soil application of the elicitor Actigard™ with or without root pruning.

Plant growth regulators (PGRs), with a mode of action involving modification of natural gibberellic acid activity, have been used in many fruit crops (e.g. apples, pears, avocado) to significantly control shoot extension growth. The commercially available PGRs B-Nine®SG (daminozide), Regalis® (prohexadione-calcium), and Sunny® (Uniconazole-P) were chosen for evaluation in a replicated trial. The PGR Moddus® (trinexapac-ethyl) was also considered in a pilot study on vines growing adjacent to the main trial. PGRs were applied to vines by "drip feed" thereby eliminating the need to spray vines. This technique was previously used to introduce PGRs into vines of 'Sparkler' and 'Meteor' (Patterson *et al* 2013a).

It should be noted however, that the use of synthetic PGR's such as those tested on male vines in this trial *is not permitted in female vines*. The use of injection and soil treatments were used here to minimise the risk of drift onto female vines, if practices were to be used on a commercial producing block. Application of any agrichemical to female vines via trunk injection is not recommended due to the risk of residues.

The PGR Actigard™ is a plant activator or elicitor that contains the active component acibenzolar-S-methyl. This compound is a functional analogue of the naturally occurring salicylic acid. It stimulates the natural defence system in plants via induced systemic resistance (ISR), and shows promise in the control of Psa-v. There have also been several observations by growers that it has growth retardant properties (smaller leaves) on kiwifruit vines. It may therefore be of additional benefit in the management of male vines. An application path via soil drench at a standard rate and a double rate was considered in the project.

Can root pruning be used to reduce vigour of male vines? Considerable experience has been gained with root pruning of kiwifruit as a technique to manipulate carbohydrate allocation to fruit and thereby increase fruit dry matter and its taste profile (Patterson *et al* 2009a, 2009b, 2011, 2013b, Pentreath *et al* 2013). Root pruning is also used for crops such as apple and pear in high density orchards in Europe and USA to control shoot growth and increase flowering. For pipfruit orchards, the timing of application is generally in late winter or early spring and significant reductions in tree vigour have been demonstrated (Ferree *et al* 1992).

For this project, root pruning just after flowering is considered. Actigard® in combination with root pruning was also included. For this treatment, Actigard™ applied one week earlier than root pruning, was a way of potentially stimulating a plant response prior to the pruning treatment.

Can extended trunk girdling be used to control male vine vigour? Summer trunk girdling is commonly applied to female vines to enhance dry matter allocation to fruit and girdles normally heal over in about 3-4 weeks. Girdles that are open for longer periods however, (extended trunk girdling) can have effects on vine vigour. When girdles remain open into the autumn/winter period, they cause severe reduction in vine vigour (Barnett *pers comm.*) Effects of extended trunk girdles on male vines has not been extensively researched but the technique does offer another potential way of limiting male vine vigour.

## 2. Methodology

### a. Plant material

A commercial male-only pollen orchard (cultivar Chieftain) located in Omanawa, Western Bay of Plenty, was used to evaluate several growth control strategies. Vines were planted at a spacing of 5 m within rows and 3.6 m between rows, with a canopy framework of main leaders orientated in a North-South direction and sub-leaders in an East-West direction.

At the time of setting up the evaluation, all vines in the trial area were Psa-v free. However, Psa-v secondary symptoms were evident in some vines within the block eastward of the trial area.

Standard vine management was used within the block to optimise flower production, with an emphasis on short shoots and spurs and a relatively open and flat canopy structure compared with conventional strip male vines. Contractors pruned vines in the trial according to this orchard's normal management protocol. This included one prune c. 3 weeks after the finish of flowering, and an annual prune in winter.

### b. Treatments and experimental design

Thirteen treatments including control were compared in a fully replicated randomised block design (Table 1). Each treatment consisted of nine vines (3 blocks of 3 vines per block).

Three Chieftain vines adjacent to the main trial, on the eastern side, were used for a pilot study into effects of the PGR Moddus®.

**Table 1 Summary of growth control treatments compared**

| | Treatment |
|---|---|
| 1 | Control – standard vine management |
| 2 | B-Nine®SG 7 g/L |
| 3 | Regalis® 1.5 g/L (Regalis® low) |
| 4 | Regalis® 3.0 g/L (Regalis® medium) |
| 5 | Regalis® 4.5 g/L (Regalis® high) |
| 6 | Sunny® 5 ml/L (Sunny® low) |
| 7 | Sunny® 10 ml/L (Sunny® medium) |
| 8 | Sunny® 15 ml/L (Sunny® high) |
| 9 | Root prune |
| 10 | Extended trunk girdle |
| 11 | Actigard™ drench x 4 @ 200 g per ha |
| 12 | Actigard™ drench x 4 @ 400 g per ha |
| 13 | Actigard™ drench x 1 @ 200 g per ha then root prune |
| Pilot | Moddus® 4 ml/L |

### c. Plant Growth Regulators (PGRs)

The product B-Nine®SG (NuFarm, NZ), contains the active ingredient daminozide at 850 g/kg. Vines were treated with one litre of B-Nine®SG at a concentration of 7 g product per litre in distilled water. This treatment was tested as a positive 'control'.

The product Regalis® (BASF, NZ), contains the active ingredient prohexadione-calcium at 100 g/kg. Vines were treated with either low, medium or high concentrations;

- Low = 1L of Regalis® at a concentration of 1.5 g product/L in distilled water
- Medium = 1L of Regalis® at a concentration of 3.0 g product/L in distilled water
- High = 1L of Regalis® at a concentration of 4.5 g product/L in distilled water

The product Sunny® (Sumitomo Chemical Co., GroChem NZ), contains the active ingredient uniconazole-P, at 50 g/L. Vines were treated with either low, medium, or high concentrations;

- Low = 1L of Sunny® at a concentration of 5 ml product/L in distilled water
- Medium = 1L of Sunny® at a concentration of 10 ml product/L in distilled water
- High = 1L of Sunny® at a concentration of 15 ml product/L in distilled water

### d. Actigard™ application

The product Actigard™ (Syngenta Crop Protection, NZ) contains the active ingredient acibenzolar-S-methyl, at 50 g/100 g. Actigard™ was applied to treated vines 4 times at 3-4 week intervals as a soil drench, beginning on 14 December 2012. Rates were equivalent to either 200 or 400 g product per canopy ha. For each application, Actigard™ was made up fresh in distilled water. Each application consisted of 10 litres of product per vine, applied by watering can to the soil surface of the complete herbicide strip under each treated vine (c. 2 x 5 m). Actual product amounts applied were;

- 200 g product per canopy ha. = 0.036 g product per litre of drench x 10 L per vine
- 400 g product per canopy ha. = 0.072 g product per litre of drench x 10 L per vine

### e. Actigard™ soil drench plus root prune

A single drench of Actigard ™ at a rate equivalent to 200 g product per canopy ha. (0.036 product per litre x 10 L – applied as above) was used to treat vines on 14 December 2012, one week before a standard root prune treatment (see below) was carried out.

### f. Injection system

A drip feed system to introduce PGRs into the sapwood (xylem) tissue of vine trunks was developed during preliminary trials at Te Puke Research Centre, and was previously used to introduce chemicals into mature vines of Sparkler and Meteor (Patterson *et al* 2013a). Chemicals were made up fresh in distilled water in 1-L plastic containers (Figure 1) to which was attached an 80-cm length of 4 mm internal diameter clear polythene tubing. This was terminated with a tee junction (4 mm barbed tees, Pope Irrigation), with two 20 cm long feeder tubes, which were then attached to two barbs (4 mm barbed connectors, Pope Irrigation) inserted into each of the vine trunks.

At the position where the holes were drilled for the injection barbs, a 1 cm x 1 cm square of bark tissue was cut with a sharp knife and carefully removed (Figure 2). This exposed the white/cream sapwood, which was then carefully drilled with a new sharp 4.6 mm diameter drill bit, slightly offset from the radial axis of the trunk to ensure the hole was drilled into sapwood and not heartwood nor directly into the pith in the centre of the trunk. The hole was cut with the drill on a high-speed setting and the drill bit removed all wood shavings cleanly. Holes were drilled to a depth of c. 2-3 cm either side of the trunk.

The irrigation barbs were gently tapped into the holes such that the flange of the barb sat flat on the wood (Figure 2). Provided the hole was neatly cut, the barb was held in place sufficiently to attach the plastic tubing without pulling it out.

Chemicals were made up fresh on the day of application and were introduced into vines over the period 1000 – 1500 h on 13 and 14 December 2012. Weather conditions were warm but somewhat overcast during the application times. Vines would have been actively transpiring during application of the chemicals.

B-Nine®SG and Regalis® were taken up via the xylem as expected. Uptake of Sunny®, however, was somewhat problematic and appeared to cease after c. 2-3 hours. A decision was made to apply remaining volumes of product to vines as a crown root zone drench. The volumes taken up via the xylem stream and volumes applied to the crown root zone were recorded (Table 2).

**Figure 1 Detail of the gravity feed drip system used to apply growth regulators to male vines. The 1L plastic container was attached to a support wire during infusion. Two ports were used to introduce the liquid to the sapwood xylem tissue**

**Figure 2 Detail of irrigation barb which was gently tapped into the sapwood following removal of a piece of bark cut slightly larger than the 1 cm x 1cm flange on the irrigation pipe joiner**

**Table 2 Volumes of Sunny® taken up via the xylem stream compared with volumes applied to the crown root zone**

| Treatment | Volume (ml) taken up via xylem (mean ± SD) | Volume (ml) applied to crown root zone (mean ± SD) |
|---|---|---|
| Sunny® 5 ml per litre | 39.8 ± 0.18 | 60.2 ± 0.18 |
| Sunny® 10 ml per litre | 20.6 ± 0.21 | 79.4 ± 0.19 |
| Sunny® 15 ml per litre | 5.1 ± 0.05 | 94.9 ± 0.06 |

### g. Root pruning

Root pruning was carried out with the root pruner built by R. and P. West, Te Puke (Figure 3). This pruner was used for previous root pruning research trials (Pentreath et al 2013, Patterson et al 2009a). Pruning was carried out on 13 December 2012 to a depth of c. 40 cm on both sides of each root-pruned vine at a distance of c. 70 cm out from the centre line of the vine row. Pruning was carried out for the full length of each treated vine (i.e. 5 m on each side of a treated vine). As with previous root pruning trials there was some disturbance of the soil during pruning, although the roller on the back of the pruner minimised this. Occasional pieces of root were brought to the surface. These were trimmed off and any exposed roots were buried. The soil was flattened back down with the tractor tyres.

**Figure 3 Root pruner designed and constructed by Russell West**

### h. Extended trunk girdling

Extended trunk girdles were used to significantly interrupt carbohydrate and hormone flow from the vine top to the lower trunk and root tissues. Research at Te Puke Research Centre has previously shown that extended trunk girdling significantly reduces vine vigour of Hort16A vines (Barnett *pers comm*.). Wide (9 mm) girdles were applied to treatment vines on 17 December 2012 using a girdling knife. These were kept open for the remainder of the growing season.

### i. Pilot evaluation of Moddus® PGR

A small sample of the PGR Moddus® (Syngenta, UK) was provided for evaluation just as the main trial was being set up. The product Moddus® contains the active ingredient trinexapac-ethyl at 250 g/L. A small pilot study using three vines adjacent to the main trial was set up to provide a preliminary evaluation of Moddus® effects on vine vigour. Moddus® was introduced to these vines via the same drip feed system used for the PGRs in the main trial. Moddus® was applied to vines at a concentration of 4 ml/L (1L per vine). Moddus® was effectively taken up via the xylem at rates comparable with B-Nine® (c. 2-3 hours). Data for these vines were obtained for comparative purposes but *should not be interpreted as being part of the main replicated trial.*

### j. Shoot growth assessment

For each treatment vine, two vegetative shoots arising from the main leader (1 in the north sector and 1 in the south sector) were tagged on January 8, 2013. Shoot lengths were measured immediately and again on February 7, March 12, and May 5. Some losses of shoots due to breakages occurred during the growing season.

During July 2013, these shoots were removed from the vines for determination of internode length. Canes were scored as terminated or not.

#### k. Winter pruning assessment

Vines were pruned as per normal orchard practice by a contract pruning gang during 26-28 July 2013. Non-terminating canes were cut back to 30-80 cm while spurs and short shoots were generally left intact. Prunings were collected for each treatment vine and weighed at the time of pruning. It was not practical to record the time required to prune each vine in the trial. Pruning weights were therefore used to give an indication of treatment effects on the amount of unwanted vegetative growth over the growing season, and thus a good indication of vine vigour and requirement for pruning cuts.

#### l. Shoot type assessment

Vines were assessed after winter pruning for treatment effects on the ratio of spurs and short shoots to large non-terminating shoots. Two long sub-leaders per vine were selected near the centre crown, one from each side of the main North-South leaders. All shoots arising from these sub-leaders were counted and recorded as either “terminated” or “cut” meaning that the shoot had been fast-growing, non-terminating and cut back to 30-80cm at pruning time.

#### m. Carry over effects on vegetative growth

Vines of several treatments continued to show vigour reductions during the 2013-2014 growing season. Vines of those treatments showing obvious vigour reduction (visual assessment) were scored for proportions of shoot types. Two sub-leaders were chosen on each vine, one each side (East & West) near to the trunk. Current season’s shoots were counted on each sub-leader and assigned to one of four categories according to length: 0-20cm, 20-60cm, 60-100cm and >100cm. Assessment were made for control vines and treatments B-Nine®@7g/L, Sunny®@10ml/L and Actigard+RootPrune.

#### n. Flowering assessment

Flower numbers in 1 m x 1m quadrats were assessed during flowering in November 2013. Unfortunately it was only possible to carry out limited flower counting. Results provide a guide to treatment effects only.

#### o. Psa-v incidence

All vines within the trial were visually checked for Psa-v incidence (primary and secondary symptoms) at the beginning of September 2013. A canker on one of the vines with an extended trunk girdle was confirmed as Psa-V positive by VLS and referred to KVH for registration. No other signs of the disease were observed at this time. Vines were again assessed in March 2014. No additional vines were showing signs of Psa infection. The vine showing secondary infection in 2013 was flagged for removal.

#### p. Metabolite sampling

At the request of Zespri, leaf samples were collected from root pruned vines at 1 week (20th Dec) and 3 weeks (8th Jan) post-root pruning date. Leaves were snap frozen in liquid nitrogen immediately after removal from the vine. Samples were then double bagged, labelled and stored at -80C at Plant and Food Research, Te Puke Research Centre.

### q. Pollen viability

During flowering in 2013, 100 flowers were collected from vines of each treatment (bulk sample). Anthers were removed and dried and pollen was then separated and stored frozen until evaluated.

Prior to viability assessment, three 20-30 mg samples of pollen were rehydrated for 2 hr at 22-25°C. Each sample was then germinated in a standard sucrose/boric acid medium for 2 hr at 22-25°C.

Aliquots were then removed and placed on a microscope slide to assess germination percentage. A total of at least 300 pollen grains were then counted, and the proportion that had a pollen tube longer than the diameter of the pollen grain was recorded.

### r. Experimental timeline

November 2012: flowering
December 2012: application of treatments
January – May 2013: shoot growth assessments
July 2013: assessment of winter pruning weights
November 2013: flowering and pollen viability assessment
March 2014: assessments of shoot types

## 3. Results and Discussion

### a. Application of PGRs

Application of B-Nine®SG, Regalis®, and Moddus® (pilot study) via the drip feed system was successful, although for Regalis® particularly at the high concentration there appeared to be some settling out of product in the containers and tubes. It was necessary to agitate the liquid occasionally to re-suspend the product. The product is technically described as "water dispersable" and for standard spray application "tank agitation" is recommended. It is possible therefore that the active ingredient does not completely dissolve and this may be related to its lack of effectiveness. In studies with spray application to female vines it was shown to be very effective at controlling shoot extension, albeit with several applications (P Martin pers comm.).

Application of Sunny® was more problematic. Under the environmental conditions, which were quite suitable for good xylem transport and uptake of B-Nine®SG and Moddus® into vines, there appeared to be significant restriction with uptake of Sunny®. The physiological/chemical reasons for this impairment are unknown. It is possible that the product is sequestered by the xylem or somehow induces blockage of xylem vessels in the sapwood. This impairment appeared to be concentration dependent. Sunny® has also been shown in other studies to control shoot growth of female vines when applied as a foliar spray, so its effectiveness on kiwifruit is not in doubt. No phytotoxicity was seen on any of the vines treated with Sunny®.

Application of Moddus® to vines was a pilot study. Ideally, it would have been prudent to carry out an evaluation of several concentrations, but only a limited quantity of product was available to test. Soon after application, leaf growth appeared to be somewhat abnormal with cupping of leaves and some toxicity of shoots. Such effects point to excess concentration in tissues and phytotoxic effects. Further work should examine lower concentrations than the 4 ml per L.

### b. Effects on shoot growth

Shoot growth was measured over the January to May 2013 period. Some inconsistencies were apparent in the shoot length data, possibly caused by measuring or transcription errors. Obvious data errors were set aside before the growth analysis was carried out.

Statistical analysis did not indicate any significant treatment effect on mean shoot length for the 8th January, 7th February, 12th March, or 5th May assessments. There was considerable variation among vines in average shoot length and for vines of the Actigard™ high drench treatment canes appeared noticeably longer when observed on 5th May. Mean shoot length for these vines was 109 cm compared with a mean shoot length of 42 cm for the control vines. The difference, however, was not statistically significant.

Shoot growth data for the four assessment dates are summarised in Figures 4 and 5. For clarity the treatments are divided into two groups for comparison.

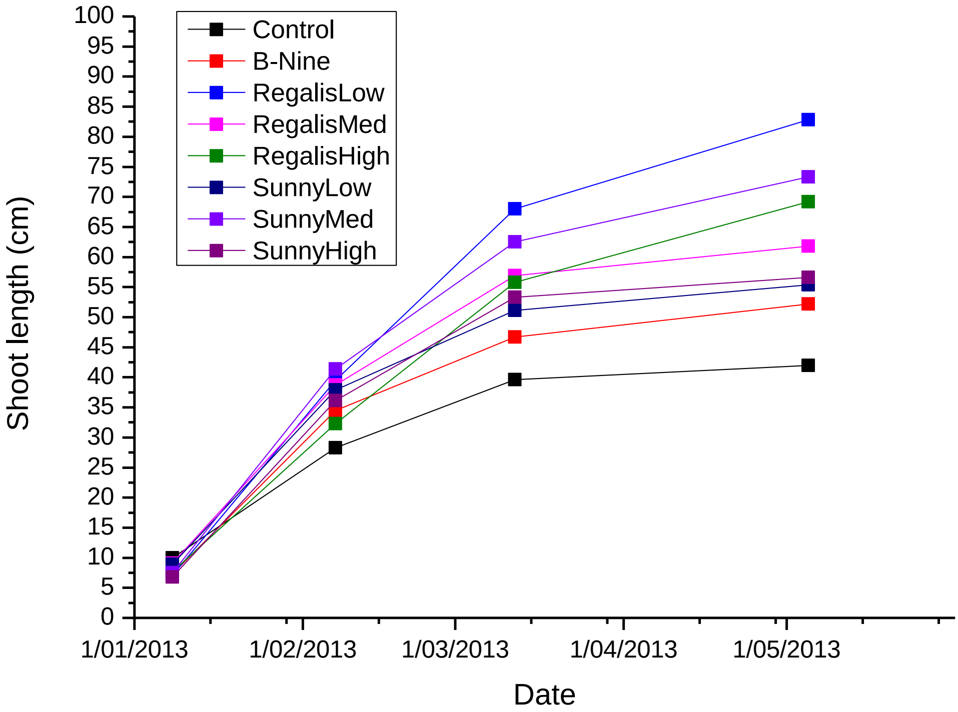


**Figure 4 Effects of PGRs B-Nine®SG, Regalis® and Sunny® on shoot extension growth of Chieftain. Each data point is the mean for nine vines. There was no significant difference between treatments on any of the measurement dates at the 5% level**

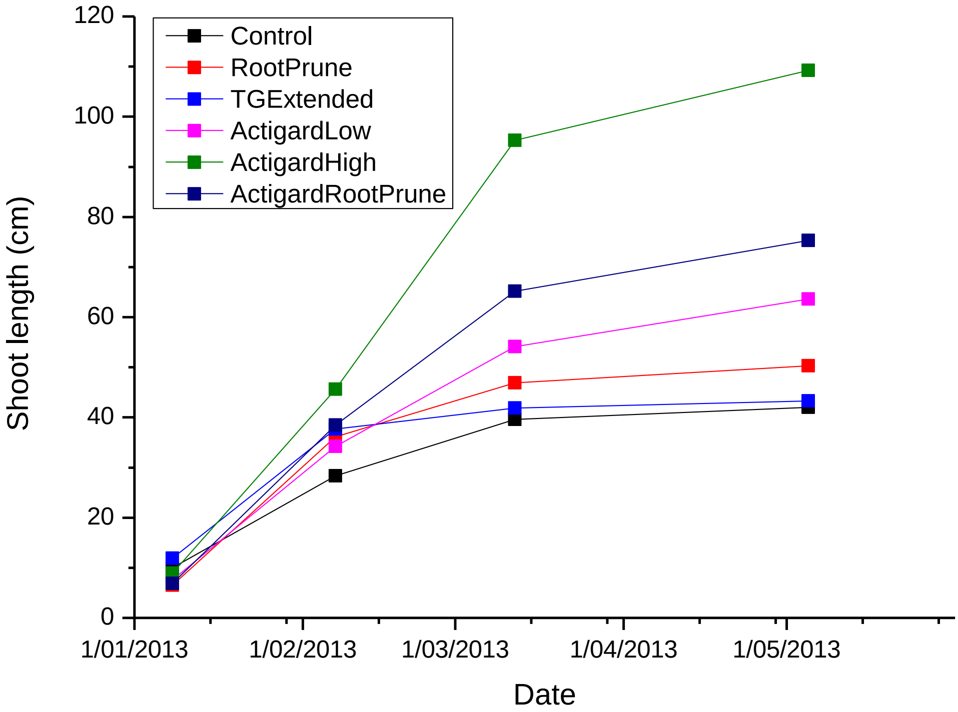


**Figure 5 Effects of root pruning, extended trunk girdling, and Actigard™ on shoot extension growth of Chieftain. Each data point is the mean for nine vines. There was no significant difference between treatments on any of the measurement dates at the 5% level**

Further analysis of the shoots used for length measurements indicated that the proportion of terminated shoots was high (80-100%) except for the shoots obtained from vines treated with the Actigard™ at the high rate. (Table 3). A much lower proportion (67%) of the measured shoots from these vines had terminated. Differences in mean internode lengths were small except for the shoots from the vines treated with the high rate of Actigard™ which had the longest internode lengths.

**Table 3 Effects of vine vigour control treatments on proportions of terminated shoots and internode lengths (n = 16-18 per treatment)**

| Treatment | Shoot attribute | |
|---|---|---|
| | Proportion terminated (%) | Mean internode length (mm) |
| Control | 89 | 2.8 |
| B-Nine®SG | 100 | 2.7 |
| Regalis® Low | 89 | 3.8 |
| Regalis® Med | 88 | 3.0 |
| Regalis® High | 78 | 3.2 |
| Sunny® Low | 94 | 3.0 |
| Sunny® Med | 82 | 3.2 |
| Sunny® High | 78 | 3.4 |
| Root prune | 83 | 3.4 |
| Trunk girdle extended | 94 | 3.1 |
| Actigard™ Low | 88 | 3.3 |
| Actigard™ High | 67 | 4 |
| Actigard plus root prune | 89 | 3.3 |

### c. Effects of treatments on winter pruning

Analysis of pruning weights removed from vines indicated that treatments involving B-Nine®SG, Sunny®Low, Sunny®Med, Sunny®High, extended trunk girdling, and root pruning (with and without Actigard™) all resulted in significantly less pruning material being removed from vines during the 2013 winter pruning (Figure 6). For the vines treated with B-Nine®SG the reduction was substantial (83%). Large reductions were also apparent for the extended trunk girdling treatment (58%) and root pruning alone (38%) or root pruning with Actigard™ (53%). For the Sunny® treatments there were also a significant reductions (Sunny®Low = 29%, Sunny®Med = 54%, Sunny®High = 42%). Vines treated with Actigard™ at the low rate also showed a reduction (33%) but this was not the case for vines treated with Actigard™ at the high rate. None of the Regalis® treatments had any statistically significant effect on pruning weights.

For the pilot study with Moddus® winter pruning weights per vine were c. 0.36 kg. This figure was also quite low and comparable with the 0.44 kg measured for B-Nine®SG. Thus Moddus® also appeared to be having a major effect on vine vigour and at this rate would be be expected to have reduced winter pruning requirements substantially.

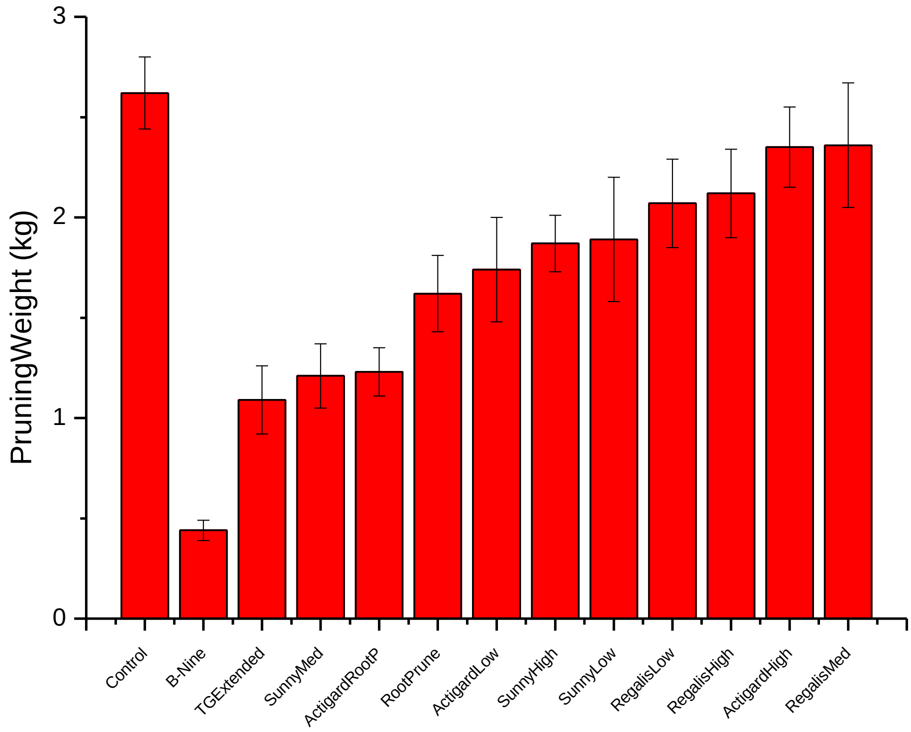


**Figure 6 Effects of vine vigour control treatments on pruning weights per vine at winter pruning 2013. Each treatment = 9 vines. (TG = Trunk girdling, RootP = Root pruning). Error bars are ±SE. B-Nine®SG, Sunny®Med, TGExtended and Actigard™plusRootPrune treatments were significantly different from Control at the 5% level**

Analysis of the proportion of shoots that required winter pruning in 2013 indicated that there were large differences between treatments. For control vines c. 23% of shoots required a pruning cut whereas for the vines treated with B-Nine®SG this proportion was reduced to c. 8% (Figure 7). This is a big difference and could be expected to significantly lower the overall number of pruning cuts required on a vine.

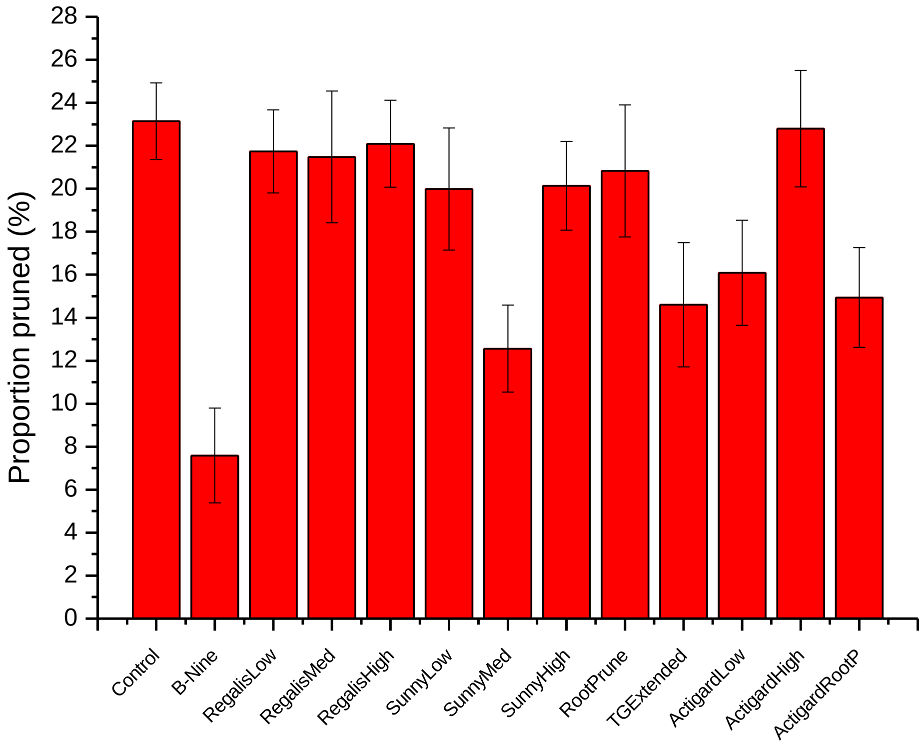


**Figure 7 Effects of vigour control treatments on proportions of shoots requiring pruning during winter 2013. Error bars = ±SE. Each treatment comprised 9 vines. Significant reductions were evident for B-Nine®SG ($p = 0.001$), SunnyMed ($p = 0.001$), TGExtended ($p = 0.023$) ActigardLow ($p = 0.033$) and ActigardplusRootPrune ($p = 0.013$)**

### d. Carry over effects on vine vigour

Analysis of proportions of short, medium, long and very long shoots on vines in March 2014 indicated that for the vines previously treated with B-Nine®SG, Sunny®Med, and Actigard™plusRootPruning the number of very long shoots was significantly reduced compared with control vines (Table 4). This was a strong indication that these treatments had significant carry over effects on vine vigour into the second growing season after application.

**Table 4 Effects of vigour control treatments on proportions of short, medium, long, and very long shoots on vines during the second growing season. Only treatments with marked vigour reductions (assessed by visual observation) and control vines were assessed. Data are means (n = 9). Values within a column followed by the same letter were not significant at the 5% level**

| Treatment | Number of short shoots (0-20 cm) | Number of medium shoots (20-60 cm) | Number of long shoots (60-100 cm) | Number of very long shoots (>100 cm) |
|---|---|---|---|---|
| Control | 20.9 a | 3.5 a | 2.3 a | 4.2 a |
| B-Nine®@7g/L, and | 20.1 a | 3.2 a | 3.1 a | 2.5 b |
| Sunny®@10ml/L | 23.3 a | 4.4 a | 1.3 a | 1.5 b |
| Actigard+RootPrune | 21.7 a | 4.2 a | 1.5 a | 1.1 b |

### e. Effects of vigour control treatments on flowering

Visual observations of vines in spring 2013 indicated that flowering was particularly heavy on B-Nine®SG treated vines and also heavy on vines that had a combination of root pruning and Actigard™. It was not possible to replicate assessments as fully as required so data should be interpreted as a guide only (Figure 8). Proportions of singles, doubles and triples also appeared to vary between treatments. For the B-Nine®SG treated vines the proportion of single flowers was very low (13%) compared with control vines (53%).

For the vines treated with Moddus® in the associated pilot study the proportion of single flowers was a very high 86%.

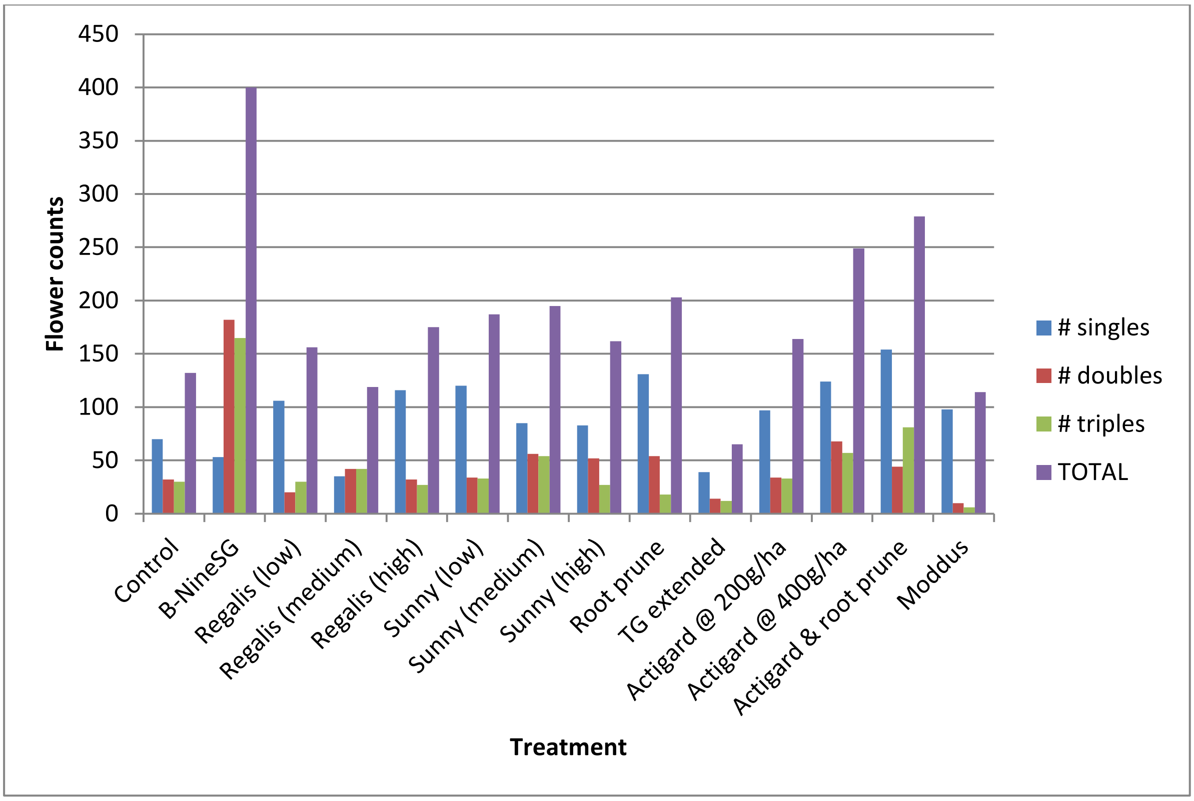


**Figure 8 Comparison of flower numbers per square meter of canopy on sample vines assessed in spring 2013. Each quadrat included at least 2 long canes, and several short canes and spurs. Insufficient data was obtained for statistical analysis. Results should be interpreted as a guide only. Data for Moddus® is included for comparative purposes**

### f. Effects on pollen viability

Results from pollen viability assays indicated that within the main trial the B-Nine®SG and extended trunk girdling treatments had significant negative effects on normal flower development (Figure 9). For the B-Nine®SG treatment, the reduction in pollen viability of 46% was considerable, and for the extended trunk girdling treatment the reduction of 15% was also marked. Both effects were statistically significant at the 1% level.

For the pollen sampled from the vines treated with Moddus®, the reduction in viability also appeared to be substantial. In comparison with untreated vines in the main trial the reduction was c. 30%.

Gibberellins are important in flower morphogenesis and perturbation of natural amounts may influence normal male or female flower development in dioecious species. It is suggested that the reduction in pollen viability observed in flowers in spring 2013 was a direct consequence of reductions in GA levels in the vines, possibly occurring much earlier in flower ontogeny.

For the extended trunk girdling treatment the reduction in pollen viability was somewhat unexpected. Trunk girdling is normally used on female vines to temporarily halt phloem transport during fruit growth and thereby to increase the allocation of carbohydrate to fruit. Girdling may however, also disrupt normal hormone transport processes within vines (Patterson and Boyd, 2013c). The effect of the extended trunk girdling treatment on pollen viability is possibly a consequence of such changes. As the treatment did reduce vine vigour it is possible that the pollen effect was hormone mediated, perhaps reflecting a significant reduction in cytokinin flow from roots because of limited carbohydrate flow from the vine parts above the girdle to developing roots.

The results indicate that the benefits of extended trunk girdling on reducing vine vigour would need to be offset against the negative effect on pollen viability. While the reduction in viability was not as great as observed for the B-Nine®Sg or Moddus® applications it would still need to be considered, particularly in male only pollen production orchards.

The Regalis® application did not affect pollen viability. The active ingredients in Regalis®, (prohexadione-Ca),  B-Nine®SG (daminozide), and Moddus® (trinexapac-ethyl) are members of the group of PGRs known as acylcyclohexanediones. These compounds all act in a similar way and inhibit the formation of highly active GAs from inactive precursors. It could therefore be expected that Regalis® might also influence pollen viability. As Regalis® did not affect flower morphogenesis and pollen viability, even at the high rate, it is a further indication that the product was ineffective at controlling growth processes within Chieftain when introduced via the xylem.

For  B-Nine®SG and Moddus® the large negative effects on pollen viability indicate that the feasibility of using these PGRs for vigour control of male vines is limited, at least for the application rates considered. Only one rate was used in the pilot study with Moddus® and lower rates of application could be considered.

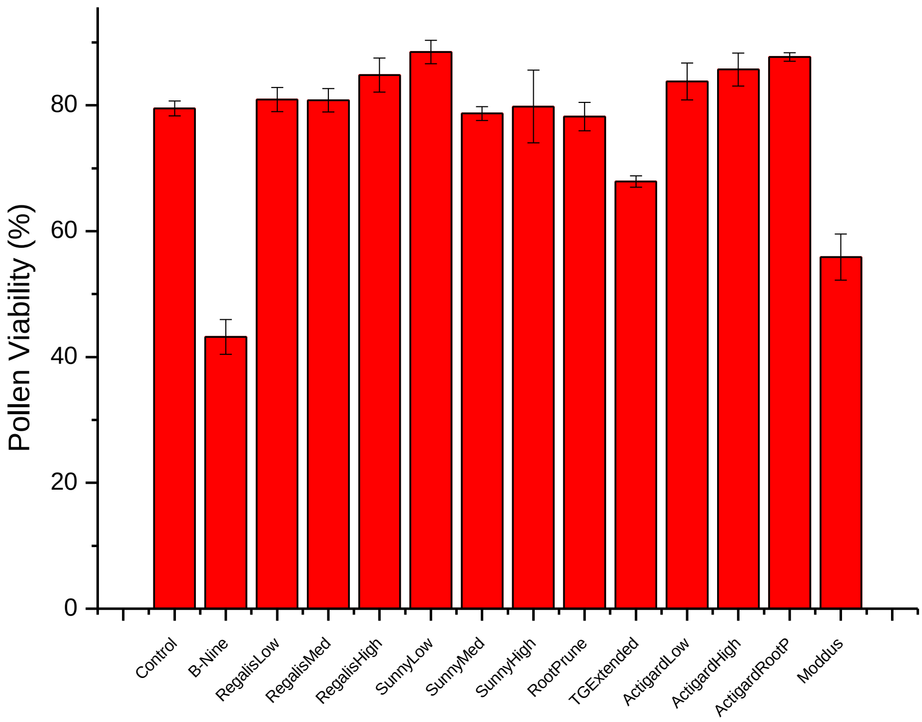


**Figure 9 Effects of PGRs, extended trunk girdling and root pruning on viability of Chieftain pollen sampled from vines during spring 2013. Error bars are ±SE. Pollen viabilities for the B-Nine®SG and extended trunk girdling treatments were significantly lower (at the 1% level) than those from control vines. Data for Moddus® are included for comparative purposes. The Moddus® treatment was not part of the main trial**

## 4. Conclusions

Uptake of B-Nine®SG, Regalis®, and Moddus® via injection into the trunk xylem was an effective way of introduction to 'Chieftain' vines. Uptake of Sunny® by this method was much less effective. Moddus® at the concentration used in the associated pilot study resulted in some leaf toxicity.

Growth control treatments including B-Nine®SG, Sunny® (medium conc.), extended trunk girdling, root pruning and Actigard™ plus root pruning significantly reduced vine vigour. The growth retardation effects translated into large reductions in winter pruning requirements. For the B-Nine®SG, extended trunk girdling, and Actigard™ plus root pruning treatments, reductions in winter pruning weights per vine were 83, 58, and 53% respectively. Reductions in vine vigour were principally from reduced proportions of vigorous non-terminating shoots (> 1 m long).

Flower numbers per unit canopy area were more than doubled by the application of B-Nine®SG. However, while several treatments had beneficial effects on vine vigour there was evidence that normal flower development leading to high pollen viability was impaired. Reductions in pollen viability of 46% for vines treated with B-Nine®SG and 30% for vines treated with Moddus® was apparent. Even for the vines treated with an extended trunk girdle, pollen viability dropped by 15%. Positive effects of vigour control therefore need to be balanced against reductions in pollen viability.

Actigard™ application via soil drench at a high rate (equivalent to 400g product per canopy ha. x 4 applications) did not reduce vine vigour whereas vines treated at the lower rate (200 g x 4) did show some evidence of reduced vigour, a result consistent with grower observations. Incidence of secondary symptoms of Psa-v within the trial area was restricted to one vine only. Analysis of effects of Actigard™ on vine health was not therefore possible as infection rates over the entire trial were very low.

Both treatments involving root pruning resulted in significant reductions in vine vigour without loss of pollen viability. This orchard management technique is relatively simple to apply and is now commonly used on female vines to enhance fruit dry matter content. It is recommended that further research into the technique for male vines be considered, particularly with respect to timing (pre- and post-flowering) and opportunities to sever higher proportions of root biomass.

## 5. References


Ferree DC 1992: Time of root pruning influences vegetative growth, fruit size, biennial bearing, and yield of 'Jonathan' apple. Journal of the American Society for Horticultural Science 117:198-202.

Patterson KJ, Currie MB, Black MZ, Martin PJ, Nicholls P, Jagielo C, Horner I 2009a: Kiwifruit Root Manipulation – Potential for Fruit Dry Matter Improvements. NZ Kiwifruit Journal, August 2009 pp 29-32.

Patterson KJ, Currie MB, Martin PJ, Black MZ, Nicholls P, Jagielo C 2009b: Taste Improvements in Kiwifruit via Root Manipulation: Year 2 (2008-2009). Report on Project TZ0847 prepared for ZESPRI Group Limited. Plant & Food Research Report No. 3288.

Patterson KJ, Martin PJ, Black M, Nicholls P, Burdon J, Haynes G 2011: Taste Improvements in Kiwifruit via Root Manipulation: Year 3 (2009-2010). Report on Project TZ0847, prepared for Zespri Group Ltd. Plant & Food Research Report No. 5202.

Patterson K J, Kashuba P, McCaughan L, Martin P J 2013a: Induced tolerance to Psa infection in male kiwifruit vines. Report on Project VI 1254 Part B, prepared for Zespri Group Ltd. Plant and Food Research Report No. 8973

Patterson KJ, Boyd L, Barnett A, Blattmann M 2013b: Options for Taste Improvements in 'Zesy002' (Gold3) Kiwifruit via Root Pruning. Report on Project 1375, prepared for Zespri Group Ltd. Plant and Food Research Report No. 8921

Patterson K J and Boyd L M 2013c: Effects of girdling on plant metabolites. Report on project VI 1377, prepared for Zespri Group Ltd. Plant and Food Research Report No. 8056.

Pentreath R, M Z Black, K J Patterson 2013: Soil ripping and root pruning. KiwiTech Bulletin No. 95, Zespri Group Ltd.