

RESEARCH PAPERS

The effects of hot water treatment, hydration and order of nursery operations on cuttings of *Vitis vinifera* cultivars

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Summary. Hot water treatment (HWT) is an effective control for endogenous pathogens, including *Phaeomonilla chlamydospora*, in grapevine propagating material. However sporadic unexplained failures of HWT material do occur. In order to determine the most reliable HWT protocols the effects of HWT at 50°C for 30 min., order of HWT and storage (store/HWT and HWT/store), and 3 hydration times (0, 4 and 6 h) on root and shoot development and final condition in dormant cuttings of Cabernet Sauvignon and Chardonnay were evaluated. After incubation callus, root and shoot development were assessed. Cuttings were potted into cardboard plant bands, grown to marketable size in a protected environment, and assessed as “A” grade, “B” grade or dead. Callus development in Chardonnay was affected by an interaction between HWT protocols and hydration times. Callus was least developed in cuttings hydrated for 15 h and stored before HWT. Callus development in all other treatments was greater ($P < 0.05$) regardless of HWT or hydration. By contrast, callus development in Cabernet Sauvignon was greater ($P < 0.05$) in HWT than in non-HWT cuttings regardless of the duration of hydration or the order of operations. Root development in Chardonnay was furthest advanced in cuttings hydrated for 15 h. (regardless of HWT) and in HWT cuttings not hydrated. HWT was the only factor that affected root development in Cabernet Sauvignon. Root development was greatest in non-HWT cuttings. There were no differences between any of the treatments in either variety at final assessment. On this evidence nurseries could apply any of the above protocols successfully. However the benign conditions of the protected environment may have enabled the cuttings to recover from the stresses imposed by the various treatments. Had the cuttings been grown in a field nursery there might have been differences between treatments at final assessment.

Key words: cold storage, Petri disease, crown gall, phytoplasmas.

Introduction

The presence of endogenous pathogens in planting material is recognised as a cause of poor vine vigour and lower than acceptable yields in newly

established vineyards with a commensurate reduction in income and return on capital (Smart, 1997; Morton, 1999). The need to control endogenous diseases such as crown gall, Petri disease and phytoplasmas in propagating and planting material has been realised both in Australia (Smart *et al.*, 1995) and elsewhere (Orffer, 1977; Crous *et al.*, 2001).

Hot water treatment at 50°C for 30 min is known to be an effective, practical and relatively

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inexpensive method for the control of a number of endogenous and exogenous grapevine pests and diseases in dormant grapevine cuttings and young rooted vines. However there continue to be regular anecdotal reports of unacceptably high rates of mortality when HWT is applied by nurseries to commercial batches of cuttings prior to callusing and to young rooted vines ready for despatch, and this has resulted in a reluctance by some nurseries to use HWT. Although HWT can be applied to young rooted vines just prior to despatch, most nurseries prefer to use it as a pre-callusing treatment to avoid the possibility of litigation from clients arising from the occasional unpredictable failure of HWT vines in the vineyard.

Currently there are no practical alternatives to HWT that can be used for large quantities of cuttings in commercial settings. Control of endogenous pathogens is difficult since traditional techniques such as chemical sprays and dips used for the control of surface pathogens do not penetrate dormant grapevine cuttings sufficiently to control organisms inhabiting the phloem and xylem tissue. However HWT of cuttings or young rooted vines at 50°C for 30 min. is regarded as an effective control of endogenous pathogens, including *Agrobacterium vitis* (Ophel *et al.*, 1990; Burr *et al.* 1996), *Phaeo-monniella clamydospora*, the primary invader in esca heart rot (Mugnai *et al.*, 1999; Crous *et al.*, 2001; Laukart *et al.*, 2001), and the phytoplasma *Flavescence dorée* (Caudwell *et al.*, 1997) since the heat is able to completely penetrate the wood, killing the pathogens, but not the marginally less sensitive vine tissue. Hot water treatment is also an effective control for external pests including nematodes (Lear and Lider, 1959; Meagher, 1960; Nicholas *et al.*, 1992) and phylloxera (Buchanan and Whiting, 1991).

The large numbers of cuttings processed by commercial nurseries in recent years has resulted in changes to storage practices, from burial in sand or sawdust-filled pits or boxes for cuttings, and covering the roots of young vines in a trench with loose soil, to storage in sealed plastic bags in cool rooms at temperatures of 1–5°C until material is required for callusing or despatch.

Some Australian nurseries report that grapevine cuttings and young vines subjected to HWT at 50°C for 30 min. after cold storage are less likely to suffer a loss of quality than material that is

HWTed before cold storage. However Wample (1993) reported that bud burst in Cabernet Sauvignon cuttings that had been HWTed (52–60°C for 10, 20 or 30 min.) post storage (3–4°C) was slower than in similar cuttings that were so treated before storage. Wample (1997) also reported that Cabernet Sauvignon cuttings treated at 52°C, 54°C and 56°C for 10, 20 or 30 min. and stored at 3–4°C thereafter generally showed better root development than cuttings treated after storage, but suggested that the results of that trial held good only for Cabernet Sauvignon grown in Washington State, where winters are very cold, and that other cultivars growing in other climates might perform differently.

In addition, it is generally accepted by the vine nursery industry in Australia that the standard practice of soaking propagating material overnight is beneficial and enhances the tolerance of cuttings to HWT (Nicholas *et al.*, 1992). However there is indirect evidence that pre-soaking plant material prior to HWT may reduce tolerance to HWT (Baker, 1962). Most other researchers investigating HWT (von Broembsen and Marais, 1978; Orffer *et al.*, 1979; Orffer and Goussard, 1980; Burr *et al.*, 1989; Burr *et al.*, 1996; Wample, 1997) do not mention periods of pre-HWT soaking, although Wample *et al.* (1991) mention a pre-HWT soaking period of 30 min.

While undertaking earlier experimental work (Waite, 2002) it was observed that cuttings that were hydrated overnight prior to HWT changed their colour from a light, bright brown to a dull black and that the buds tended to be soft and mushy. On the basis of this evidence and the indirect evidence of Baker (1962a) it was thought that soaking had the potential to damage grapevine cuttings. A small preliminary trial was designed to test the effects of pre-HWT soaking on the quality of Semillon cuttings. In this preliminary trial 3 bundles of 10 Semillon cuttings were soaked for 0, 6 and 15 hours prior to HWT and callused in clean coarse sand. They showed suppression of rooting after 6 and 15 hours soaking compared with bundles that were not soaked (Fig. 1).

These observations suggested that a more thorough examination of the effects of HWT, soaking and storage on the performance of grapevine cuttings was warranted. This paper reports

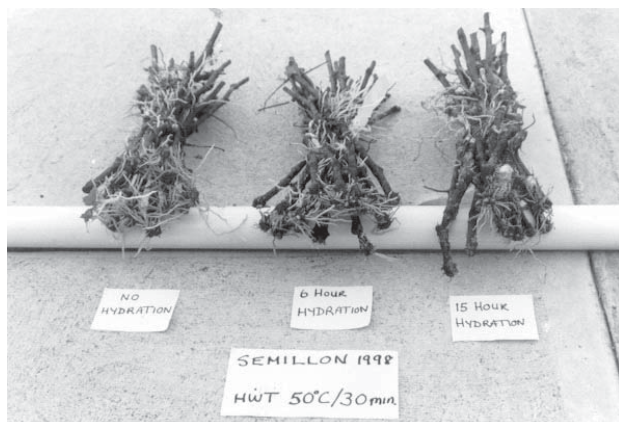


Fig. 1. Callused Semillon cuttings subjected to 0, 6 and 15 hours hydration prior to callusing.

on investigations into the effects of HWT, hydration and order of HWT and cold storage on dormant cuttings to determine the HWT protocols likely to be the most successful in commercial nurseries.

Materials and methods

A total of 900 cuttings, 450 each of the varieties Cabernet Sauvignon and Chardonnay, were collected from the Victorian and Murray Valley Vine Improvement Association (VAMVVIA) mother vine source blocks at Irymple in north west Victoria (Australia) on 21.6.1999. Cabernet Sauvignon and Chardonnay were chosen as they had been used in a preceding trial (Waite, 2002) and, in the case of Cabernet Sauvignon, to use the same variety as Wample (1991) in order that comparisons might be drawn between that work and this study. The cuttings were selected to conform to VAMVVIA standards: 7–12 mm in diameter and approximately 350–400 mm long, without obvious signs of disease or damage. The cuttings were allocated at random to 90 bundles of 10 cuttings, 45 bundles of each variety. Fifteen bundles (150 cuttings) of each variety were assigned at random to either no-HWT, pre-storage HWT, or post-storage HWT. Each group of 15 bundles was further divided into 3 groups of 5 bundles (50 cuttings) and subjected to either 0 or 4 or 15 hours hydration in the commercial hydration tanks at the VAMVVIA facility. Clean potable water was used as per standard industry practice.

Following hydration, the 15 bundles of each

variety that were to undergo pre-storage HWT were treated at 50°C for 30 min. as part of a commercial lot at the VAMVVIA hot water treatment facility at Irymple. On removal from the HWT tank the cuttings were immediately plunged into a cool-down tank of clean potable water at ambient temperature for 30 minutes. The cuttings were then removed from the cool-down tank and allowed to drain until there was no free moisture on the surface of the cuttings. All cuttings, including the group for post storage HWT and the group that did not receive any HWT, were then sealed in new plastic bags and stored in the VAMVVIA cool room at 1–2°C for 2 months. All cuttings were removed from storage on 24.8.1999 and left in the bags overnight to allow the material to stabilize to ambient temperature (approx. 10–12°C). The following day (25.8.1999) the 15 bundles of each variety to be HWTed post storage received HWT (50°C/30 min.) at the VAMVVIA facility following the same protocols that were used for the pre-storage HWT bundles.

All cuttings were then packed in moist vermiculite in new polystyrene vegetable boxes with the tops cut out and incubated in the callusing room of a commercial nursery at Irymple at 27°C (+/-0.5°C) and 95% humidity for 2 weeks as per standard industry practice. The cuttings were removed from the callusing room on 8–9.9.1999, assessed and potted into cardboard plant bands (bottomless containers 50 mm square packed in polystyrene boxes) using a standard nursery potting mix and placed in a glasshouse with other potted grapevine cuttings and cared for in the same manner. Fungicidal sprays were applied at regular intervals and shoot growth trimmed once with a brush cutter. By the time of final assessment on 6.10.1999, the cuttings had become established in the plant bands and were hardened off ready for removal to the holding area for saleable plants.

Assessment criteria

At the end of the callusing phase, callus development was assessed by estimating the percentage of callus formed around the basal end of each cutting. A complete ring of well-developed callus tissue was scored as 100%. For cuttings where the callus did not form a complete ring the score was calculated as a percentage to the nearest 5%. Root initials for each cutting were scored as 1, absent,

no visible sign of root initiation; 2, present, root initials present but less than 3 mm long; and 3, extended, root initials present and longer than 3 mm. Means of callus and root development were calculated for each bundle of 10 cuttings.

Since the cuttings in this experiment were propagated in a commercial nursery specialising in container-grown grapevine rootlings, final condition was assessed at the time the cuttings would normally be ready for sale as green potted vines for planting in the vineyard in late spring or early summer. The cuttings were assessed on 6.10.1999 when they had been hardened off and were ready for planting. They were assessed as either "A" grade, with a well developed root system emerging from the pot and two or more well developed shoots; "B" grade, with a strong root system that had not yet emerged from the pot and at least one moderately well developed shoot; "C" grade, with poor root and shoot development, or dead. "A" grade cuttings were assigned a score of 3, "B" grade cuttings were scored as 2 and "C" grade or dead cuttings were scored as 1, since for commercial purposes, both "B" and "C" groups are not saleable and are discarded.

Data analysis

Data were analysed using the Minitab statistical package and Microsoft Excel 97. Details of the analyses used are given in the Results section.

Results and discussion

Cultivar Chardonnay

Initially the data were analysed using a General Linear model ANOVA with the factors HWT and hydration. The levels of HWT were no HWT, HWT before storage and HWT after storage. The levels of hydration were no hydration, 4 hours hydration and 15 hours hydration.

Callus development

Although there was a significant main effect of HWT protocols on callus development in Chardonnay in this experiment, the interpretation of this effect is hampered by a significant interaction between HWT protocols and hydration. Discussion of these results is therefore based on one-way ANOVA with 9 individual treatments covering all possible combinations of HWT protocols and hydration times. Means separation was performed using Tukey's pairwise comparisons test ($P < 0.05$).

The development of callus tissue in Chardonnay in this experiment was consistently, but not always significantly ($P < 0.05$) greater in HWT cuttings that were stored before HWT than in either the group that was not HWTed or the group that was HWTed before storage, and this regardless of hydration time. In the post-storage HWT group there was also less variation in callus development between hydration times than there was in the group that received HWT before storage, or in the group that did not receive HWT. There was a significant difference ($P < 0.05$) in callus development between cuttings subjected to 4 and 15 hours hydration and cuttings HWTed before storage. The cuttings that were hydrated for 4 hours showed the most advanced callus development (82%) compared to cuttings that were hydrated for 15 h (53%). However neither group was significantly different ($P < 0.05$) from the group that received no hydration (69%) (Table 1).

Although callus development and root development are independent events (Hartmann *et al.*, 1990), the extent of callus development may be a useful indicator of the physiological state of the material at the time of assessment. In this experiment 15 hours hydration applied to cuttings that were then HWTed before storage suppressed or delayed callus development. The prolonged period

Table 1. Cultivar Chardonnay - means of callus development scores expressed as the percentage of the complete ring of callus formed at the basal circumference of each cutting.

Hydration time (h)	Hot water treatment/store	Store/hot water treatment	Store/no hot water treatment
0	69 ab	83 a	74 a
4	82 a	86 a	68 ab
15	53 b	81 a	81 a

Numbers followed by the same letter are not significantly different ($P < 0.05$).

in the relatively low-oxygen environment of the hydrating tank (Gray, 1999) may have resulted in intracellular flooding (Baker, 1962b) and the activation of anaerobic respiration that may persist since the cuttings are saturated during hydration (Baker and Chandler, 1957; Baker, 1962b). Under these conditions respiratory efficiency might be compromised (Raven and Johnson, 1992), resulting in delayed recovery from the stress of HWT and suppressed or delayed callus development.

The significantly greater ($P < 0.05$) callus development in 15-hour hydrated cuttings that were stored before HWT compared to the cuttings that were stored after HWT, suggests that in storage the cuttings may have recovered from the effects of prolonged hydration before they were subjected to the additional stress of HWT. The relatively warm (27°C) environment of the callusing box and the resulting increased respiratory rate may also have enabled quicker recovery from the stress of HWT in cuttings that were HWTed after storage compared with cuttings that were HWTed before storage. It is interesting to note that, unlike cuttings HWTed before storage, callus development in cuttings that were not HWTed, or that were stored before HWT, was not significantly affected ($P < 0.05$) by variations in hydration times. However, callus development in cuttings that were not HWTed was more variable than in cuttings that were HWTed post storage, indicating that the interaction between HWT and hydration is possibly affected by the order of storage and HWT.

Root development

In this experiment, root development in Chardonnay was significantly affected ($P < 0.05$) by HWT protocols and hydration time at the time of assessment. However interpretation of these effects is hampered by a significant interaction between HWT protocols and hydration time. Discussion of

these results is therefore based on one-way ANOVA with 9 individual treatments covering all combinations of the 2 factors (HWT protocols and hydration times). Means separation was performed using Tukey's pairwise comparisons. The results of this experiment indicate that root development at the time of assessment was most advanced in material that was HWTed but not hydrated, or hydrated for 15 hours. By contrast, root development in material that was neither hydrated nor HWTed, or that was hydrated for 15 hours regardless of the HWT protocol, was significantly less advanced ($P < 0.05$). It is interesting that 15 hours hydration appeared to stimulate root development, but 4 hours hydration apparently had no effect. It is also interesting that in this variety in this experiment, HWT apparently had no effect on root development in hydrated material. However, HWT did affect root development in material that had not been hydrated. The mean score of root development for material that was not hydrated or HWTed was significantly ($P < 0.05$) lower than that for either group of cuttings that were HWTed regardless of order of storage. By contrast, the results of a previous experiment (Waite, 2002) in which all cuttings were hydrated overnight (15–16 hours) and then HWTed, root development in Chardonnay cuttings collected in mid-season (at a date similar to the collection date in this experiment) was less advanced than in cuttings that were not HWTed. Since the cuttings used in the present experiment were collected from the same block of vines as those in the previous experiment, it would appear that one or more unidentified factors not examined in these experiments may affect the physiological state of the cuttings. Such factors might include climatic variations such as temperature and rainfall, or variations in management practices such as irrigation scheduling and fertiliser applications.

Table 2. Cultivar Chardonnay - means of root development where 1, roots absent; 2, roots <3 mm; 3, roots >3mm.

Hydration time (h)	Hot water treatment/store	Store/hot water treatment	Store/no hot water treatment
0	2 a ^a	1.9 a	1 b
4	1 b	1 b	1 b
15	2 a	1.8 a	2 a

Numbers followed by the same letter are not significantly different ($P < 0.05$).

Final condition

There were no significant ($P<0.05$), or observable differences between any of the treatment groups when the cuttings, now green potted vines, were assessed for final condition on 6.10.1999. All the cuttings in each treatment group were alive and healthy when assessed and had well developed roots and shoots. Means of final condition of all combinations of HWT and hydration are shown in Table 3.

There were no significant ($P<0.05$), or observable differences between any of the treatment groups when the cuttings, now green potted vines, were assessed for final condition on 6.10.1999. All the cuttings in each treatment group were alive and healthy when assessed and had well developed roots and shoots. Means of final condition of all combinations of HWT and hydration are shown in Table 3.

Although there was a significant interaction effect between HWT protocols and hydration on callus development and root development in this variety, the mean final condition scores for all treatment groups were relatively high (Table 3) indicating that the effects of hydration, HWT and order of HWT and cold storage on the cuttings at callusing were transient and did not affect the capacity of the cuttings to develop into sound rootlings.

At final assessment all the rootlings were sound and vigorous green potted vines and had they been planted out in a permanent vineyard as would happen in normal commercial practice, it is unlikely that there would have been any significant differences in vigour between the treatment groups during the establishment phase.

Cultivar Cabernet Sauvignon

Callus development

In this experiment there were significant main effects of both the HWT protocols and hydration

times on callus development when the results were analysed as a two-way factorial. There was no interaction between the 2 factors. To identify which levels of each factor were significantly different, a one-way ANOVA was run for each factor. In this analysis, only HWT protocols had a significant effect ($P<0.05$) on callus development. Means separation was performed using Tukey's pairwise comparisons test. When the data were analysed as one-way ANOVA for hydration time there was no significant effect of hydration time on callus development. Mean callus development in cuttings not hydrated was 60%; in cuttings that were hydrated for 4 and 15 hours it was 72 and 69% respectively.

Callus development in cuttings that were not HWTed (mean 46%) was significantly less ($P<0.05$) than that in either of the groups that were HWTed. Greatest callus development occurred in cuttings that were HWTed after cold storage (mean 83%), followed by cuttings that were hydrated before cold storage (mean 73%). In this variety HWT may provoke a wounding response in the tissue exposed by the basal cut that results in the stimulation of callus tissue. It is interesting that the pattern of callus development in Cabernet Sauvignon was broadly similar to that of Chardonnay in this experiment.

Root development

In this experiment root development in Cabernet Sauvignon cuttings at callusing was significantly affected by variations in HWT protocols, but not by hydration. There was no interaction between the 2 factors. Discussion of these results is therefore based on one-way ANOVA with 3 individual treatments covering all possible combinations of HWT protocols. Means separation was performed using Tukey's pairwise comparisons test ($P<0.05$). The mean root development score in cuttings that were not HWTed (2.9) was significantly ($P<0.05$) greater than

Table 3. Cultivar Chardonnay - means of final condition scores where 1, dead; 2, "B" grade; 3, "A" grade.

Hydration time (h)	Hot water treatment/store	Store/hot water treatment	Store/no hot water treatment
0	2.9	2.8	2.9
4	2.7	2.8	2.8
15	2.9	2.8	2.8

There are no significant differences ($P<0.05$) between any means in this table.

that of cuttings that were HWTed. The mean root development score for cuttings that received HWT before cold storage was 2.5; for cuttings that received HWT after cold storage it was 2.4.

The order of HWT and storage did not affect root development in the present experiment. Wample (1997) reported increased root numbers in Cabernet Sauvignon cuttings receiving pre-storage HWT. However in this experiment root development was assessed rather than root numbers since root initials are very fragile and easily knocked off during handling in commercial situations making it difficult to measure root numbers accurately.

It is interesting that HWT suppressed root development but stimulated callus development. The results of a regression analysis (Fig. 2) showed a strong negative relationship between callus and root development ($r^2=-0.86$) in Cabernet Sauvignon in this experiment. The results of this experiment contradict the perception in industry that well developed callus is a good indicator of potential root development.

Final condition

There were no significant ($P<0.05$), or observable differences between any of the treatment groups of Cabernet Sauvignon when the cuttings, now green potted vines, were assessed for final condition on 6.10.1999. All the cuttings in each treatment group were alive and healthy when assessed and had well developed roots and shoots. Means of final condition of all combinations of HWT and hydration are shown in Table 4.

As with Chardonnay in this experiment, the high mean final condition score for all treatment groups indicates that the effects of HWT protocols on Cabernet Sauvignon cuttings at callusing were transient and did not affect the capacity of the cuttings to develop into sound rootlings. As was the case with the Chardonnay cuttings in this experiment, all the rootlings were sound and vigorous at final assessment and it is unlikely that there would have been any significant differences in vigour between treatment groups once the rootlings were exposed to the harsher conditions in the vineyard.

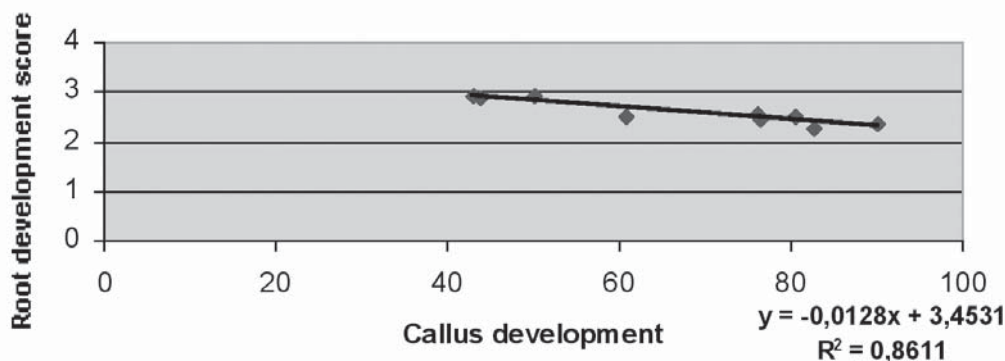


Fig. 2. Cultivar Cabernet Sauvignon regression plot – root development versus callus development.

Table 4. Cultivar Cabernet Sauvignon - means of final condition scores where 1, dead; 2, “B” grade; 3, “A” grade.

Hydration time (h)	Hot water treatment/store	Store/hot water treatment	Store/no hot water treatment
0	3	2.9	2.8
4	3	3	3
15	2.9	3	2.9

There are no significant differences ($P<0.05$) between any means in this table.

Conclusion and recommendations

The effects of different hydration times and HWT protocols on cuttings of Chardonnay and Cabernet Sauvignon at callusing were variable with different responses observed between varieties. In Chardonnay, callus development was consistently greater in cuttings that were HWTed post storage than in cuttings that were HWTed pre storage, or stored without HWT. Callus development was suppressed in cuttings that were HWTed pre storage and hydrated for 15 hours. In Cabernet Sauvignon callus development was greatest in cuttings that were hydrated for 4 hours and cuttings that were stored before HWT showed greater callus development than either cuttings that were HWTed post storage or cuttings that received no HWT.

Root development in Chardonnay was significantly affected by the HWT protocols, by the hydration times, and by the interaction between these two factors. Root development was most advanced in HWT cuttings that received no hydration, and in all cuttings that were hydrated for 15 hours regardless of HWT. These results are very interesting in that they seem to show that HWT had no effect on root development in Chardonnay cuttings that were hydrated, but stimulated root development in cuttings that were HWTed but not hydrated. In Cabernet Sauvignon cuttings, root development was affected by HWT protocols, but not by hydration. Most advanced root development in this variety at the time of assessment was in the group that were not HWTed, indicating that HWT suppressed early root development in this variety.

The reasons for the variable responses of the 2 varieties to HWT and hydration are difficult to explain in more specific terms other than that there clearly are complex interactions between each variety and the treatments applied. It is interesting to note that although callus and root development were affected by the treatments in this experiment, the final condition of the cuttings of both varieties was not affected by any of the treatments applied. Consequently the effects of HWT and hydration time observed at callusing were not a useful indicator of final condition. No cuttings of either variety from any treatment died and all the cuttings were of good commercial quality and considered saleable by the host nursery.

We therefore conclude that all the protocols tested in this experiment could be used successfully in a commercial situation. However the favourable growing conditions of the glasshouse and shade house compared to the more rigorous conditions in a field nursery where cuttings are exposed to climatic fluctuations may have aided recovery from any stress induced by HWT, hydration, or storage. Had the callused cuttings been planted in the field nursery instead of the more protected nursery environment, the final condition of the cuttings may have been different.

If, as nursery industry experience indicates, HWT has detrimental effects on grapevine cuttings, post-callusing handling is likely to play an important role in grapevine propagation. The high quality of the rooted vines resulting from the experiment reported here indicate that the protected environments of glasshouses and shade houses reduce the stress on cuttings during the establishment phase, resulting in fewer losses. Although the capital costs are relatively high, nurseries that do not have these facilities may wish to consider investing in glass houses and shade houses as a means of increasing the percentage of saleable vines resulting from HWT cuttings.

Although nurseries sometimes use HWT as a point-of-sale treatment, the effects of HWT on rooted vines have not yet been investigated, the issue of the occasional, unexplained failure of young HWT vines in the vineyard has not been resolved, and HWT as a pre-callusing treatment is generally preferred to avoid the risk of litigation. However it is possible that exposure to untreated water, contaminated soil and insect vectors could result in reinfection with *P. chlamydospora*, *A. vitis*, and phytoplasmas such as Flavescence dorée in the nursery during the post callusing phase (3–12 months), particularly if callused cuttings are planted in an open field nursery instead of being grown to maturity in sterile potting soil in enclosed glasshouses and shade houses. Until the reasons for the failure of HWT vines in the vineyard are fully understood nurseries may prefer to continue with the present practice of applying HWT to cuttings at the pre-callusing stage, before or after cold storage, and to introduce post-callusing practices such as water treatment and pest control to minimise the chances of reinfection in the nursery. Consistent with our current understanding of the effects

of HWT on grapevine cuttings and rooted vines, this strategy is the most practical method of ensuring the availability of healthy planting material for growers with least risk of litigation for nurseries.

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