Ultralow Oxygen Treatment for Control of *Planococcus ficus* (Hemiptera: Pseudococcidae) on Grape Benchgrafts

YONG-BIAO LIU,[1] LARRY J. BETTIGA,[2] AND KENT M. DAANE[3]

J. Econ. Entomol. 103(2): 272–276 (2010); DOI: 10.1603/EC09221

ABSTRACT Controlled atmosphere treatments with ultralow oxygen (ULO treatments) were developed successfully for control of vine mealybug, *Planococcus ficus* Signoret (Hemiptera: Pseudococcidae), on dormant grape (*Vitis* spp.) benchgrafts. At 30 ppm oxygen, 3-d ULO treatment at 25°C and 4-d ULO treatment at 15°C achieved complete control of all life stages of *P. ficus*. At a much lower oxygen level (<1 ppm), the two ULO treatments with the same exposure periods of 3 d at 25°C and 4 d at 15°C were tested on six table and wine grape cultivars grafted on rootstocks along with *P. ficus*. The benchgrafts were then potted in a greenhouse, together with untreated controls, to determine treatment effects on rootstock viability. Both ULO treatments achieved complete control of *P. ficus* and did not have any negative effects on vine growth, compared with the control. Results indicate that ULO treatments can be used to control *P. ficus* on dormant grape benchgrafts. The advantages of the ULO treatments are also discussed with respect to hot water treatments.

KEY WORDS controlled atmosphere, ultralow oxygen, *Planococcus ficus*, mealybug, grape benchgraft

The vine mealybug, *Planococcus ficus* Signoret (Hemiptera: Pseudococcidae), a major pest of in vineyards (Walton and Pringle 2004), is now found in most grape (*Vitis* spp.) production regions in California (Daane et al. 2008), although within each region not all vineyards are infested. The mealybug was first identified in southern California in 1994 (Gill 1994), but it had most likely entered the state earlier. There is circumstantial evidence that its rapid spread throughout the state, from 1998 to 2002, was the result of movement of infested nursery stock produced in southern California regions where the pest had become established (Daane et al. 2004). When populations in the vineyard are left untreated, this invasive mealybug has the potential to build up to large numbers and cause significant damage as a direct pest (Gutierrez et al. 2008), as well as a vector of viral leafroll diseases (Engelbrecht and Kasdorf 1990, Tsai et al. 2008). For this reason, control of *P. ficus* on nursery stock is critical to prevent further dissemination within California and other grape-growing regions where the pest has yet to be found. Most foliar insecticide treatments do not provide effective control for nursery stocks as a portion of the *P. ficus* population resides underneath the bark or on the roots. Dormant benchgrafts are resistant rootstocks grafted to a fruiting cultivar and field grown for one season and are the most common planting stocks for vineyards. Currently, *P. ficus* on dormant nursery cuttings is controlled using hot water treatment (Haviland et al. 2005). Hot water treatment can be labor-intensive and expensive, and excessive high temperatures can cause plant damage. Here, we investigated controlled atmosphere treatments as alternative control of *P. ficus* for grape nursery cuttings.

Controlled atmosphere with ultralow oxygen (ULO) treatment has been shown to be an effective postharvest treatment for black widow spiders on table grape clusters, lettuce aphid on head lettuce, and western flower thrips on lettuce and broccoli (Liu 2005, 2007, 2008a, 2008b, Liu et al. 2008), suggesting ULO treatment may also have potential to control *P. ficus* on grape nursery stocks. For fresh vegetables and fruits, any ULO treatment injury to the products, such as discoloration or lesions, would reduce quality and value of the products. For grape benchgrafts, budbreak and growth of benchgrafts are important. Therefore, we examined both the effects of ULO treatments on the mortality of *P. ficus* as well as on the growth of the exposed grape nursery stock material. Results are discussed with respect to the potential use of ULO treatments for *P. ficus* control on grape benchgrafts as an alternative to hot water dip treatment.

Materials and Methods

Mealybug Cultures. *P. ficus* was reared on sprouted potatoes (*Solanum tuberosum* L.) and butternut squash (*Cucurbita moschata* [Duchesne] Poir). The mealybug cultures were held in an environmental chamber at 22°C and a photoperiod of 14:10 (L:D) h.

[1] Corresponding author, USDA–ARS, Crop Improvement and Protection Unit, Salinas, CA 93905 (e-mail: yongbiao.liu@ars.usda.gov).
[2] UC Cooperative Extension, Salinas, CA 93901.
[3] Department Environmental Science, Policy and Management, UC Berkeley, CA 94720-3114.

New potatoes with sprouts and squash were added periodically to provide fresh host plant materials as the mealybug population density increased and the older plant materials dried out. Both potatoes and butternut squash were used because potatoes fit more easily into the jars for small-scale tests, whereas butternut squash was a better host and produced more *P. ficus.* The colonized host plant materials with the presence of mealybugs in the crawler (unsettled first instars), nymphal, and adult stages were selected for ULO treatments. Infested potatoes were placed, individually, in glass jars (0.95 liter) and sealed with paper towels. Infested butternut squashes were either placed directly in treatment chambers for small scale tests or placed in plastic containers (33 by 20 by 14 cm) and sealed with paper towels for larger scale tests on grape benchgrafts, respectively.

The mealybugs in the colonies typically deposited 100–300 eggs in an ovisac, a waxy cotton-like mass that protects the eggs. An accurate count of individual eggs in each ovisac before treatment exposure was difficult as the majority of eggs were buried in the cotton-like mass and attempting to count all eggs might damage eggs. For this reason, the number of eggs in each ovisac was estimated and, for each treatment, similar-sized ovisacs were divided evenly into groups, which were then placed into plastic vials (8.5 cm in height by 5.0 cm in diameter) on a piece of Kimwipe tissue (Kimberly-Clark, Roswell, GA). Each vial had two to four ovisacs. The vials were then sealed with screened lids lined with Kimwipe tissue to allow air circulation and assigned randomly to ULO treatments and the control. More than 150,000 mealybug crawlers and 45,000 mealybug nymphs and adults were tested. In total, 81 vials of mealybug eggs were used.

Response of Mealybug to ULO Treatment. ULO treatments were conducted in small treatment chambers modified from 7.6-liter (8-quart) pressure cookers. Infested potatoes (in glass jars), squash, and excised ovisacs (in vials) were placed in the treatment chambers for testing. In each treatment either ovisacs, infested hosts alone, or both were included depending on condition of the mealybug culture and availability of enough ovisacs. A small vial half filled with water was placed in each chamber to maintain air moisture during the ULO treatment. Treatment chambers were placed in temperature cabinets, with external temperature controllers and circulation fans to maintain uniform and accurate temperature (±0.5°C). ULO treatments with 30 ppm (0.003%) O_2 were conducted at 15, 25, and 35°C, with exposure periods ranging from 2 to 5 d, depending on temperature. To achieve 30 ppm oxygen level, the treatment chambers were initially flushed with bottled nitrogen. ULO treatments were maintained by constantly flowing pure nitrogen at 0.5 liter/min mixed with generated nitrogen gas with ≈0.2% O_2 from a nitrogen generator (Balston 75-7820, Parker Hannifin Co., Tewksbury, MA) by using two digital mass flow controllers (16 Series, Alicat Scientific, Tucson, AZ). Oxygen levels in treatment chambers were monitored periodically with an oxygen analyzer (Series 800, IL Instruments, Inc., Johnsburg, IL) with a detection limit of 0.01 ppm. During ULO treatment, the ovisacs (in vials) and infested potatoes (in jars) for the control were kept at 22°C and a photoperiod of 14:10 (L:D) h in an environmental chamber.

After ULO treatments were completed, treated host plant materials were kept overnight in an environmental chamber at 22°C and a photoperiod of 14:10 (L:D) h and then examined for mealybug survival by recording the number of live and dead nymphs and adults. When mealybug densities were high, nymphs and adults were brushed off from subsample areas of host plant onto butcher paper and examined under a dissecting microscope to record the numbers of live and dead mealybugs. Crawler numbers were often high and, in these cases, the numbers of dead crawlers were estimated by counting dead crawlers on small sections of surfaces of host materials. Treated mealybug ovisacs were held in an environmental chamber at 22°C and a photoperiod of 14:10 (L:D) h together with the controls for at least three weeks after ULO exposure periods to allow all viable eggs to hatch. A yellow sticky card was placed in each vial at the end of ULO treatment for both treatment and control to trap crawlers, after egg hatch, as they moved out of the ovisacs. The yellow sticky cards were then examined under a dissecting microscope, and the number of crawlers on each card was recorded. In total, seven ULO treatments were conducted, with two to five replicates for each treatment, except for the 5-d treatment at 15°C.

Effects of Ultralow Oxygen Treatment on Grape Benchgraft Growth. The effects of ULO treatments on *P. ficus* mortality and on grape benchgraft growth were examined using a 4-d exposure period at 15°C and a 3-d exposure period at 25°C. The treatments were conducted in plastic box chambers (56 by 41 by 25 cm). The box chamber had a sleeve made of plastic film and was sealed by folding and tying the sleeve. Rootstocks grafted with six grape cultivars: table grapes 'Autumn Royal', 'Crimson', and 'Flame Seedless'; wine grapes 'Chardonnay' and 'Merlot'; and the multiuse grape 'Thompson Seedless' were supplied by Sunridge Nurseries (Bakersfield, CA) and stored in plastic bags with wet sawdust at 2°C before being used in the ULO treatments. In each test, two benchgrafts from each of the six cultivars were pooled and placed in a plastic bag, with roots buried in wet sawdust. The bag was then placed in the treatment chamber. A butternut squash infested with *P. ficus* (sealed in a plastic box with a paper towel), and vials with *P. ficus* ovisacs also were placed in the bag and the opening of the bag remained unsealed. A fan was used to circulate air inside the treatment chamber.

The treatment chamber was placed in a temperature cabinet equipped with a circulation fan and an external temperature control, as described previously. The ULO treatment was established by flowing pure nitrogen, without mixing with generated nitrogen, which resulted in a low (<1 ppm) O_2 level, as compared with 30 ppm O_2 in the small-scale tests for the mealybug alone. Pure nitrogen from compressed cylinders was continuously released into the chamber

through a flow meter equipped with a control valve. The flow rate was initially 3–5 liter/min, to establish the ULO condition, and once the level dropped below 10 ppm O_2, nitrogen gas was released into the chamber at a flow rate of 0.8–1.0 liter/min. The O_2 level in the chamber was monitored with the oxygen analyzer as stated above and showed that 8 h after start of the treatment, the O_2 level had declined to the desired level of <1 ppm O_2.

Both treatments were replicated three times, and a total of 108 benchgrafts from the six cultivars were used. Benchgrafts used as controls were also pooled and stored in a plastic bag, with roots buried in wet sawdust and stored at 2°C in a walk-in cooler during the ULO treatment. Vine mealybug cultures and ovisacs (in vials) used as controls were kept in an environmental chamber at 22°C and a photoperiod of 14:10 (L:D) h. At the end of ULO exposure period, mealybug mortality was recorded using the same procedures as described above. For controls, most nymphs and adults were alive and the total number of nymphs and adults and their mortality were estimated to be >3,000 and <10%, respectively.

After the ULO treatment of benchgrafts, benchgrafts were stored at 2°C for 1–2 wk before planting. Benchgrafts were planted in potting soil in 18.9-liter buckets with drainage holes. For each cultivar, three benchgrafts (one from each of the two treatments and one from the control) were planted in one pot. The plants were watered twice a week. Budbreak and growth of the benchgrafts were evaluated at 30 and 60 d after planting, recording the presence or absence of budbreak and scoring the level of growth as low, medium or high (1, 2, and 3, respectively).

Data Analysis. Treatment comparisons for mealybug egg mortality were calculated based on the numbers of crawlers caught on yellow sticky cards. We assumed an equal number of viable eggs for each treatment and control in each replicate of ULO treatment and calculated relative mortality rates for each ULO treatment as ((1 − number of crawlers in ULO/number of crawlers in control) × 100). There was no difference in egg mortality between small-scale and box chambers for similar treatments (exposure period and temperature) and, for this reason, these data were pooled. Analysis of variance was used to determine treatment effects on benchgraft growth ratings, using JMP Discovery Statistics software (SAS Institute 2008).

Results and Discussion

ULO treatments at moderate temperatures (15 and 25°C) were effective against all life stages of *P. ficus.* Complete control of *P. ficus* eggs was achieved in 4-d and 5-d ULO treatments at 15°C and in 3-d ULO treatment at 25°C (Table 1). Complete control of all life stages of vine mealybug was achieved in all of the small-scale ULO treatments with infested potatoes or butternut squash only. In the large-scale ULO treatments with both infested hosts and dormant grape benchgrafts, complete control of all life stages of vine mealybug was achieved (Table 2). In comparison, crawler, nymphal, and adult stages were more susceptible to ULO treatments than egg stage (Tables 1 and 2). At 35°C, 2-d ULO treatment resulted in 99.8% egg mortality. This indicated that 2-d ULO treatment at 25°C would not produce complete control of *P. ficus* eggs because the potency of ULO treatment decreases with reduced temperature. Although the 3-d ULO treatment at 35°C also achieved complete control of *P. ficus* eggs, the treatment of the same duration at 25°C is preferred as the temperature is easier to achieve, and the treatment is likely to cause less stress to the benchgrafts.

ULO treatments of 3 d at 25°C and 4 d at 15°C did not have a negative effect on the growth of grape benchgrafts. All benchgrafts germinated and growth ratings at 30 and 60 d were similar among the three treatments (3 d at 25°C, 4 d at 15°C, and control; Table 3). The shoot length of vines ranged from 5 to 20 cm and 20 to 50 cm at 30 and 60 d after planting, respectively. There were no interactions between cultivars and treatments for growth rating scores; therefore, the main effects of treatments and cultivars were analyzed. There was no significant differences among treatments in the growth rating score at 30 d (F = 2.801; df = 2, 2; P = 0.066) or at 60 d (F = 1.329; df = 2, 2; P = 0.270). There were, however, significant differences among the six cultivars in the growth rating score with Flame Seedless performed significantly poorer compared with others at both 30 d (F = 4.410; df = 5, 5; P = 0.001) and 60 d (F = 19.861; df = 5, 5;

Table 1. Mortality of *P. ficus* eggs in response to ultralow oxygen treatments

Temp (°C)	Time (d)	Rep. (vial)[a]	Hatched eggs[b]	Mortality (%)[c]
15	3	2 (5)	1,183	52.5 ± 9.0
	4	5 (10)	0	100
	5	1 (3)	0	100
25	3	7 (21)	0	100
	4	3 (7)	0	100
35	2	2 (4)	1	99.8 ± 0.22
	3	2 (4)	0	100
Control		10 (27)	4,454	0

[a] Numbers of tests and total no. of vials of *P. ficus* eggs.
[b] Total number of crawlers caught on yellow sticky cards.
[c] Relative mortality based on numbers of crawlers caught on yellow sticky cards for specific treatment in comparison with the numbers of crawlers for the controls from the same tests.

Table 2. Mortality of *P. ficus* nymphs and adults in response to ultralow oxygen treatments in small scale tests and large scales tests with grape benchgrafts

Temp (°C)	Time (d)	Rep.	*N*	Mortality (%)
15	3	2	3,586	100
	4	7	27,900	100
	5	3	2,724	100
25	3	7	9,500	100
	4	7	3,335	100
Control	Potato	4	519	12.5 ± 2.9
	Squash	3	>3,000	<10

Table 3. Growth ratings of grape benchgrafts from ultralow oxygen treatments for control of *P. ficus* at 30 and 60 d after planting

Cultivar	Treatment	*N*	Growth rating	
			30 d	60 d
Autumn Royal	Control	6	3.0 ± 0.0	3.0 ± 0.0
	ULO (4 d/15°C)	6	3.0 ± 0.0	2.8 ± 0.2
	ULO (3 d/25°C)	6	3.0 ± 0.0	2.7 ± 0.2
Crimson	Control	6	3.0 ± 0.0	3.0 ± 0.0
	ULO (4 d/15°C)	6	3.0 ± 0.0	3.0 ± 0.0
	ULO (3 d/25°C)	6	2.7 ± 0.3	3.0 ± 0.0
Flame Seedless	Control	6	2.3 ± 0.3	2.0 ± 0.0
	ULO (4 d/15°C)	6	2.5 ± 0.2	1.8 ± 0.2
	ULO (3 d/25°C)	6	2.0 ± 0.4	1.8 ± 0.2
Thompson Seedless	Control	6	3.0 ± 0.0	3.0 ± 0.0
	ULO (4 d/15°C)	6	2.8 ± 0.2	2.8 ± 0.2
	ULO (3 d/25°C)	6	2.7 ± 0.3	2.7 ± 0.3
Chardonnay	Control	6	2.7 ± 0.3	2.7 ± 0.2
	ULO (4 d/15°C)	6	3.0 ± 0.0	3.0 ± 0.0
	ULO (3d/25°C)	6	2.8 ± 0.2	2.7 ± 0.2
Merlot	Control	6	2.5 ± 0.2	2.8 ± 0.2
	ULO (4d/15°C)	6	2.8 ± 0.2	2.8 ± 0.2
	ULO (3 d/25°C)	6	2.2 ± 0.4	2.8 ± 0.2
All cultivars	Control	36	2.7 ± 0.2	2.7 ± 0.2
	ULO (4 d/15°C)	36	3.0 ± 0.2	3.0 ± 0.2
	ULO (3 d/25°C)	36	2.8 ± 0.2	2.7 ± 0.2

$P < 0.001$; Tables 3 and 4). Although growth was measured only up to the first 60 d, there was no indication that there will be significant differences among treatments in the growth of the benchgrafts later on. Because the two ULO treatments tested on benchgrafts had a lower oxygen level than the ULO treatments tested on *P. ficus* alone, the data suggest that ULO with O_2 levels ranging from <1–30 ppm could be used to control of *P. ficus* with no detrimental effect on benchgrafts. This will also probably simplify the treatment process because there is no need to maintain a constant oxygen level as long as oxygen is at or below 30 ppm. Industrial nitrogen usually contains trace amount of oxygen. However, during ULO treatment, benchgrafts and microbes on sawdust consume oxygen and thereby reduce oxygen to the undetectable level.

Commercial hot water treatment protocols were developed for dormant grape cuttings that provided >99.8% control of *P. ficus* nymph and adults (Haviland et al. 2005). The recommended hot water protocol has additional benefits as a treatment for other pests, such as root knot nematodes (*Meloidogyne* spp.) (Barbercheck 1986) and grape phylloxera, *Daktulosphaira vitifoliae* (Fitch) (Stonerod and Strik 1996), and several bacterial pathogens, including Pierce's disease (*Xylella fastidiosa*) (Goheen et al. 1973) and *Agrobacterium* spp. (Burr et al. 1996, Ophel et al. 1990). However, these treatments can be labor-intensive and expensive, and higher temperature or longer exposure can cause damage to the grape plant (Wample 1997).

Table 4. ANOVA results on growth ratings of grape benchgrafts from ultralow oxygen treatments at 30 and 60 d after planting

Source	Nparm	df	30 d		60 d	
			F	*P*	*F*	*P*
Treat	2	2	2.801	0.066	1.329	0.270
Cultivar	5	5	4.410	0.001	19.861	<0.0001
Treat × cultivar	10	10	0.488	0.894	0.570	0.835

It is possible that the ULO treatment for control of *P. ficus* is also effective against other pests on grape benchgrafts. Because grape benchgrafts were able to tolerate all ULO treatments tested, it is likely that ULO treatments can be developed to control other pests that are more tolerant to ULO than *P. ficus* on grape benchgrafts without reducing viability. The tolerance of benchgrafts to the extreme low oxygen (anoxic) conditions makes the ULO treatment practical for commercial use. For large-scale commercial ULO treatment, dormant planting stocks could be held in a sealed enclosure or controlled atmosphere room and flushed with nitrogen gas and sealed for a specified time period at a selected temperature to accomplish the treatment to control *P. ficus.* The economics and effectiveness of such large-scale operations have not yet been investigated. Still, ULO treatment may have advantages over other grape-cutting treatments; for example, removal of all soil on dormant vines is not required as it is with hot water dips. Therefore, further development of ULO treatments is warranted and the treatment has potential for commercial use in the control of *P. ficus* and other pests on dormant grape planting stocks.

Acknowledgments

We thank C. Torres and J. Wasson for technical assistance and Sunridge Nurseries (Bakersfield, CA) for supplying grape benchgrafts. We also thank Drs. Peter Follett, Jim McCreight, and Xinzhi Ni for reviewing an earlier draft of the paper. This study was supported in part by funding from the Consolidated Central Valley Table Grape Pest and Disease Control District of California and the California Table Grape Commission.

References Cited

Barbercheck, M. 1986. Control of *Meloidogyne javanica* in dormant grapevine nursery stock. Phytophylactica 18: 39–40.

Burr, T. J., C. L. Reid, D. F. Splittstoesser, and M. Yoshimura. 1996. Effect of heat treatments on grape bud mortality and survival of *Agrobacterium vitis* in vitro and in dormant grape cuttings. Am. J. Enol. Viticul. 47: 119–123.

Daane, K. M., E. A. Weber, and W. J. Bentley. 2004. Vine mealybug – formidable pest spreading through California vineyards. Practical Winery & Vineyard. May/June. (www.practicalwinery.com).

Daane, K. M., M. L. Cooper, S. V. Triapitsyn, V. M. Walton, G. Y. Yokota, D. R. Haviland, W. J. Bentley, K. E. Godfrey, and L. R. Wunderlich. 2008. California vineyard managers and researchers seek sustainable solutions for mealybugs, a changing pest complex. Calif. Agric. 62: 167–176.

Engelbrecht, D. J., and G.G.F. Kasdorf. 1990. Transmission of grapevine leafroll disease and associated closteroviruses by the vine mealybug, *Planococcus ficus.* Phytophylactica 22: 341–346.

Gill, R. 1994. Vine mealybug. California Plant Pest and Disease Report, January–June. California Department of Food and Agriculture, Sacramento, CA.

Goheen, A. C., G. Nyland, and S. K. Lowe. 1973. Association of a Rickettsialike organism with Pierce's disease of grapevines and alfalfa dwarf and heat therapy of the disease in grapevines. Phytopathology 63: 341–345.

Gutierrez, A. P., K. M. Daane, L. Ponti, V. M. Walton, and C. K. Ellis. 2008. Prospective evaluation of the biological control of vine mealybug: refuge effects and climate. J. Appl. Ecol. 45: 524–536.

Haviland, D. R., W. J. Bentley, and K. M. Daane. 2005. Hot-water treatments for control of *Planococcus ficus* (Homoptera: Pseudococcidae) on dormant grape cuttings. J. Econ. Entomol. 98: 1109–1115.

Liu, Y.-B. 2005. Ultralow oxygen treatment for postharvest control of *Nasonovia ribisnigri* (Homoptera: Aphididae) on iceberg lettuce. J. Econ. Entomol. 98: 1899–1904.

Liu, Y.-B. 2007. Ultralow oxygen treatment for postharvest control of western flower thrips, *Frankliniella occidentalis*, on broccoli. J. Econ. Entomol. 100: 717–722.

Liu, Y.-B. 2008a. Ultralow oxygen treatment for postharvest control of western flower thrips, *Frankliniella occidentalis* (Thysanoptera: Thripidae), on iceberg lettuce, I. Effects of temperature, time, and oxygen level on insect mortality and lettuce quality. Postharvest Biol. Technol. 49: 129–134.

Liu, Y.-B. 2008b. Ultralow oxygen treatment for postharvest control of western flower thrips, *Frankliniella occidentalis* (Thysanoptera: Thripidae), on iceberg lettuce, II. Effects of pre-treatment storage on lettuce quality. Postharvest Biol. Technol. 49: 135–139.

Liu, Y.-B., K. M. Daane, J. S. Tebbets, and L. J. Bettiga. 2008. Ultralow oxygen treatment for control of *Latrodectus hesperus* (Araneae: Theridiidae) on harvested table grapes. J. Econ. Entomol. 101: 1515–1518.

Ophel, K., P. R. Nicholas, P. A. Magarey, and A. W. Bass. 1990. Hot water treatment of dormant grape cuttings reduces crown gall incidence in a field nursery. Am. J. Enol. Viticul. 41: 325–329.

SAS Institute. 2008. Introductory guide JMP 8. SAS Institute, Cary, NC.

Stonerod, P., and B. Strik. 1996. Hot-water dipping eradicates phylloxera from grape nursery stock. HortTechnology 6: 381–383.

Tsai, C.-W., J. Chau, L. Fernandez, D. Bosco, K. M. Daane, and R.P.P. Almeida. 2008. Transmission of grapevine leafroll-associated virus 3 by the vine mealybug (*Planococcus ficus*). Phytopathology 98: 1093–1098.

Walton, V. M. and K. L., Pringle. 2004. Vine mealybug, *Planococcus ficus* (Signoret) (Hemiptera: Pseudococcidae), a key pest in South African vineyards. A review. S. Afr. J. Enol. Viticul. 25: 54–62.

Wample, R. L. 1997. Influence of pre- and post-treatment storage on rooting hot-water treated cuttings of Cabernet Sauvignon. Am. J. Enol. Vitcul. 48: 131–136.

Received 7 July 2009; accepted 23 November 2009.