1 *Article*

2 Identification of *Vitis* Cultivars, Rootstocks and
3 Species Expressing Resistance to a *Planococcus*
4 Mealybug

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11 **Abstract:** Mealybugs cause economic loss to vineyards through physical damage, fouling fruit and
12 leaves with honeydew, and the transmission of viruses. *Planococcus ficus* is one of several mealybug
13 species in vineyards, and one that causes economic damage over a relatively large global range. To
14 develop novel management tools, host resistance to *P. ficus*, which has not previously been
15 identified for any grape cultivars, was studied. Ten grape lines (species, cultivars, and rootstocks)
16 were evaluated for *P. ficus* resistance across two separate potted plant assays. Significant differences
17 were detected among cultivars and rootstocks in the recorded number of *P. ficus* juveniles, adults
18 and egg sacs. Cabernet Sauvignon and Chardonnay were two of the most susceptible grape cultivars
19 for mealybug population growth, whereas rootstocks IAC 572, 10-17A and RS-3 all demonstrated
20 some level of resistance. Southern fire ant (*Solenopsis xyloni*) was positively associated with
21 mealybug populations, but did not have a negative effect on the observed presence of other
22 arthropod species including potential predators.

23 **Keywords:** host plant resistance; pest management; *Planococcus ficus*; vineyard

24

25 1. Introduction

26   Grapes have a long history of cultivation and breeding for a wide range of soils, climates and
27 commodities (e.g., table grapes for fresh consumption and processed grapes that are dried into raisins
28 or pressed for grape juice or wine) [1]. While there have been numerous studies on the development
29 of resistant cultivars to fungal and viral pathogens [2-4] and nematodes [5], with the exception of
30 grape phylloxera [6], there has been little work on grape cultivars that are resistant to key arthropod
31 pests. Globally, mealybugs (Hemiptera: Coccoidea: Pseudococcidae) are one of the more important
32 arthropod pests in vineyards [1] and economic losses resulting from mealybugs have dramatically
33 increased in the past decades, in part, as a result of globalization [7] despite the fact that many
34 countries impose regulations on the movement of vine material [8].
35   Vineyard mealybugs are phloem-feeding pests that can cause economic loss through feeding
36 damage to leaves, resulting in reduced photosynthetic capability, and the excretion of carbohydrate-
37 rich honeydew that can further foul the leaves, stems and fruit and lead to the accumulation of sooty
38 molds [9,10] (Fig. 1). In addition to losses attributed directly to feeding, mealybugs can transmit
39 grapevine leafroll associated viruses (GLRaVs) resulting in grape leafroll disease (GLD) [11,12] (Fig.
40 1), which has been estimated to cost grower between $12,106 to $91,623 per acre annually in California
41 [13]. Of that expenditure, mealybug control costs were estimated to range from $50 per acre for
42 vineyards with low mealybug population densities, and up to $500 per acre for vineyards with
43 moderate to large population densities [13]. At least ten mealybug species have been identified
44 globally that have risen to the level of economic pest in vineyards [9]. *Planococcus ficus* (Signoret) is

45 one of the most important vineyard mealybugs that has a global distribution [14], is a known vector
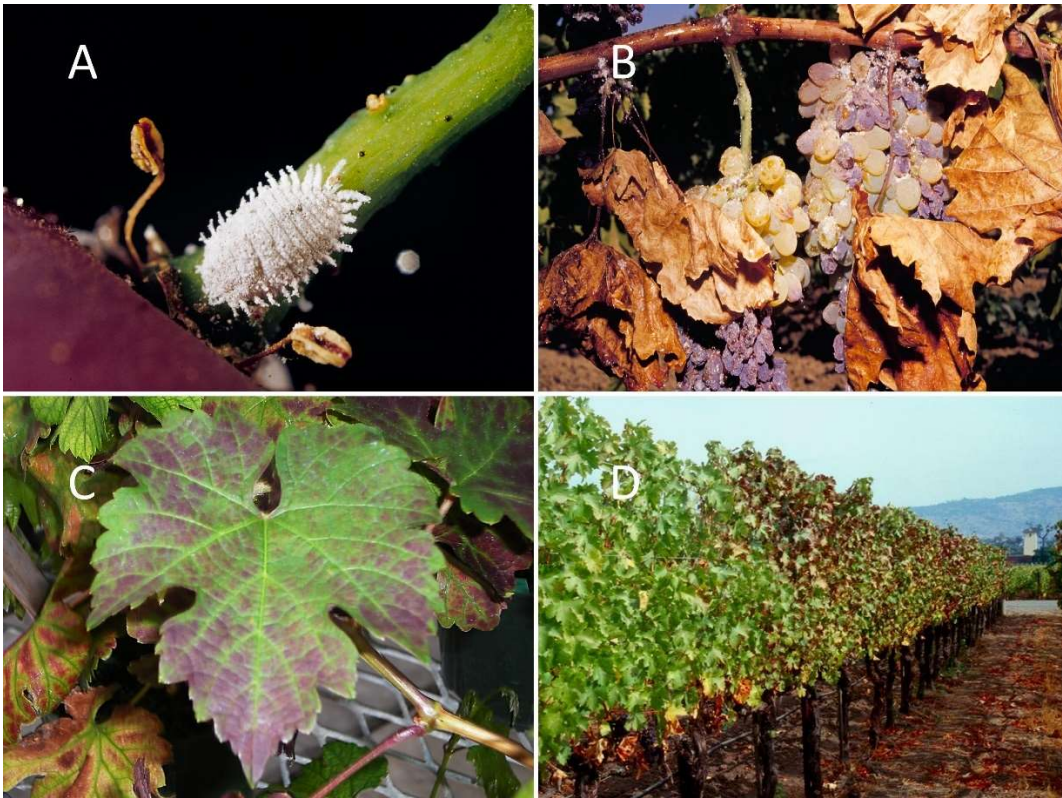46 of GLRaVs [15-18], and has become the primary pest in California vineyards [19].
47

49 **Figure 1.** Globally, mealybugs have become some of the more important vineyard pests; shown here
50 (**A**) an adult *Planococcus ficus* on a grape berry petiole, (**B**) direct damage from mealybugs feeding
51 on grape leaves, showing defoliation, and fruit clusters, showing berry damage and raising
52 (drying), (**C**) a single leaf showing grape leafroll disease (GLD) on a red-cultivar wine grape caused
53 by grape leafroll associated viruses transmitted by mealybugs, caused feeding damage to an
54 almond showing style puncture and kernel damage, and (**D**) a GLD-infested vine row.

55 Integrated pest management (IPM) systems are integral for mealybug management primarily in
56 the table and wine grape markets, and include cultural practices, such as cluster thinning and bark
57 stripping, but most farmers still rely on chemical controls to minimize exposure of the clusters to
58 mealybugs [19,20]. More sustainable tools for *P. ficus* control include mating disruption, which is
59 currently being used or tested worldwide as an alternative or complement to insecticide sprays [21-
60 24]. Biological controls are another tool to help suppress *P. ficus* populations, with a number of
61 predators that attack mealybugs, including the mealybug destroyer, *Cryptolaemus montrouzieri*
62 Mulsant, lacewings (e.g., *Chrysoperla* spp.), cecidomyiid flies (predaceous midges such as *Diadiplosis*
63 *koebelei* (Koebele)) [25-29]. Most successful biological control programs rely primarily on encrytid
64 parasitoids [30], such as *Anagyrus pseudococci* Signoret, a parasitoid of *Pl. ficus* and other related
65 mealybugs [26,29,31-33]. Even in organic or sustainable vineyards, natural enemies may not provide
66 complete control - ants have been shown to disrupt mealybug biological control in vineyards [33-36]
67 and *Pl. ficus* can find refuge from some natural enemy species under the vines bark [37]. For these
68 reasons, additional control tools should still be investigated.
69 Host resistance to *P. ficus* has not yet been developed or even investigated for grape. Classic
70 development of plant host resistance to insects typically occurs through antixenosis or antibiosis [38].
71 In antibiosis, the host adversely effects the insect resulting in increased mortality, reduced fecundity
72 or longevity, whereas antixenosis affects the behavior of the insect resulting in migration to a more
73 favorable host [39]. Resistance to other pests like phylloxera (*Daktulosphaira spp.*) and nematodes
74 (*Meloidogyne spp.*) have been identified in grape, primarily in native American species, which may

75　serve as a useful source of resistance to other pests [5,40,41]. Few sources of plant host resistance to
76　mealybugs have been identified, although antibiotic components of resistance were reported in
77　cassava cultivars to the cassava mealybug, *Phenacoccus manihoti* Matile-Ferrero, reducing the insect's
78　reproductive capacity [42]. Similarly, antiobiosis resistance was described for a grape rootstock to the
79　citrus mealybug, *Planococcus citri* (Risso), that had a reduction in the number of viable offspring
80　compared to susceptible cultivars [43] and these results were later reproduced with the pineapple
81　mealybug, *Dysmicoccus brevipes* (Cockerell) [44]. Our aim was to evaluate the potential of grape
82　rootstocks to impart resistance to *P. ficus* to improve vineyard IPM.

83　**2. Materials and Methods**

84　*2.1. Germplasm evaluation*

85　　　Own-rooted cuttings were collected from mature field-grown grapevines including rootstocks
86　and species at the San Joaquin Valley Agricultural Sciences Center (SJVASC), Parlier, CA or the
87　University of California's Kearney Agricultural Research and Extension Center (KARE), Parlier, CA
88　(Table 1). Plant material was selected based on known resistance to nematodes and suspected
89　resistance to mealybugs, as well as other agronomic traits of value. Ten replicate rooted plants were
90　transplanted into round 15.24 × 30.48 cm² black tree-pots (CP612R, Stuewe and Sons Inc, Tangent,
91　OR) and maintained in a screened cage at SJVASC. Potted plants were treated every two weeks with
92　sulfur to control powdery mildew, but did not receive any other insecticide treatments. Plants were
93　watered as needed.

94　　　**Table 1. Grape germplasm evaluated for resistance to vine mealybug in cage experiment**

Cultivar	*Vitis* species	Features [1]
USDA 1-2	*V. champinii*	Nematode resistance
PCO-349-11	Interspecific hybrid	Nematode resistance
IAC 572	*V. carabaea x* 101-14	Citrus mealybug resistance
10-17A	Interspecific hybrid	Nematode resistance
Australis [2]	*V. longii*	Phylloxera resistance
Cabernet Sauvignon	*V. vinifera*	Wine grape control

95　　　　　　[1] Special characteristics (insect resistance) of each genotype selected.
96　　　　　　　　　　[2] Australis is a cultivar name [45].

97　　　*Planococcus ficus* used was from an established colony reared on butternut squash at a KARE
98　insectary; the material originated from *P. ficus*-infested vines in Fresno County. To inoculate potted
99　vines, a single egg sac was placed onto a 60 mm piece of filter paper that was then attached to each
100　grape plant by wrapping the filter paper around the base of the stem and securing it with a stapler.
101　One week later, a second egg sac was placed onto each plant using the same method. The number of
102　mealybugs placed onto each plant, was estimated by the number of first instars that hatched per
103　ovisac from 20 randomly selected egg sacs.
104　　　Plants were evaluated every 1-2 weeks for a total of twelve weeks, for the number of mealybugs
105　(recorded as juveniles, or adult females) and ovisacs counted during a 1-minute rating period. A pre-
106　existing Southern Fire ant colony was located near the study, and the total number of ants were
107　recorded for each plant. Possible *P. ficus* predators, including lacewings, spiders, robber flies, and
108　other species were counted as presence/absence based on observation of the animal or parasitized
109　mealybugs. Southern fire ants, *Solenopsis xyloni* McCook, were observed tending mealybugs and were
110　not considered predatory. The experiment was repeated using a separate cage, approximately one
111　week after the initiation of the first experiment.
112　　　For each plant, an area under the insect growth curve (AIGC) value was determined based on the
113　area under the disease progress curve (AUDPC) calculation by Shaner and Finney [46]. In brief, the
114　number reflects insect population growth on each plant by accounting for the rate of change between
115　sample dates based on:

116
117
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$$AIGC = \sum_{i=1}^{N_i-l} \frac{y_i + y_i + 1}{2} \times (t_{i+1} - t_i)$$

119 Where, for each rating period, the number of insects observed (y_i) and the difference from the next
120 rating period (y_i+1) are averaged and compared to the amount of time (t) between the rating periods.
121 The sum of these calculations for the total number of observations (N) is the AIGC.
122   Data were compared for each line and cage using LSMeans implemented within SAS statistical
123 analysis software v 9.3 (Cary, NC). Significant differences were noted between the experiment cages
124 ($p = 0.0019$) and a significant line by cage interaction ($p = 0.0084$). Data were square root transformed
125 (mealybugs and ants) prior to analyses to improve normality, means were separated using Least
126 Significant Differences (LSD) at $p < 0.05$. Pearson's Correlation Coefficient was calculating using
127 PROC Corr implemented within SAS.

128

129   **Figure 2.** (**A**) Field design for the cultivar preference evaluating seven *Vitis* lines for resistance to
130   *Planococcus ficus* and (**B**) southern fire ants, *Solenopsis xyloni*, tending mealybugs in the trial, which
131   become an inadvertent but important aspect of mealybug response to *Vitis* cultivars.

132 *2.2. Cultivar preference*

133   A second experiment was conducted to determine differences in the number of mealybugs
134 among cultivars, with data recorded including different mealybug life stages. Own-rooted cuttings
135 were generated from mature field-grown grapevines at the SJVASC (Table 2). Ten replicate rooted
136 plants were transplanted into black tree pots (CP612R) and placed into similar sized pots buried into
137 the ground with an 8 cm block in the bottom to raise the internal pot height (7.62 x 7.62 cm^2) (Fig 2).
138 Two of the cultivars were only represented by 5 plants due to their availability (Table 2). To minimize
139 the presence and impact of predators and parasitoids, each vine was covered with a paint strainer
140 bag. Plants were treated every two weeks with sulfur to control powdery mildew, but did not receive
141 any insecticide treatments during the trial or 8 months prior to the start of the experiment. Plants
142 were watered as needed.
143   For inoculations, two hundred crawlers (first or second instar) were transferred to 60 mm filter
144 paper using a paintbrush, placed onto the base of each plant. A second set of two hundred crawlers
145 was placed onto each plant using the same method one week later, for a total of 400 crawlers per
146 plant. The total number of mealybugs were counted on each plants every two weeks during a 1-
147 minute timed search [47] recording mealybug numbers and their developmental stage (first, second
148 and third instars, adults, and ovisacs. A pre-existing southern fire ant population was located near
149 the study, and the total number of ants were recorded for each plant on each sample date. Plant health
150 was also monitored using a 1-5 scale with 1 being dead and 5 being completely healthy.

151   **Table 2. Grape germplasm evaluated for resistance to vine mealybug in cultivar preference**
152   **experiment**

Cultivar	No. Plants [1]	Species	Features
Autumn King	10	*V. vinifera*	table grape control
Cabernet Sauvignon	10	*V. vinifera*	wine grape control
IAC 572	10	Interspecific hybrid	*P. citri* and *D. brevipes* resistance
RS-3	5	Interspecific hybrid	mealybug resistance (anecdotal)[2]
Flame Seedless	5	*V. vinifera*	table grape
Chardonnay	10	*V. vinifera*	wine grape
Valley Pearl	10	*V. vinifera*	table grape

153 [1] Number of plants included in the study and used for analyses.

154 [2] Based on observations by Dr. M. Mckenry (*personal communication*)

155 *2.3. Data Analysis*

156   Results are presented as sample means (±SEM). For each plant, an area under the insect growth
157 curve (AIGC) value was determined based on the AUDPC calculation by Shaner and Finney for both
158 ants and vine mealybugs [46]. The number of third instars to adults were combined for analyses (e.g.,
159 first instars and third instars to adults were analyzed separately). Data were compared for each line
160 using LSMeans implemented within SAS statistical analysis software v9.3 (Cary, NC). Data were log
161 transformed (vine mealybugs and ants) prior to analyses to improve normality, means were
162 separated using Tukey's Honest Significant Difference at $p < 0.05$. Pearson's Correlation Coefficient
163 was calculating using PROC Corr implemented within SAS.

164 **3. Results**

165 *3.1. Annual generations and seasonal development*

166   For the cage experiment, significant differences were detected among cultivars ($p < 0.0001$) for
167 mealybug and ant population growth over time. Cabernet Sauvignon, the susceptible control,
168 consistently had higher numbers of mealybugs and ants throughout the experiment than any of the
169 other materials evaluated (Table 3, Fig 4). Population growth (AIGC values) was lower, on average,
170 in the second run of the experiment compared to the first experiment. No significant differences were
171 detected among the rootstock cultivars or wild species in the first run, and only minor differences in
172 the second ($p = 0.05$). The number of ants detected per rootstock and wild species had a significant
173 cultivar ($p < 0.0001$) and cage ($p < 0.0001$) effect, but no significant interaction was detected between
174 cultivar and cage. The number of predators detected was significantly different among cultivars ($p <$
175 0.0001), but not by cage or the interaction between cultivar and cage. A greater numbers of ants was
176 associated with higher numbers of mealybugs across all lines evaluated (r = 0.62115, $p < 0.0001$);
177 however, ant density was not associated with predator presence (r = 0.17420, $p = 0.0581$).
178   For the cultivar preference experiment, significant differences were detected among cultivars for
179 first instars ($p < 0.0001$) and third instars and adults ($p = 0.0007$). Chardonnay had the greatest number
180 of mealybugs (juveniles, adults, and ovisacs) and was significantly different compared to both IAC
181 572 and RS-3 rootstocks (Table 4). Most of the commercially available scion cultivars were not
182 significantly different from each other. Valley Pearl had a lower number of crawlers compared to the
183 other cultivars, but was not significantly different in the number of adult female mealybugs visible.
184 Rootstocks IAC 572 and RS-3 both had fewer mealybugs (juveniles and adults), than cultivars
185 Chardonnay, Autumn King and Cabernet Sauvignon, but high variability in mealybug populations
186 were observed within scion cultivars. Strong correlations were observed between the number of ants
187 detected and mealybug populations (r = 0.37087, $p = 0.0001$ and r = 0.4864, $P < 0.0001$ for crawlers and
188 adults, respectively.)

189 **Table 3. Vine mealybug and ant population growth on grapevines evaluated in cage experiment**

Cultivar	Trial [1]	Mealybug AIGC [2]	Ant AIGC	Predators [3]
10-17A	1	192 c[4]	184.8 d	68% a

	2	127.75	cd	19.15	e	45%	bcd
Cabernet Sauvignon	1	733.1	b	869.4	a	48%	abc
	2	1026.1	a	549.0	b	53%	a
IAC 572	1	89.5	cd	242.9	cd	23%	e
	2	8.8	e	27.5	e	33%	bcde
PCO-349	1	151.75	c	379.4	bc	30%	cde
	2	53	de	36.0	e	33%	bcde
Australis	1	205.75	c	291.2	cd	25%	de
	2	78	cd	29.5	e	30%	cde
USDA 1-2	1	99.8	cd	340.9	cd	33%	bcde
	2	8	e	27.1	e	23%	e

[1] Indicates the first or second experimental trial

[2] Area under the insect growth curve based on the formula from Shaner and Finney [46].

[3] Percentage of plants with predatory insects or arachnids or evidence of them present.

[4] Numbers followed by the same letter within a column are not significantly different (P = 0.05).

Table 4. Population growth of vine mealybug and ants on grapevines evaluated in cultivar study

Cultivar	Immature mealybugs AIGC [1]		Adult mealybugs AIGC [2]		Mealybug ovisacs AIGC		Ant AIGC		Plant health	
Autumn King	530.4	ab [3]	547.6	a	98.8	ab	53.7	a	4.6	a
Cabernet Sauvignon	542.5	abc	279.3	ab	56	b	46.9	a	3.4	b
IAC 572	75.6	c	54.6	b	9.1	b	17.5	c	4.9	a
RS-3	7.0	c	9.8	b	1.4	b	2.8	c	3.2	b
Flame Seedless	95.2	abc	133.0	ab	30.8	b	5.6	bc	4.8	a
Chardonnay	1463	a	532.7	a	161.7	a	39.2	a	3.8	ab
Valley Pearl	100.8	c	272.3	ab	32.9	b	37.1	ab	4.0	b

[1] Area under the insect growth curve based on the formula from Shaner and Finney [46].

[2] Adult female mealybugs and third instar juveniles.

[3] Numbers followed by the same letter within a column are not significantly different (P = 0.05).

4. Discussion

Planococcus ficus is a serious insect pest of grapes with no management tools that provide complete control [14,20,23,48-51]. We evaluated ten grape cultivars, rootstocks and species for their relative resistance to *P. ficus* population growth. Each of the rootstocks evaluated showed reduced mealybug population numbers compared to *V. vinifera* controls (*cv*. Cabernet Sauvignon and Chardonnay) and we suggest the reduced population growth could result from some level of antibiosis or antixenosis resistance mechanisms. Female mealybugs while sessile when adults, can travel several feet or more to find a host during early stages of development. In contrast to previous studies by Bertin et al. [44] and Filho et al. [43], IAC 572 did have some mealybugs visible throughout the study, suggesting that viable offspring were produced, though in low numbers. This could be, in part, due to differences in the three mealybug species in host preference and reproduction methods. RS-3, which had been suspected to protect roots against vine mealybug [52], showed few crawlers or adults throughout the study. These data suggest that sources of resistance to vine mealybug do exist, and that there may be differences in mechanisms of resistance among *Vitis* spp. Ants effect natural enemy effectiveness, although their impact depends on the ant and natural enemy species [34,35,53,54]. Surprisingly, in our study ant populations were associated with higher mealybug numbers, but had little effect on presence/absence of natural enemies.

Though all *Vitis vinifera* cultivars appear to be susceptible to vine mealybug, scion variability exists. In our results, table grape cultivars Valley Pearl and Flame Seedless had fewer adult mealybugs and egg sacs over time compared to the wine grape cultivars Chardonnay and Cabernet Sauvignon and the table grape cultivar Autumn King. This is similar to previous studies evaluating

219 mealybug resistance in cassava, mango, and buffalo grass where cultivar differences were observed
220 [42,55].
221   In summary, we identified at least two sources of resistance to vine mealybug under potted plant
222 conditions in a semi-natural environment. The commercially available, though not widely used
223 rootstocks IAC 572 and RS-3 may be useful in grape growing regions with high mealybug pressure
224 as part of an IPM program. Further studies to identify additional sources of resistance and determine
225 if these mechanisms are acting through anti-biosis or xenosis are needed.

5. Conclusions

227   *Planococcus ficus* is one of several mealybug species found in grape vineyards globally. Resistant
228 grape cultivars, which are an important component of IPM, are not available to manage this insect
229 pests. Evaluation of grape rootstocks and cultivars identified differences in mealybug population
230 growth. Both juvenile and adult female mealybug, and Southern fire ant populations were lower on
231 rootstocks than on cultivated varieties. Because of the variability in mealybug growth even on the
232 rootstocks, it is likely that there are cultivar-specific mechanisms contributing to mealybug resistance.
233 These mechanisms could be physical or chemical features that affect feeding and host attractiveness
234 to mealybugs. While several ant species are associated with *P. ficus*, the specific effect of each species
235 on mealybug population growth has not been evaluated. The presence of ants was correlated with
236 higher numbers of mealybugs, but not the absence of mealybug predators. Here we confirm the
237 existence of mealybug resistance in *Vitis spp.*, and identify rootstocks useful for breeding and IPM.
238 Follow-up studies should include a multi-year evaluation of these rootstocks in a vineyard setting
239 under high and low vine mealybug pressure.

240 **Author Contributions:** R.P.N. secured funding, conceived the article, and supervised data collection. K.M.D.
241 and P.C. provided advice throughout the study. R.P.N analyzed the data and wrote the initial draft; all authors
242 contributed to the final draft.

249 **Conflicts of Interest:** The authors declare no conflict of interest.

References

251 1.   Daane, K.M.; Vincent, C.; Isaacs, R.; Ioriatti, C. Entomological opportunities and challenges for
252   sustainable viticulture in a global market. *Annu. Rev. Entomol.* **2018**, *63*, 193-214, doi:10.1146/annurev-
253   ento-010715-023547.
254 2.   Ramming, D.W.; Gabler, F.; Smilanick, J.; Cadle-Davidson, M.; Barba, P.; Mahanil, S.; Cadle-Davidson,
255   L. A single dominant locus, ren4, confers rapid non-race-specific resistance to grapevine powdery
256   mildew. *Phytopathology* **2011**, *101*, 502-508, doi:10.1094/phyto-09-10-0237.
257 3.   Xu, K.; Riaz, S.; Roncoroni, N.C.; Jin, Y.; Hu, R.; Zhou, R.; Walker, M.A. Genetic and QTL analysis of
258   resistance to Xiphinema index in a grapevine cross. *Theor. Appl. Genet.* **2008**, *116*, 305-311,
259   doi:10.1007/s00122-007-0670-6.
260 4.   Gadoury, D.M.; Seem, R.C.; Ficke, A.; Wilcox, W.F. Ontogenic resistance to powdery mildew in grape
261   berries. *Phytopathology* **2003**, *93*, 547-555, doi:10.1094/phyto.2003.93.5.547.
262 5.   Ferris, H.; Zheng, L.; Walker, M.A. Resistance of grape rootstocks to plant-parasitic nematodes. *J.*
263   *Nematol.* **2012**, *44*, 377-386.
264 6.   Granett, J.; Walker, M.A.; Kocsis, L.; Omer, A.D. Biology and management of grape phylloxera. *Annu.*
265   *Rev. Entomol.* **2001**, *46*, 387-412, doi:10.1146/annurev.ento.46.1.387.
266 7.   Vila, M.; Basnou, C.; Pysek, P.; Josefsson, M.; Genovesi, P.; Gollasch, S.; Nentwig, W.; Olenin, S.; Roques,
267   A.; Roy, D., et al. How well do we understand the impacts of alien species on ecosystem services? A
268   pan-European, cross-taxa assessment. *Front Ecol Environ* **2010**, *8*, 135-144, doi:10.1890/080083.

269   8.   Golino, D.A.; Fuchs, M.; Al Rwanih, M.; Farrar, K.; Martelli, G.P. Regulatory aspects of grape viruses
270   and virus diseases: certification, quarantine, and harmonization. In *Grapevine Viruses: Molecular Biology,*
271   *Diagnostics And Management*, Springer: New York, in press.
272   9.   Daane, K.M.; Almeida, R.P.P.; Bell, V.A.; Botton, M.; Fallahzadeh, M.; Mani, M.; Miano, J.L.; Sforza, R.;
273   Walton, V.M.; Zaveizo, T. Biology and management of mealybugs in vineyards. In *Arthropod*
274   *Management in Vineyards*, Bostanian, N.J., Isaacs, R., Vincent, C., Eds. Springer: Netherlands, 2012;
275   10.1007/978-94-007-4032-7_12pp. 271-308.
276   10.   Charles, J.G. Economic damage and preliminary economic thresholds for mealybugs (*Pseudococcus*
277   *longispinus* T-T.) in Auckland vineyards. *N. Z. J. Agric. Res.* **1982**, *25*, 415-420,
278   doi:10.1080/00288233.1982.10417905.
279   11.   Engelbrecht, D.J.; Kasdorf, G.G.F. Transmission of grapevine leafroll disease and associated
280   closteroviruses by the vine mealybug, *Planococcus ficus*. *Phytophylactica* **1990**, *22*, 341-346.
281   12.   Almeida, R.P.P.; Daane, K.M.; Bell, V.A.; Blaisdell, G.A.; Cooper, M.L.; Herrbach, E.; Pietersen, G.
282   Ecology and management of grapevine leafroll disease. *Frontiers in Microbiology: Virology* **2013**, *4*, doi:
283   10.3389/fmicb.2013.00094.
284   13.   Ricketts, K.D.; Gomez, M.I.; Atallah, S.S.; Fuchs, M.F.; Martinson, T.E.; Battany, M.C.; Bettiga, L.J.;
285   Cooper, M.L.; Verdegaal, P.S.; Smith, R.J. Reducing the economic impact of grapevine leafroll disease
286   in California: Identifying optimal disease management strategies. *Am. J. Enol. Vitic.* **2015**, *66*, 138-147,
287   doi:10.5344/ajev.2014.14106.
288   14.   Daane, K.M.; Middleton, M.C.; Sforza, R.F.H.; Kamps-Hughes, N.; Watson, G.W.; Almeida, R.P.P.;
289   Correa, M.C.G.; Downie, D.A.; Walton, V.M. Determining the geographic origin of invasive
290   populations of the mealybug *Planococcus ficus* based on molecular genetic analysis. *Plos One* **2018**, *13*,
291   doi:10.1371/journal.pone.0193852.
292   15.   Mahfoudhi, N.; Digiaro, M.; Dhouibi, M.H. Transmission of grapevine leafroll viruses by Planococcus
293   ficus (Hemiptera: Pseudococcidae) and Ceroplastes rusci (Hemiptera: Coccidae). *Plant Dis.* **2009**, *93*,
294   999-1002, doi:10.1094/pdis-93-10-0999.
295   16.   Tsai, C.W.; Bosco, D.; Daane, K.M.; Almeida, R.P.P. Effect of Host Plant Tissue on the Vector
296   Transmission of Grapevine Leafroll-Associated Virus 3. *J. Econ. Entomol.* **2011**, *104*, 1480-1485,
297   doi:10.1603/ec10412.
298   17.   Blaisdell, G.K.; Zhang, S.; Bratburd, J.R.; Daane, K.M.; Cooper, M.L.; Almeida, R.P.P. Interactions
299   Within Susceptible Hosts Drive Establishment of Genetically Distinct Variants of an Insect-Borne
300   Pathogen. *J. Econ. Entomol.* **2015**, *108*, 1531-1539, doi:10.1093/jee/tov153.
301   18.   Bertin, S.; Pacifico, D.; Cavalieri, V.; Marzachi, C.; Bosco, D. Transmission of Grapevine virus A and
302   Grapevine leafroll-associated viruses 1 and 3 by *Planococcus ficus* and *Planococcus citri* fed on mixed-
303   infected plants. *Ann. Appl. Biol.* **2016**, *169*, 53-63, doi:10.1111/aab.12279.
304   19.   Daane, K.M.; Bentley, W.J.; Smith, R.J.; Haviland, D.R.; Weber, E.; Gispert, C.; Battany, M.C.; Millar, J.G.
305   *Planococcus* mealybugs (Vine mealybug). In *Grape Pest Management, Publication 3343*, 3rd ed.; Bettiga, L.,
306   Ed. University of California, Division of Agriculture and Natural Resources: Oakland, CA, 2013; pp.
307   246-260.
308   20.   Mansour, R.; Belzunces, L.P.; Suma, P.; Zappala, L.; Mazzeo, G.; Grissa-Lebdi, K.; Russo, A.; Biondi, A.
309   Vine and citrus mealybug pest control based on synthetic chemicals. A review. *Agron. Sustain. Dev.*
310   **2018**, *38*, doi:10.1007/s13593-018-0513-7.
311   21.   Walton, V.M.; Daane, K.M.; Bentley, W.J.; Millar, J.G.; Larsen, T.E.; Malakar-Kuenen, R. Pheromone-
312   based mating disruption of *Planococcus ficus* (Hemiptera : Pseudococcidae) in California vineyards. *J.*
313   *Econ. Entomol.* **2006**, *99*, 1280-1290.
314   22.   Cocco, A.; Lentini, A.; Serra, G. Mating Disruption of Planococcus ficus (Hemiptera: Pseudococcidae)
315   in Vineyards Using Reservoir Pheromone Dispensers. *Journal of Insect Science* **2014**, *14*,
316   doi:10.1093/jisesa/ieu006.
317   23.   Sharon, R.; Zahavi, T.; Sokolsky, T.; Sofer-Arad, C.; Tomer, M.; Kedoshim, R.; Harari, A.R. Mating
318   disruption method against the vine mealybug, *Planococcus ficus*: effect of sequential treatment on
319   infested vines. *Entomol. Exp. Appl.* **2016**, *161*, 65-69, doi:10.1111/eea.12487.
320   24.   Mansour, R.; Grissa-Lebdi, K.; Khemakhem, M.; Chaari, I.; Trabelsi, I.; Sabri, A.; Marti, S. Pheromone-
321   mediated mating disruption of *Planococcus ficus* (Hemiptera: Pseudococcidae) in Tunisian vineyards:
322   Effect on insect population dynamics. *Biologia* **2017**, *72*, 333-341, doi:10.1515/biolog-2017-0034.
323   25.   Daane, K.M.; Cooper, M.L.; Triapitsyn, S.V.; Walton, V.M.; Yokota, G.Y.; Haviland, D.R.; Bentley, W.J.;
324   Godfrey, K.E.; Wunderlich, L.R. Vineyard managers and researchers seek sustainable solutions for
325   mealybugs, a changing pest complex. *Calif. Agric.* **2008**, *62*, 167-176, doi:10.3733/ca.v062n04p167.
326   26.   Charles, J.G.; Bell, V.A.; Lo, P.L.; Cole, L.M.; Chhagan, A. Mealybugs (Hemiptera: Pseudococcidae) and
327   their natural enemies in New Zealand vineyards from 1993-2009. *N. Z. Entomol.* **2010**, *33*, 84-91.

27. Abbas, M.S.T. Studies on *Dicrodiplosis manihoti* Harris (Diptera, Cecidomyiidae), a common predator of mealybugs. *J. Pest Sci.* **1999**, *72*, 133-134.

28. Tauber, M.J.; Tauber, C.A.; Daane, K.M.; Hagen, K.S. Commercialization of predators: recent lessons from green lacewings (Neuroptera: Chrysopidae: *Chrysoperla*). *Am. Entomol.* **2000**, *46*, 26-38.

29. Walton, V.M.; Pringle, K.L. A survey of mealybugs and associated natural enemies in vineyards in the Western Cape Province, South Africa. *S. Afr. J. Enol. Vitic.* **2004**, *25*, 23-25.

30. Noyes, J.S.; Hayat, M.S. *Oriental mealybug parasitoids of the Anagyrini (Hymenoptera: Encyrtidae)*; C. A. B. International Press: Wallingford, UK., 1994; pp. 576.

31. Daane, K.M.; Malakar-Kuenen, R.D.; Walton, V.M. Temperature-dependent development of *Anagyrus pseudococci* (Hymenoptera : Encyrtidae) as a parasitoid of the vine mealybug, *Planococcus ficus* (Homoptera: Pseudococcidae). *Biol. Control* **2004**, *31*, 123-132, doi:10.1016/j.biocontrol.2004.04.010.

32. Franco, J.C.; Silva, E.B.; Cortegano, E.; Campos, L.; Branco, M.; Zada, A.; Mendel, Z. Kairomonal response of the parasitoid *Anagyrus* spec. nov near *pseudococci* to the sex pheromone of the vine mealybug. *Entomol. Exp. Appl.* **2008**, *126*, 122-130.

33. Sime, K.R.; Daane, K.M. A comparison of two parasitoids (Hymenoptera: Encyrtidae) of the vine mealybug: Rapid, non-discriminatory oviposition is favored when ants tend the host. *Environ. Entomol.* **2014**, *43*, 995-1002, doi:10.1603/en13192.

34. Mgocheki, N.; Addison, P. Interference of ants (Hymenoptera: Formicidae) with biological control of the vine mealybug *Planococcus ficus* (Signoret) (Hemiptera: Pseudococcidae). *Biol. Control* **2009**, *49*, 180-185, doi:10.1016/j.biocontrol.2009.02.001.

35. Daane, K.M.; Sime, K.R.; Fallon, J.; Cooper, M.L. Impacts of Argentine ants on mealybugs and their natural enemies in California's coastal vineyards. *Ecol. Entomol.* **2007**, *32*, 583-596.

36. Beltra, A.; Navarro-Campos, C.; Calabuig, A.; Estopa, L.; Wackers, F.L.; Pekas, A.; Soto, A. Association between ants (Hymenoptera: Formicidae) and the vine mealybug (Hemiptera: Pseudococcidae) in table-grape vineyards in Eastern Spain. *Pest Manag. Sci.* **2017**, *73*, 2473-2480, doi:10.1002/ps.4640.

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

38. Oki, N.; Komatsu, K.; Sayama, T.; Ishimoto, M.; Takahashi, M.; Takahashi, M. Genetic analysis of antixenosis resitnace to the common cutworm (*Spodoptera litura* Fabricus) and its relationship with pubescence characteristics in soybean (*Glycine max* (L.) Merr.). *Breed. Sci.* **2012**, *61*, 608-217.

39. Jayaraj, S.; Uthamasamy, S. Aspects of insect resistance in crop plants. *Proc. Indian Aca. Sci.* **1999**, *99*, 211-214.

40. Anwar, S.A.; McKenry, M.; Ramming, D. A search for more durable grape rootstock resistance to root-knot nematode. *Am. J. Enol. Vitic.* **2002**, *53*, 19-23.

41. Lider, L.A. Inheritance of resistance to a root-knot nematode (*Meloidogyne incognita* var. *acrita* Chitwood) in *Vitis* spp. *Proceedings of the Helminthological Society of Washington* **1954**, *21*, 53-60.

42. Tertuliano, M.; Dossougbete, S.; Leru, B. Antixenotic and antibiotic components of resistance to the cassava mealybug, *Phenacoccus manihoti* (Homoptera: Pseudococcidae) in various host-plants. *Insect Sci. Appl.* **1993**, *14*, 657-665, doi:10.1017/s1742758400018087.

43. Filho, M.; Grutzmacher, A.D.; Botton, M.; Bertin, A. Biology and fertility life table of *Plannococcus citri* in different vegetative structures of grape cultivars. *Pesqu. Agropecu. Bras.* **2008**, *43*, 941-947.

44. Bertin, A.; Bortoli, L.C.; Botton, M.; Parra, J.R.P. Host plant effects on the development, survival, and reproduction of *Dysmicoccus brevipes* (Hemiptera: Pseudococcidae) on grapevines. *Ann. Entomol. Soc. Am.* **2013**, *106*, 604-609, doi:10.1603/an13030.

45. Hedrick, U.P. Grapes of New York: Report of the New York Agricultural Experiment Station for the Year 1907 II. In *J.B. Lyon Company, State Printers Albany, NY*, 1908; p. 439.

46. Shaner, G.; Finney, R.E. The effect of nitrogen fertilization on the expression of slow-mildewing resistance in Knox wheat. *Phytopathology* **1977**, *67*, 1051-1056.

47. Geiger, C.A.; Daane, K.M. Seasonal movement and distribution of the grape mealybug (Homoptera: Pseudococcidae): Developing a sampling program for San Joaquin Valley vineyards. *J. Econ. Entomol.* **2001**, *94*, 291-301.

48. Japoshvili, G.; Erkilic, L.; Caliskan, A.F.; Kaydan, M.B. Parasitoid complex of Planococcus ficus (Signoret) (Hemiptera: Pseudococcidae) in Turkey. *Phytoparasitica* **2018**, *46*, 481-485, doi:10.1007/s12600-018-0679-1.

49. Cocco, A.; Muscas, E.; Mura, A.; Iodice, A.; Savino, F.; Lentini, A. Influence of mating disruption on the reproductive biology of the vine mealybug, Planococcus ficus (Hemiptera: Pseudococcidae), under field conditions. *Pest Manag. Sci.* **2018**, *74*, 2806-2816, doi:10.1002/ps.5067.

50. Fallahzadeh, M.; Japoshvili, G.; Saghaei, N.; Daane, K.M. Natural enemies of *Planococcus ficus* (Hemiptera: Pseudococcidae) in Fars province vineyards, Iran. *Biocontrol Sci. Technol.* **2011**, *21*, 427-433.

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

52. McKenry, M. Nematological problems in grapevine and use of rootstocks in integrated plant-parasitic nematodes management. In *XXXV Congresso Brasileiro de Nematologia.*, 2018.

53. Cooper, M.L.; Daane, K.M.; Nelson, E.H.; Varela, L.G.; Battany, M.C.; Tsutsui, N.D.; Rust, M.K. Liquid baits control Argentine ants sustainably in coastal vineyards. *Calif. Agric.* **2008**, *62*, 177-183.

54. Guindani, A.N.; Nondillo, A.; da Silva, V.C.P.; Andzeiewski, S.; de Azevedo, W.S.; Bueno, O.C.; Bolton, M. Interaction between *Linepithema micans* (Hymenoptera: Formicidae) and the vine mealybug *Planococcus ficus* (Hemiptera: Pseudococcidae): trophobiosis or predation? *Environ. Entomol.* **2018**, *47*, 1209-1215, doi:10.1093/ee/nvy089.

55. Johnson-Cicalese, J.; Baxendale, F.; Riordan, T.; Heng-Moss, T. Identification of mealybug (Homoprera : Pseudococeidae) resistant turf-type buffalograss germplasm. *J. Econ. Entomol.* **1998**, *91*, 340-346, doi:10.1093/jee/91.1.340.