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# EVALUATING EFFECTIVENESS OF MASS RELEASES OF THE VINE MEALYBUG (*PLANOCOCCUS FICUS*) PARASITOID *COCCIDOXENOIDES PEREGRINUS* IN WESTERN CAPE PROVINCE VINEYARDS, SOUTH AFRICA

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## INTRODUCTION

Chemical applications are currently used to control *P. ficus*. Several attempts of classical biological control have been made with importation and release of *Chrysoplaticyercus splendens* Howard (Joubert, 1943), *Cryptolaemus montrouzieri* Mulsant (Greathead *et al.*, 1971), *Scymnus guttulatus* (Le Conte) and *Scymnus sordidus* (Horn) (Joubert, 1943), *Pseudaphycus angelicus* (Howard), and *Anagyrus pseudococci* (Girault) from Israel (Urban, 1985). In a survey of natural enemies associated with vine mealybug (Urban, 1985), it was found that the parasitoids *Coccidoxenoides peregrinus* (Timberlake), *Anagyrus* spp. and *Leptomastix dactylopii* (Howard) and predatory beetles in the genus *Nephus* were the dominant natural enemies. In addition, it was found that these parasitoids played an important role in biological control of *P. ficus*. However, the level of biological control was not sufficient to keep *P. ficus* infestations below economically acceptable levels.

Biological control by mass releases of natural enemies has contributed to the control of several pseudococcid pests (Mineo and Viggiani, 1977; Longo and Benfatto, 1982; Summy *et al.*, 1986; Smith *et al.*, 1988; Nagarkatti *et al.*, 1992; Smith, 1991; Reddy and Bhat, 1993; Smith *et al.*, 1996; Fronteddu, 1996; Raciti *et al.*, 1997). *Coccidoxenoides peregrinus* is a parasitoid of *P. ficus* (Trjapitsin, 1989), but no reference could be found on biological control of *P. ficus* by mass releases of this parasitoid. However, *P. citri* has been successfully controlled using mass releases of *C. peregrinus* on citrus (Hattingh *et al.*, 1999). This study was conducted to investigate the effectiveness of mass releases of *C. peregrinus* as an alternative to chemical control of *P. ficus*.

## MATERIALS AND METHODS

The nine experimental vineyard locations were divided equally among one table grape area (the Hex River Valley) and two wine grape areas (Stellenbosch and Robertson), all in South Africa. Each vineyard consisted of a release block (1 ha), an adjacent buffer block (1 ha), and a control block (1 ha) adjacent to the buffer block. No pesticide treatments were allowed in the release blocks, except for ant control. Stem barrier treatments of alpha-cypermethrin SC at 20 ml/liter (Table 1) were used for this purpose. All vines and trellis systems were treated with 50 ml of this pesticide (Ueckermann, 1998). Dormant IPM-compatible ant and mealybug treatments (100-200 ml/liter chlorpyrifos two weeks apart before bud burst) were applied in the buffer and control areas (Table 1). These treatments were applied before the first parasitoid releases. The normal fungicide treatments were applied in all blocks.

**Table 1.** Insecticide treatments applied in the nine trial sites

Management Practices	Mealybug Management Programs, by Plot Type		
	Parasitoid Release Areas	Buffer Zones	Control Plots
Chemicals used	No insecticide applications	Dursban EC (chlorpyrifos) 100-200 ml/L	Dursban EC (chlorpyrifos) 100-200 ml/L
Time of treatment		2 weeks before bud burst (September)	2 weeks before bud burst (September)
Method of treatment		broadcast spray of bare vines	
	Ant Management Programs, by Plot Type		
	Parasitoid Release Areas	Buffer Zones	Control Plots
Chemicals used	Fastac (Alpha-cypermethrin) SC at 20 ml/L	Fastac (Alpha-cypermethrin) SC at 20 ml/L	Fastac (Alpha-cypermethrin) SC at 20 ml/L
Time of treatment	Early season (October)	Early season (October)	Early season (October)
Method of treatment	applied to form a stem barrier	applied to form a stem barrier	applied to form a stem barrier

Parasitoids were reared and placed in the field in paper bags by stapling one bag in the crown of the vine. Twenty bags were spaced evenly in an experimental block. Six releases were made at monthly intervals starting in November. Release of  $\pm 20,000$  parasitoids each were made at all sites on November 11 and December 7, 1999; on January 4, February 3, March 8, April 6, November 1, and December 27, 2000; and January 31, February 28, and March 30, 2001.

### Evaluation of Parasitoid Releases

Effectiveness of released parasitoids was evaluated by determining levels of vine mealybug stem infestation, crop loss due to vine mealybug, monthly *C. peregrinus* counts on yellow sticky traps, and monthly percentage parasitism of mealybugs on infested pumpkins deployed in vineyards as baits.

*Stem infestation levels.* Sampling in the blocks was done in 20 evenly spaced plots, each consisting of five vines. A central systematic sampling system was used. The lateral branches of each of these vines were inspected for *P. ficus* in the area closest to the main stem (up to 20 cm from the main stem) where new growth occurred. One basal leaf in the same area was inspected for mealybugs on the same vine. All bunches on the fifth vine in each of these plots were inspected for the presence of *P. ficus*. The proportion of each infested plant part (lateral branches, leaves and bunches) was recorded in each block. Therefore, in each plot, five vines, five leaves, and all bunches on the fifth vine were classified as infested or uninfested. Sampling was conducted throughout the year for two seasons at intervals of one to four weeks depending on the time of year.

*Infestation at harvest (crop loss).* The three assessments of bunch infestation closest to harvest were summed and averaged as an estimate of crop loss for the season.

*Yellow sticky traps.* Yellow sticky traps were used to sample adult *C. peregrinus*. Two sticky traps were used, one on the edge and one in the middle of each trial block. These were left in the field for one month, after which they were replaced.

*Percentage parasitism.* Sampling natural enemies was done on a monthly basis using two mealybug-infested butternut squash, one placed on the edge and one in the middle of each trial block. Each squash bore at least 100 mealybugs at various stages of development. Squash were placed in polystyrene containers with entry holes smeared with petroleum jelly which effectively excluded ants. After exposure in the field, squash were taken to the laboratory and placed in emergence cages and held for one and two months, after which emerged natural enemies were identified and counted. Accuracy of the calculation of percent parasitism was compromised, but this method enabled the identification of natural enemies. Parasitoid identification was done by G. Prinsloo at the Plant Protection Research Institute in Pretoria.

After this period at least 100 mealybugs or mummies were randomly selected on each of the butternut squash samples. Parasitism was determined by looking for mealybugs with emergence holes (mummies) and dissecting the remainder of mealybugs (looking for the remaining live immature parasitoid stages). No eggs and newly emerged crawler stages were used in the determination of percentage parasitism. No counts were made of small hosts that died during the rearing. Percent parasitism (%PA) was estimated using the following formula (Van Driesche, 1983):

$$\%PA = \frac{EMP + LP}{EMP + LP + UMH}$$

where EMP = emerged parasitoid species, LP = all live parasitoid stages, and UMH = unparasitized mealybug hosts.

## ANALYSIS OF DATA

Data from stem infestation, percentage parasitism, and trap catches were transformed by averaging data from two consecutive sampling dates and multiplying by the number of days between these dates. The resultant figures from these were summed to give the total number of insect days (Ruppel 1985). Insect days represented the area under each of the data curves. These data were used in a split plot analysis with the three areas as the main plots and treatments and years as the main effects in the sub-plots.

Data pertaining to percentage crop loss were analysed in the same way.

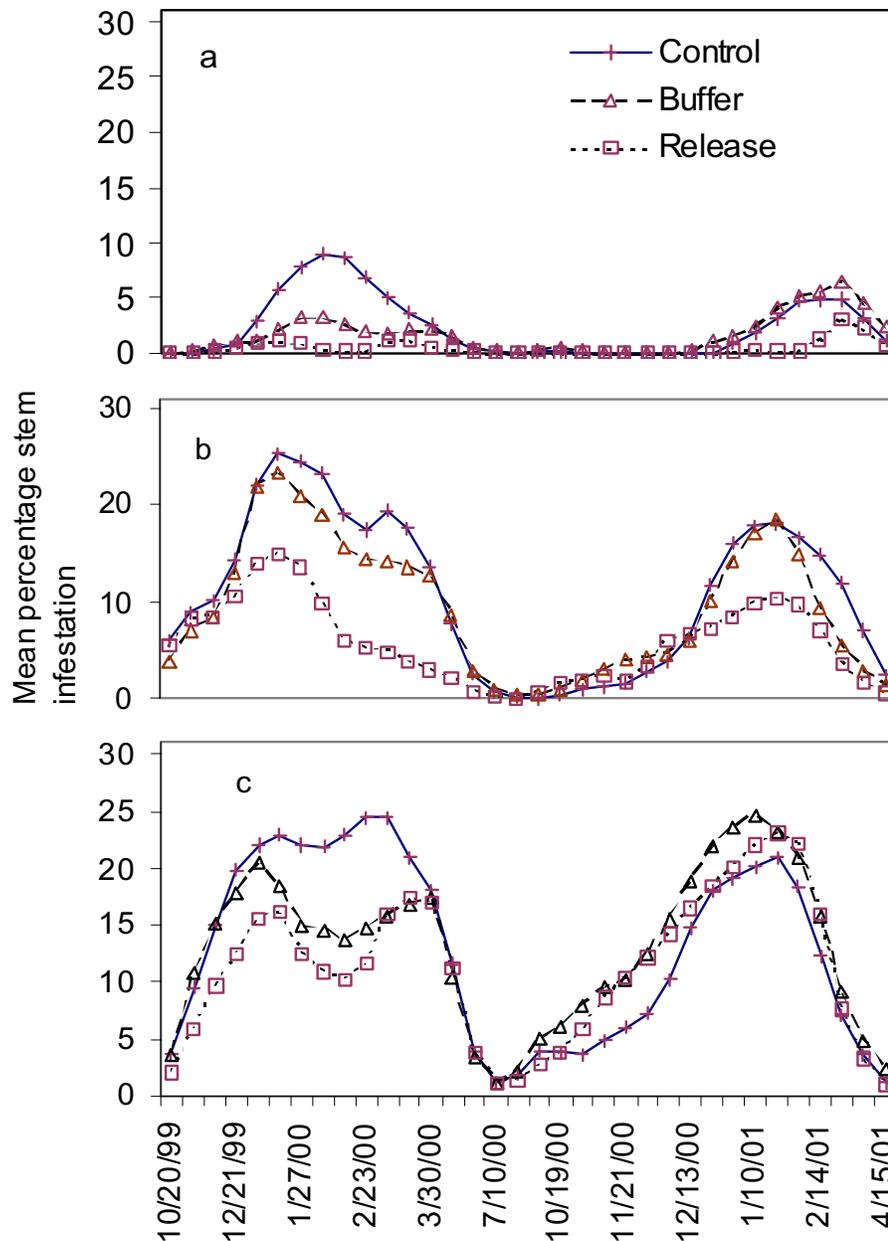
## RESULTS

### Stem Infestation

Stem infestation by *P. ficus* was lower in the Hex River Valley than in Stellenbosch and Robertson ( $P < 0.01$ ) (Fig. 1).

### Infestation at Harvest (Crop Loss)

There were significant differences in *P. ficus* bunch infestations ( $P < 0.01$ ) (Table 2) between treatments. There were also differences between areas with less crop loss due to *P. ficus* infestations in the Hex River Valley than in Stellenbosch and Robertson ( $P < 0.01$ ) (Fig. 1; Table 2). There were also interactions between areas and treatments ( $P < 0.01$ ) with less crop loss due to *P. ficus* infestations in the Hex River Valley than in Stellenbosch and Robertson (Table 4).



**Figure 1.** Average stem infestations by *Planococcus ficus* during two seasons in blocks into which *Coccidoxenoides peregrinus* was released and in buffer and control blocks in three vineyards in the Hex River Valley (a); Stellenbosch (b); Robertson (c).

### Yellow Sticky Traps

There was no significant difference in the number of *C. peregrinus* caught on yellow sticky traps between the three areas ( $P = 0.21$ ) or between the treatments ( $P = 0.19$ ). There were differences between seasons ( $P < 0.01$ ). More parasitoids were caught on the yellow sticky traps during the second season than during the first (Table 3, Fig. 2). The differences were not as marked in the Hex River Valley as in the Stellenbosch and Robertson areas. This discrepancy resulted in interactions between area and season ( $P < 0.01$ ) (Table 3, Fig. 2).

**Table 2.** Mean percentage crop loss in release, buffer and control vineyards due to vine mealybug (*Planococcus ficus*) infestation at harvest in three grape growing areas during two seasons

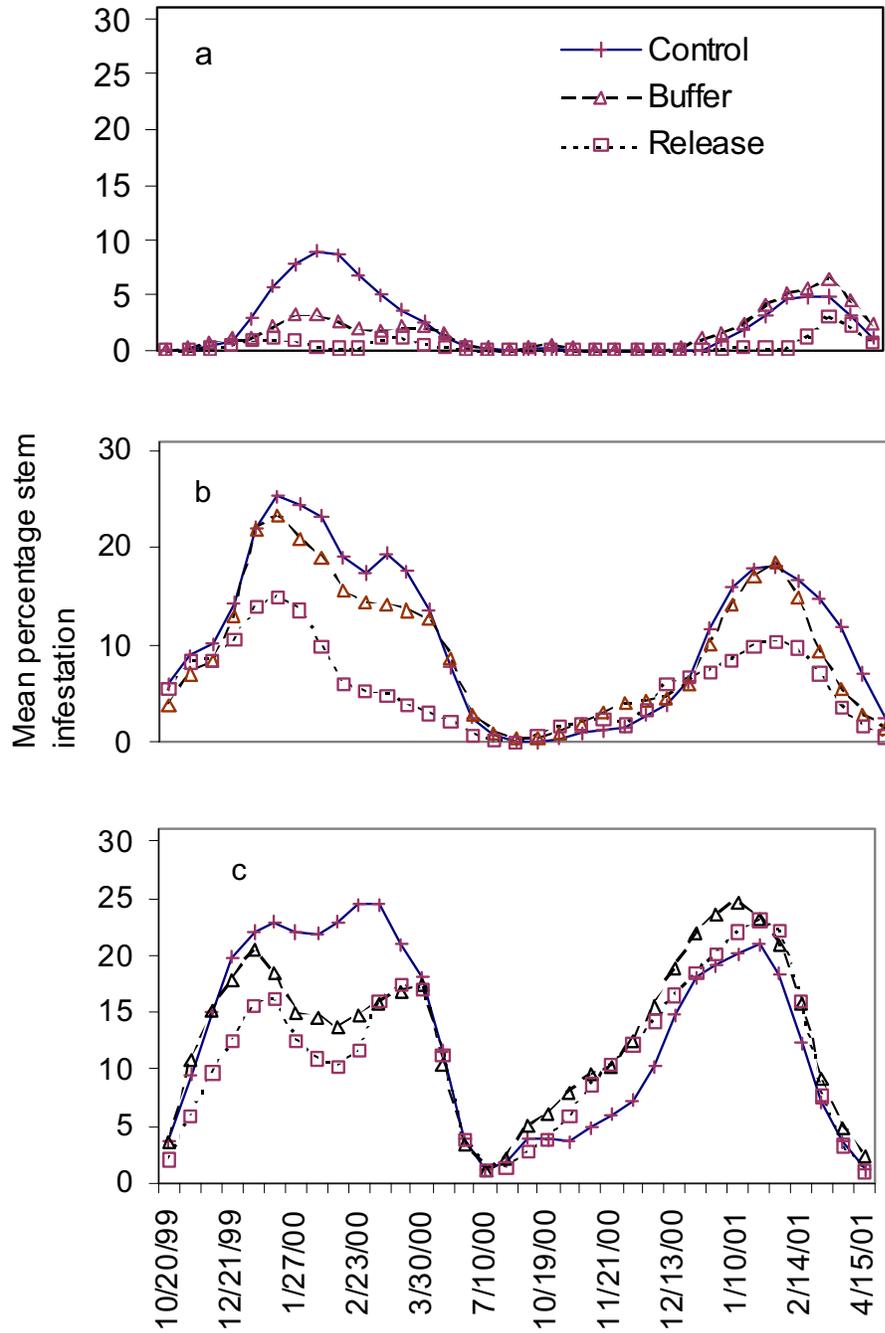
Grape growing area	Mean % crop loss					
	Control		Buffer		Release	
	1999-2000	2000-2001	1999-2000	2000-2001	1999-2000	2000-2001
Hex River	2.3	0.03	1.11	0.03	0.05	0
Stellenbosch	8.6	7.3	4.05	3.9	6.5	5.8
Robertson	8.5	8.2	8.01	8.5	6.9	8.3
Average	6.47	5.18	4.39	4.14	4.48	4.7

**Table 3** Cumulative yellow sticky trap counts of *Coccidoxenoides peregrinus* starting from September and ending in April during 1999-2000 and 2000-2001

Area	Control		Buffer		Release	
	1999-2000	2000-2001	1999-2000	2000-2001	1999-2000	2000-2001
Hex River	22	31	51	63	44	76
Robertson	1	136	18	264	18	332
Stellenbosch	55	50	84	169	93	179
Total	78	217	153	496	115	587

**Table 4.** Average percentage *Planococcus ficus* parasitism for the control, buffer and release treatments during the two seasons

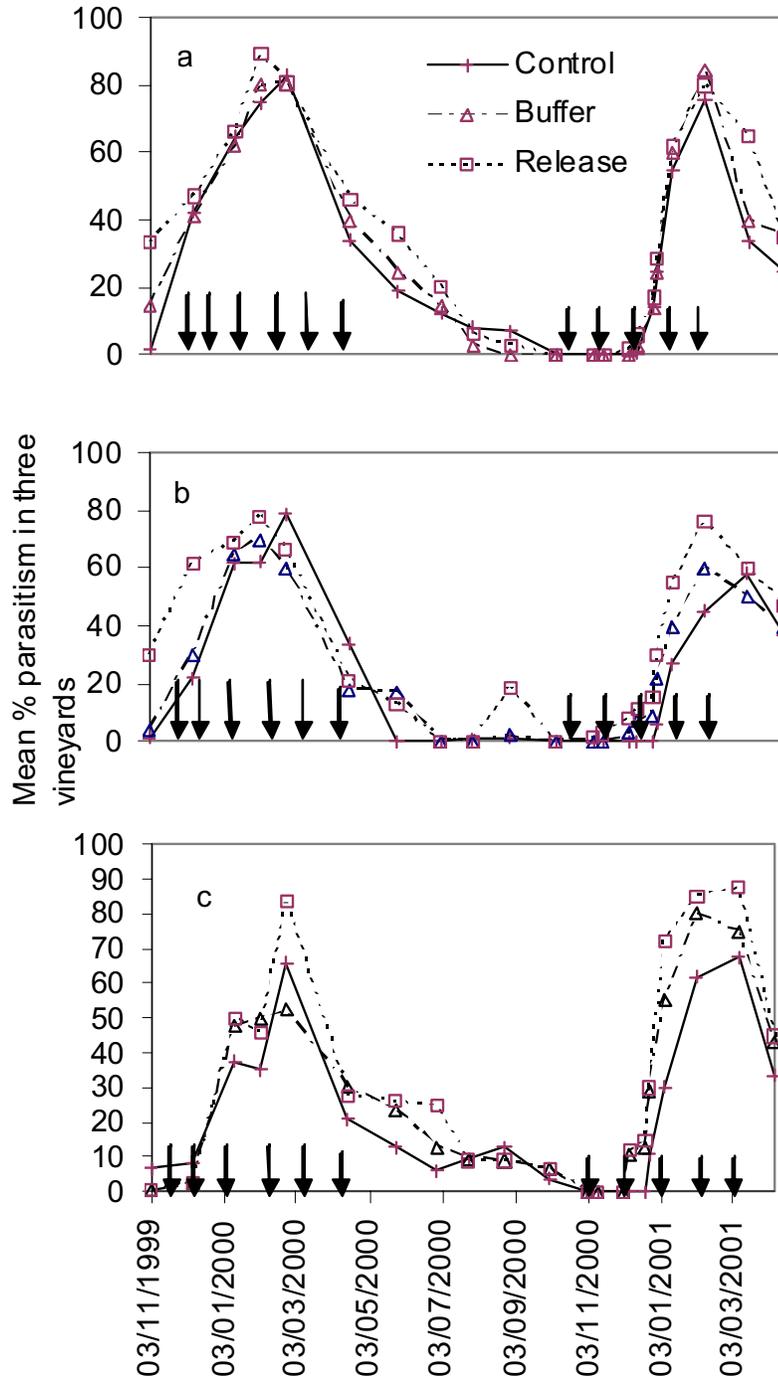
Area	Control		Buffer		Release	
	1999-2000	2000-2001	1999-2000	2000-2001	1999-2000	2000-2001
Hex River	35.4	25.4	42.4	32	44.3	32.7
Robertson	28.7	19.2	37.2	31.2	37.6	32.7
Stellenbosch	20.7	22.6	25.2	37.9	26.2	38.4
Total average	28.3	22.4	34.9	33.7	36	34.6



**Figure 2.** Average number of *Coccidoxenoides peregrinus* caught on yellow sticky traps during two seasons in blocks in which *C. peregrinus* was released and buffer and control blocks in three vineyards in (a), Hex River Valley; (b), Stellenbosch; (c), Robertson. Arrows indicate the release of 20000 *C. peregrinus*/ha.

### Percentage parasitism

There were no differences in percent parasitism between areas ( $P = 0.55$ ) or treatments ( $P = 0.35$ ). There was a difference between years ( $P < 0.01$ ), with a slightly higher percent parasitism during the first season than during the second (Fig. 3).



**Figure 3.** Percentage parasitism during two seasons in blocks where *Coccidoxenoides peregrinus* was released and buffer and control blocks in three vineyards in Hex River Valley (a); Stellenbosch (b); and Robertson (c). Arrows indicate a release of 20,000 *C. peregrinus* per hectare.

## DISCUSSION

No differences were detected in the percentage *P. ficus* stem or bunch infestation, *C. peregrinus* counts on yellow sticky traps, or percentage parasitism among the release, buffer, and control blocks. The large plot size meant that the treatments might not have been increasing the variance and, therefore, the experimental error. The large plots also made it logistically difficult to increase the number of replicates, which would have increased the degrees of freedom, providing more sensitive tests. In addition the large plot size may have meant that the treatments were ecologically heterogeneous, increasing the experimental error.

*Planococcus ficus* stem infestation in the Hex River Valley was significantly lower than in Robertson and Stellenbosch. The lower stem infestations in the Hex River Valley did not influence the number of parasites caught or percent parasitism. Generally *P. ficus* stem infestation levels remained lower in the release than in the buffer and control blocks during both seasons in all areas (Fig. 1), although this was not reflected in the formal analysis. This may indicate that the additional released *C. peregrinus* aided biological control in the release blocks.

*Planococcus ficus* bunch infestations at harvest (Table 2) in the release and buffer treatments were lower than in the control, suggesting that the releases limited crop loss to a greater extent than the chemical control program. The average crop loss in the buffer and release plots was similar. Therefore, it appeared that the buffer plots also benefited from the parasitoid releases. The higher numbers of *C. peregrinus* and percentage parasitism recorded in the release and buffer blocks may therefore have resulted in the lower overall bunch infestation levels in these treatments at the end of the season.

More *C. peregrinus* were caught on yellow sticky traps in the release and buffer blocks than in the control in all three areas (Fig. 2). However, no differences among treatments were detected by the formal analysis. The higher numbers of *C. peregrinus* in release and buffer blocks in all three areas may have added to biological control as was found by Smith *et al.* (1988) using early mass releases of *L. dactylopii* against *P. citri*. The slightly higher *C. peregrinus* counts in the buffer blocks than in the control could be explained by gradual movement of the parasitoid to the surrounding areas over time. Continued higher counts of *C. peregrinus* were made in all release areas compared with the buffer and control blocks throughout the season. Therefore, it appeared as if the releases successfully supplemented naturally occurring *C. peregrinus* populations.

No significant differences were found between percentage parasitism between any of the grapegrowing areas. The general trend in all three areas was that of a higher percent parasitism in the release and buffer blocks (Table 4, Fig. 3) than in the control blocks.

Mass releases of *C. peregrinus* controlled the pest adequately in the Hex River Valley. The low infestation levels of *P. ficus* appeared to be more suitable for biological control than the high *P. ficus* infestation encountered in Robertson and Stellenbosch.

In Stellenbosch and Robertson a measure of control was evident but not sufficient to keep *P. ficus* populations below economic injury levels. High initial *P. ficus* infestation levels appeared to be less suitable for biological control. Future strategies should include more effective ant control by chemical stem barrier treatments, and initial suppression of high mealybug population levels through the use of dormant season chemical treatments.

Mass releases of *C. peregrinus* in all three areas resulted in *P. ficus* control similar to that achieved using chemical sprays. This method of control is therefore at least as effective as chemical control. The main problem encountered in the use of this strategy in the Hex River Valley was the high cost and lack of available high quality parasitoids. Risks using this method of control include the injudicious use of chemicals during the release period, the lack of ant control, and lack of technical support.

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