# DEVELOPMENT OF AN INTEGRATED PEST MANAGEMENT SYSTEM FOR VINE MEALYBUG, *PLANOCOCCUS FICUS* (SIGNORET), IN VINEYARDS IN THE WESTERN CAPE PROVINCE, SOUTH AFRICA.

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Dissertation submitted in partial fulfillment of the requirements for the degree of Doctor in Philosophy at the University of Stellenbosch.

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# **DECLARATION**

I, the undersigned, hereby declare that the work contained in this dissertation is my own original work and that I have not previously in its entirety or in part submitted it at any university for a degree.

#### **ABSTRACT**

A survey was conducted in the Western Cape Province during the 1999/2000 and 2000/2001 seasons on mealybugs occurring in vineyards. *Planococcus ficus* (Signoret) was the dominant mealybug in vineyards during this time.

During this study *P. ficus* was recorded for the first time on roots of grapevines, which has far reaching implications for the control of this important vine leafroll virus vector as control actions were focused on above ground control. Other mealybugs presently recorded in local vineyards included *Pseudococcus longispinus* (Targioni) and *Ferrisia malvastra* (McDaniel). *Pseudococcus viburni* (Maskell) and *Ps. solani* Ferris were found on weeds in vineyards. Natural enemies of *P. ficus* recorded most frequently were species of *Nephus* predatory beetles, and the parasitoids *Coccidoxenoides peregrinus* (Timberlake), *Anagyrus* sp. and *Leptomastix dactylopii* (Howard).

Developmental studies on P. ficus and C. peregrinus indicated that the intrinsic rate of increase  $(r_m)$  was similar, peaking at 25°C  $(r_m = 0.169 \text{ for } P)$ . ficus;  $r_m = 0.149 \text{ for } C$ . peregrinus). The net replacement rate  $(R_0)$  was higher for P. ficus than for C. peregrinus at all five temperatures tested. The  $R_0$  for P. ficus reached a maximum at 21°C (308.87) and C. peregrinus at 25°C for C. peregrinus (69.94). The lower and upper thresholds for development of P. ficus were estimated at 16.59 and 35.61°C respectively. The lower threshold for development of C. peregrinus was 8.85°C. These parameters indicated that both insects were well adapted to temperatures in the Western Cape

Province. The lower minimum threshold temperature of *C. peregrinus* in relation to that of *P. ficus* suggests that *C. peregrinus* should be more active during winter and early spring than *P. ficus*.

A central systematic presence-absence sampling system was developed for *P. ficus*. Monitoring three different plant parts on the vine indicated that new growth areas on vines adjacent to the main stem could serve as an early warning system for pending *P. ficus* bunch infestations. Intervention should be planned when 2 % of the stems are infested with *P. ficus* when using this system.

Seasonal population studies of *P. ficus* and its natural enemies showed that stem infestation by *P. ficus* reached peak levels during January in Robertson and Stellenbosch and during February in the Hex River Valley. Vine mealybugs colonised new growth early in the season, followed by the leaves and eventually the bunches towards the end of the season. High stem infestations early in the season resulted in high bunch infestation levels at harvest. A density dependent relationship was evident between *P. ficus* populations and parasitoid populations, suggesting that the parasitoids played a mayor role in the biological control of *P. ficus* populations. Biological control was however only achieved towards the end of the season when damage to the crop had already occurred.

Mass releases of *C. peregrinus* on *P. ficus* populations were done in order to augment biological control as an alternative to chemical control. Between five

and six releases of 20 000 *C. peregrinus* per release were done at monthly intervals in three grapegrowing areas. Mass released *C. peregrinus* controlled *P. ficus* adequately in the Hex River Valley. Control of *P. ficus* using this approach was no worse than using chemical control in Robertson and Stellenbosch. *C. peregrinus* is commercially available and can therefore be used as an alternative to chemical control by producers.

Degree day estimation was used to predict development of *P. ficus* populations. This information was used as an input in a *P. ficus* pest management model. Data acquired from *P. ficus* and ant monitoring were used as components to construct a decision chart. This chart can be used by producers to optimise the control of *P. ficus* populations using either chemical control or mass releases of *C. peregrinus*.

#### **OPSOMMING**

'n Studie is gedurende die 1999/2000 en 2000/2001 seisoene gedoen met die doel om die witluisspesies wat in wingerde voorkom, te identifiseer.

Planococcus ficus (Signoret) is tans die dominante witluisspesie in wingerde in die Wes Kaap Provinsie. P. ficus kolonies is op wingerdwortels gevind.

Dié bevinding kan verreikende gevolge hê vir die beheer van dié plaag as 'n belangrike rolbladvirus vektor aangesien beheer tot dusver gefokus het op bogrondse gedeeltes. Ander witluisspesies wat in wingerde gevind is, sluit in Pseudococcus longispinus (Targioni) en Ferrisia malvastra (McDaniel).

Pseudococcus viburni (Maskell) en Ps. solani Ferris is op onkruide in wingerde gevind. Dominante natuurlike vyande van P. ficus sluit predatoriese kewertjies van verskeie Nephus spp. en die parasitoïede Coccidoxenoides peregrinus (Timberlake), Anagyrus sp. en Leptomastix dactylopii (Howard) in.

Ontwikkelingstudies op *P. ficus* en *C. peregrinus* het aangetoon dat die inhirente voortplantingstempo (r<sub>m</sub>) soortgelyk was vir beide insekte met 'n maksimum by 25°C (0.169 vir *P. ficus*, 0.149 vir *C. peregrinus*). Die netto vervangingstempo (R<sub>o</sub>) was in vergelyking met *C. peregrinus* hoër vir *P. ficus* by al vyf temperature getoets. Die R<sub>o</sub> van *P. ficus* het 'n maksimum bereik teen 21°C (308.87) en die van *C. peregrinus* by 25°C (69.94). Die teoretiese hoër en laer drempels vir ontwikkeling van *P. ficus* was onderskeidelik 16.59 en 35.61°C. Die teoretiese laer drempelwaarde van ontwikkeling vir *C. peregrinus* was 8.85°C. Hierdie parameters dui aan dat beide insekte goed aangepas is by temperature in die Wes Kaap Provinsie. Die laer minimum

drempel vir ontwikkeling van *C. peregrinus* in verhouding tot *P. ficus* impliseer dat *C. peregrinus* in die winter en vroeë lente meer aktief sal wees as *P. ficus*.

'n Sentrale sistematiese aan-afwesig moniteringsisteem met bekende vlakke van steekproefnemingsfout is ontwikkel in kommersiële wingerde vir *P. ficus*. Monitering van drie verskillende dele op die wingerdstok het aangedui dat die nuwe groei areas kan dien as 'n vroeë waarskuwing vir latere *P. ficus* trosinfestasies. Dié sisteem sal produsente in staat stel om te bepaal wanneer optrede noodsaaklik is. Daar word voorgestel dat optrede noodsaaklik is by 'n *P. ficus* besmettingsvlak van 2 % op die nuwe groei areas op stokke.

Stambesmetting deur *P. ficus* het in Januarie piekvlakke bereik in Stellenbosch en Robertson, en in Februarie in die Hex Rivier Vallei. *P. ficus* koloniseer nuwe groei vroeg in die seisoen waarna blare en trosse aan die einde van die seisoen gekoloniseer word. Dié data dui aan dat *P. ficus* besmetting op nuwe groei vroeg in die seisoen 'n aanduiding kan gee van hoë trosbesmetting aan die einde van die seisoen. 'n Digtheidsafhanklike verwantskap was waarneembaar tussen *P. ficus* plaagpopulasies en parasitoïed populasies. Dié verwantskap dui aan dat parasitoïede die belangrikste rol speel in biologiese beheer van *P. ficus* populasies.

Biologiese beheer van witluis is egter eers aan die einde van die seisoen bereik toe die oes reeds beskadig was.

Massavrylatings van *C. peregrinus* is in *P. ficus* besmette blokke gedoen om biologiese beheer aan te help en sodoende as alternatief tot chemiese beheer

te dien. Tussen vyf en ses vrylatings met 20 000 *C. peregrinus* is een keer per maand gedurende die seisoen gedoen. Die vrygelate *C. peregrinus* het *P. ficus* populasies voldoende beheer in die Hex Rivier Vallei. Beheer van *P. ficus* deur massavrylatings van *C. peregrinus* was soortgelyk as chemiese beheer in Robertson en Stellenbosch. *C. peregrinus* is kommersieel beskikbaar en kan om hierdie rede as alternatief tot chemiese beheer gebruik word.

Graaddag bepaling is gebruik om die ontwikkeling van *P. ficus* populasies te voorspel. Hierdie inligting is gebruik as 'n verdere hulpmiddel in die *P. ficus* plaagbeheermodel. Inligting verkry vanuit *P. ficus* en mier monitering is gebruik as komponente in die opstel van 'n besluitnemingstabel. Hierdie tabel kan gebruik word deur produsente om beheer van *P. ficus* plaagpopulasies te optimaliseer deur chemiese beheer of massavrylatings van *C. peregrinus*.

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#### **CHAPTER 1**

### INTRODUCTION AND LITERATURE REVIEW

## 1.1 HISTORY OF THE PEST IN SOUTH AFRICA

Planococcus ficus (Signoret) was initially identified in the Western Cape Province as Planococcus citri (Risso) by Joubert (1943), Kriegler (1954) and Whitehead (1957) after introduction to the area, probably with plant material. De Lotto (1975) subsequently identified it as Planococcus ficus. The most recent samples of the insect collected during 1999/2000 were identified as Planococcus ficus (Signoret) by I.M. Millar, Plant Protection Research Institute in Pretoria. It was recorded by Joubert (1943) in the Boland during 1930. By 1935 P. ficus had spread to the Hex River Valley and subsequently to all other major grape producing areas (Joubert 1943) in this region. Kriegler (1954) regarded it as one of the most important pests of the grape industry in South Africa. Other pseudococcid species recorded from vines in the Western Cape Province included Pseudococcus longispinus (Targioni) and Ferrisia malvastra (McDaniel) also identified by I.M. Millar, Plant Protection Research Institute in Pretoria. However, they had as yet not attained pest status on grapes in the Western Cape Province.

#### 1.2 TAXONOMIC STATUS

The most recent classification was done by Ben-Dov (1994) who classified *P. ficus* in the Order Hemiptera, Suborder Homoptera, Coccoidea and

Pseudococcidae. The species was well described by De Lotto (1975), Cox (1981, 1989) and Williams & Granara de Willink (1992). Keys for the female of this species were given in Williams & Moghaddam (1999) (Iran), Williams & Granara de Willink (1992) (Central and South America), Cox (1989) (World), Cox & Ben-Dov (1986) (Mediterranean basin) and Cox & Wetton (1988) (West Indies). *P. ficus* was initially described as *Coccus vitis* by Nedzilskiii (1869) (Cox & Ben-Dov 1986). Lichtenstein (1870) subsequently placed this species in *Dactylopius* (Cox 1989). Signoret (1875) described it as *Planococcus ficus*. Thereafter various synonyms were used, many of which were the result of misidentification (Ben-Dov 1994) (Table 1.1).

TABLE 1.1. Synonyms, used for Planococcus ficus (Ben-Dov 1994).

Synonym	Author	Comment
Coccus vitis	Nedzilskii (1869),	Incorrect due to
	Lindinger (1912),	misidentification (Cox &
	Borchsenius (1942)	Ben-Dov 1986). True
		identity unknown.
Dactylopius vitis	Lichtenstein (1870),	Misidentification (Cox
	Signoret (1895)	1989)
Dactylopius ficus	Signoret (1875),	Type material lost (Ben-
	Borchsenius (1949)	Dov & Matile-Ferrero
		1995).
Dactylopius subterraneus	Hempel (1901)	On roots of cultivated
		grapes
Pseudococcus ficus	Fernald (1903)	Change of combination

Table 1.1 continued

Synonym	Author	Comment
Pseudococcus vitis	Fernald (1903), Leonardi (1920), Bodenheimer (1924)	-
Pseudococcus citriodes	Ferris (1922)	New name
Pseudococcus citri	Balachowsky & Mesnil (1935)	Misidentification
Dactylopius ficus	Borchsenius (1949)	Synonymised with  Pseudococcus citri  (Risso)
Planococcus citroides	Ferris (1950)	Change of combination
Planococcus vitis	Ezzat & McConnell (1956), Matile-Ferrero (1984)	
Planococcus ficus	Ezzat & McConnell (1956)	Change of combination
Pseudococcus praetermissus	Ezzat (1962)	Synonym

# 1.3 VERNACULAR NAMES

Vernacular names given by Balachowsky & Mesnil (1935) include 'cocciniglia farinosa della vite', 'cochonilha algodeo da vinha', 'cotonet de la vid', 'grapevine mealybug', 'la cochenille farineuse de la vigne'. Berlinger (1977) described P. ficus as the 'Mediterranean vine mealybug', Bodenheimer (1924) as 'subterranean vine mealy bug' and De Lotto (1975) as 'vine mealybug'.

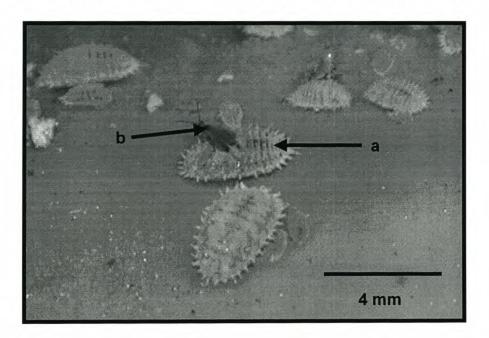
## 1.4 MORPHOMETRICS

Criteria for age distinction of the different developmental stages of *P. ficus* were described by Kriegler (1954). This information was used in studies on the developmental biology of this pest (Chapter 4). Kriegler (1954) made use of a combination of colour, size and other characteristics to distinguish between the different stages. Certain criteria were selected and presented in Table 1.2.

**TABLE 1.2**. Morphometric characters for distinguishing life stages of *P. ficus* (Kriegler 1954) in developmental biology studies (Chapter 4).

Stage	Average length (mm)	Average width (mm)	Characteristics/Colour
Egg	0.41	0.21	Light straw
First nymphal instar	0.46	0.22	Light to dark yellow, six antennal segments
Second nymphal instar	0.68	0.35	Yellowish brown
Third nymphal instar	1.13	0.66	Seven antennal segments
Male prepupa	0.95		One pair of lateral ocelli. Visible wingbuds
Male pupa	1.05		Three pairs of lateral ocelli. Wingbuds reaching to third abdominal segment
Adult male	1.05		Wings fully developed
Adult female	1.69	0.99	Wingless, eight antennal segments

In the survey work (Chapter 3) it was concluded that *P. ficus* was the dominant mealybug species in vineyards. Adult female mealybugs were approximately 4 mm in length, slightly more than 2 mm wide and about 1.5 mm thick. The adult female and immature stages were ovate, humpbacked, light slate- to flesh-coloured and covered by a fine, white powdery wax secretion which was more evident on the later stages. The body of the adult female was clearly segmented, and had a fringe of short, fingerlike wax filaments around its edge (Kriegler 1954) (Fig. 1.1). After mating egg sacs covered by waxy threads started to appear.



**Fig. 1.1**. Adult female (indicated by arrow a) and male (indicated by arrow b) *P. ficus*.

This species was easily distinguished from *Ps. longispinus* which was about 3 mm long, 1 mm wide, ovate and yellowish grey in colour. Adult females and

younger stages of this species had exceptionally long posterior filaments and no egg sacs as this species was ovoviviparous (El-Minshawy et al. 1974). A single adult female Ferrisia malvastra (McDaniel) 7 mm long and 4 mm wide with a light orange colour was for the first time recorded from a vineyard in Stellenbosch (Chapter 3).

P. ficus was misidentified by several authors as mentioned earlier in this chapter. The main reasons for this was because of the lack in qualitative characteristics (De Lotto 1975) which could be used to differentiate between this and other closely related species such as P. citri. Identification was based on minor differences in the number and arrangement of glandular ducts of the dermis. P. ficus was found to have fewer groups and smaller ducts than P. citri (De Lotto 1975). Other less apparent differences between these species were described by De Lotto (1975). However, P. citri has not yet been found on vines in South Africa.

## 1.5 LIFE CYCLE

Kriegler (1954) studied the lifecycle of *P. ficus* in detail. Developmental stages studied were eggs, first, second and third nymphal instars. The male characteristics appeared after the third nymphal instar. During subsequent development, differentiation between the sexes occurred. In the case of the male, the prepupa stage was followed by the pupa from which the winged male emerged (Fig. 1.1). Males were characterised by long filamentous anal setae

and no mouthparts (Kriegler 1954). The adult female started releasing pheromones at sexual maturity, attracting adult males for copulation (Hinkens *et al.* 2001). Subsequent to copulation there was a pre-oviposition period, after which the female layed eggs in an egg sac made up of filamentous waxy hairs. Kriegler (1954) recorded an average of 362 eggs per female.

## 1.6 HOSTS

P. ficus is a polyphagous insect and apart from the economic damage on Vitis vinifera Linn. it has been found on various other host plants (Table 1.3).

**TABLE 1.3**. Recorded findings of *P. ficus* on host plants other than *V. vinifera*.

Family	Genus/Species	Reference
Anacardiaceae	Mangifera indica Blume	Ezzat & McConnel (1956), Cox
		(1989), Ben-Dov (1994)
Apocynaceae	Nerium oleander Linn.	Ezzat & McConnel (1956)
Asteraceae	Dahlia spp.	Ezzat & McConnel (1956)
Juglandaceae	Juglans spp.	Ezzat & McConnel (1956)
Lauraceae	Persea americana Mill.	Cox (1989), Ben-Dov (1994)
Labiaceae	Dichrostachys glomerata	Cox (1989), Ben-Dov (1994)
	Linn.	
	Prosopis farcata Linn.	Cox (1989), Ben-Dov (1994)
	Tephrosia purpurea Pers.	Cox (1989), Ben-Dov (1994)
Moraceae	Ficus benjamina Linn.	Williams & Granara de Willink
		(1992), Ben-Dov (1994)

Table 1.3 continued

Family	Genus/Species	Reference
Palmae	Phoenix dactylifera Linn.	Cox (1989),
		Ben-Dov (1994)
Platanaceae	Platanus orientalis Linn.	Martin-Mateo (1985), Williams &
		Moghaddam (1999)
Poaceae	Bambusa spp.	Ezzat & McConnel (1956)
Rhamnaceae	Zizyphus spina-christi Georgi	Cox (1989), Ben-Dov (1994)
Rosaceae	Cydonia oblonga Mill.	Granara de Willink et al. (1997)
Rosaceae	Malus domestica Baumg.	Granara de Willink et al. (1997)
	Malus pumila Mill.	Cox (1989),
		Ben-Dov (1994)
Salicaceae	Salix spp.	Cox (1989), Ben-Dov (1994)
Sterculiaceae	Theobroma cacao Linn.	Ezzat & McConnel (1956).
Styracaceae	Styrax officinalis Walt.	Cox (1989), Ben-Dov (1994)

None of the above host plants were found in close proximity to the vineyards sampled in the present study. A variety of weeds was, however, sampled for mealybugs in vineyards during the current study but no *P. ficus* were found on any of them (Chapter 5).

# 1.7 GEOGRAPHICAL DISTRIBUTION AND ECONOMIC IMPORTANCE

P. ficus has been found in most grape production areas throughout the world and caused economic damage (Table 1.4). P. ficus is of particular economic importance on grapevines in the Mediterranean region, South Africa, Pakistan and Argentina (Ben-Dov 1994).

**TABLE 1.4.** Geographical areas where *Planococcus ficus* has been recorded on vines (Ben-Dov 1994).

Geographical	Reference
area	
Afrotropical:	Ezzat & McConnel (1956), De Lotto (1975), Cox (1989), Ben-
South Africa	Dov (1994)
Mauritius	Ezzat & McConnel (1956)
Nearctic:	Ezzat & McConnel (1956)
United States of	
America	
Neotropical:	Hempel (1901), Ezzat & McConnel (1956), , Granara de
Argentina	Willink (1991), Williams & Granara de Willink (1992), Ben-
	Dov (1994), Trjapitzyn & Trjapitzyn (1999)
Brazil	Williams & Granara de Willink (1992), Ben-Dov (1994)
Chile	Ezzat & McConnel (1956)
Dominican	Ezzat & McConnel (1956)
Republic	
Trinidad and	Ezzat & McConnel (1956)
Tobago	
Uruguay	Granara de Willink et al. (1997)
Oriental:	Varshney (1992), Ben-Dov (1994)
India	
Pakistan	Cox (1989), Ben-Dov (1994)
Palearctic:	Kozár, Fowjhan & Zarrabi (1996)
Afghanistan	
Azerbaijan	Rzaeva (1985), Ben-Dov (1994)
Azores	Ezzat & McConnel (1956)
Canary Islands	Carnero Hernandez & Pérez Guera (1986),
	Pérez Guerra & Carnero Hernandez (1987), Ben-Dov (1994)

Table 1.4. continued

Geographical	Reference	
area		
Palearctic:	Argyriou (1983), Cox (1989), Ben-Dov (1994)	
Crete		
Cyprus	Cox (1989), Ben-Dov (1994)	
Egypt	Ezzat & McConnel (1956), Ezzat & Nada (1987), Cox (1989),	
	Ben-Dov (1994)	
France	Signoret (1875), Ben-Dov (1994)	
Greece	Ezzat & McConnel (1956)	
Hyeres Islands	Foldi (2000)	
Iran	Cox (1989), Ben-Dov (1994), Kozar, Fowjhan & Zarrabi	
	(1996), Williams & Moghaddam (1999)	
Iraq	Cox (1989), Ben-Dov (1994)	
Israel	Bodenheimer (1924), Avidov (1961), Avidov & Harpaz 1969),	
	Cox & Ben-Dov (1986), Ben-Dov (1994)	
Italy	Leonardi (1920), Tranfaglia (1976), Marotta (1987),	
	Rosciglione & Castellano (1985), Duso (1990), Ben-Dov	
	(1994)	
Lebanon	Cox (1989), Ben-Dov (1994)	
Libya	Ferris (1922), Ben-Dov (1994)	
Portugal	Ezzat & McConnel (1956)	
Sardinia	Melis (1930), Ben-Dov (1994), Longo et al. (1995), Pellizzari-	
	Scaltriti & Fontana (1996)	
Saudi Arabia	Beccari (1971), Matile-Ferrero (1984), Ben-Dov (1994)	
Sicily	Longo et al. (1995), Russo & Mazzeo (1997)	
Spain	Gómez-Menor Ortega (1937), Ezzat & McConnel (1956),	
	Martin-Malteo (1985), Ben-Dov (1994)	
Syria	Ezzat & McConnel (1956)	
Tunisia	Cox (1989), Ben-Dov (1994)	
Turkmenistan	Achangelskaya (1930), Ben-Dov (1994)	

Engelbrecht & Kasdorf (1985) and Cabaleiro & Segura (1997) found that *P. ficus* transmitted the grapevine leafroll associated virus 3 (GLRa V-3). Initially, the mealybug specimens studied by Cabaleiro & Segura (1997) were identified as *Planococcus citri* (Risso) but later identified by Ben-Dov as *P. ficus* (Signoret) (Yair Ben-Dov, unpublished data, July 1998). Transmission of GLRa V-3 by *P. ficus* and positive identification of GLRa V-3 was further confirmed using PCR methods by Acheche *et al.* (1999).

The transfer of the vine leafroll virus caused inefficient photosynthesis which resulted in reduced fruit production, inability to produce sufficient sugar and higher than normal acidity levels, delaying harvest. In addition, infested vines were less drought resistant (Cabaleiro *et al.* 1999; Manini 2000). Manini (2000) showed that uninfected seedlings showed increased vegetative vigour and higher propagation potential than infected seedlings. In addition, *P. ficus* has been found to be a vector of corky-bark disease virus in vines (Engelbrecht & Kasdorf 1985; Tanne *et al.* 1989) and Shiraz disease (Engelbrecht & Kasdorf 1985).

Apart from being a vector of GLRa V-3, high infestations of *P. ficus* infested table grape bunches resulting in direct crop loss and progressive weakening of vines through early leaf loss (Kriegler 1954; Whitehead 1957; Berlinger 1977; Charles 1982).

1.8 SEASONAL POPULATION DYNAMICS, PHENOLOGY AND INFLUENCE OF TEMPERATURE ON *P. FICUS* 

Kriegler (1954) and Whitehead (1957) studied the population dynamics and seasonal abundance of *P. ficus* in South Africa. Berlinger (1977) did similar studies in Israel and Duso (1990) in Italy. Kriegler (1954) found that there were six generations during the year in which he studied this insect while Duso (1990) recorded three generations a year.

Upward migration on the trunk began from spring or early summer (October in South Africa, March/April in Israel and Italy) (Kriegler 1954, Berlinger 1977, Duso 1990). Populations started to develop on new growth and the population peak was recorded between the end of January and the beginning of February, after which numbers declined (Kriegler 1954, Whitehead 1957). Mealybugs found in the vine canopy after harvest formed the nuclei of winter colonies (Whitehead 1957). Similar observations were made in Israel and Italy (Berlinger 1977, Duso 1990). Berlinger (1977) noted that winter population levels were low in Israel and consisted mainly of non-ovipositing adult females.

The influence of temperature on the development of *P. ficus* under fluctuating temperatures was studied by Kriegler (1954) on potatoes and by Duso *et al.* (1985) and Berlinger (1977) in the field. Berlinger (1977) found that cool early summer temperatures delayed upward migration which delayed the population peak. No life table studies at constant temperatures have been reported.

However, indications were that the optimum temperatures for ranged from 23°C to 27°C (Duso *et al.* 1985).

# 1.9 BIOLOGICAL CONTROL

Many natural enemies associated with *P. ficus* have been reported. Some of these were hyperparasitoids (Table 1.5).

**TABLE 1.5.** Natural enemies associated with *P. ficus*.

Order and	Species	Reference	Comment
Family			
Diptera:	Leucopis sp.	Rzaeva (1985)	
Chamameyidae			
Hymenoptera:	Pachyneuron concolor	Rzaeva (1985)	Possible
Encyrtidae	Forster		hyperparasitoid
	Allotropa mecrida	Rzaeva (1985)	
	Walker		
	Anagyrus pseudococci	Rzaeva (1985),	
	(Girault)	Urban (1985),	
		Trjapitzyn &	
		Trjapitzyn (1999)	
	Chartocerus subaeneus	Rzaeva (1985)	Possible
	Forster		hyperparasitoid
	Clausenia josefi Rosen	Rosen (1965),	
		Berlinger (1977),	
		Trjapitzyn (1989)	
	Coccidoxenoides	Berlinger (1977),	Synonym:
	peregrinus (Timberlake)	Urban (1985),	Pauridia
		Trjapitzyn (1989)	peregrina

Table 1.5 continued

Order and	Species	Reference	Comment
Family			
Hymenoptera:	Leptomastix flavus	Berlinger (1977)	
Encyrtidae	Mercet		
	Leptomastidea abnormis	Berlinger (1977),	
	(Girault)	Urban (1985),	
		Trjapitzyn (1989),	
		Trjapitzyn &	
		Trjapitzyn (1999)	
	Prochiloneurus bolivari	Trjapitzyn (1989)	Possible
	(Mercet)		hyperparasitoid
	Prochiloneurus	Trjapitzyn (1989)	Possible
	pulchellus (Silvestri)		hyperparasitoid
	Chrysoplatycerus	Identified in current	
	splendens (Howard)	study	
Neuroptera:	Chrysoperla carnea	Rzaeva (1985)	
Chrysopidae	(Stephens)		
Coleoptera:	Nephus reunioni Fürsch	Rzaeva (1985)	
Coccinellidae	Cryptolaemus	Orlinskii et al. (1989)	
	montrouzieri Mulsant		
	Hyperaspis felixi	Whitehead (1957),	
	Mulsant	Urban (1985)	
	Nephus angustus Casey	Whitehead (1957),	
		Urban (1985)	
	Nephus binaevatus	Whitehead (1957),	
	Mulsant	Urban (1985)	
Coleoptera:	Nephus quadrivittatus	Whitehead (1957),	
Coccinellidae	Mulsant	Urban (1985)	

Table 1.5 continued

Order and Family	Species	Reference	Comment
	Rhizobiellus sp.	Whitehead (1957)	
	Cydonia lunata F.	Whitehead (1957)	
	Scymnus nubilis	Identified in current	
	Mulsant	study	

From the list it is clear that *P. ficus* populations are attacked by a range of natural enemies, many of which are from the Western Cape Province (Whitehead 1957, Urban 1985). These include, in descending order of abundance,

#### Parasitoids:

Anagyrus spp., Coccidoxenoides peregrinus, Leptomastix dactylopii, (Whitehead 1957, Urban 1985)

## Predatory beetles:

Nephus bineavatus, N. angustus and N. quadrivittatus (Whitehead 1957, Urban 1985).

Berlinger (1977) also found that the parasitoids and predators mentioned above were the dominant ones in Israel. Whitehead (1957) believed that the predatory beetles played a major part in biological control and that the parasitoids were of lesser importance. Predatory beetle populations were found to peak early in the season (from September to November) and declined after this. However, mealybug population levels did not decrease while the predators were present (Berlinger 1977, Urban 1985) both in the Western Cape and in Israel. Parasitoid

numbers reached a peak later in the season (from November), which resulted in the destruction of most of the mealybug colonies (Berlinger 1977, Urban 1985) towards the end of the season (February to March). This suggested that the parasitoid complex played a major role in reducing *P. ficus* numbers.

Biological control was severely hampered by the presence of a variety of ant species (Kriegler 1954, Whitehead 1957, Ueckermann 1998) in vineyards in the Western Cape Province. This was also reported in Israel (Berlinger 1977). Ant control has been achieved using chemical stem barrier treatments (Ueckermann 1998).

## 1.10 CHEMICAL CONTROL

During the past number of years chemical control of *P. ficus* in South Africa has been based on either two treatments of chlorpyrifos two weeks apart, or prothiophos just before bud burst. These treatments are applied during the dormant period. An additional supplementary treatment of a chemical with a short residual period, such as dichlorvos or methidathion, has sometimes been applied prior to harvest from January to April (Nel *et al.* 1999). However, *P. ficus* colonies are protected by wax threads and are not easily controlled by these routine sprays. Populations usually occur under bark and in crevices on the main stem as well as on roots, making it difficult to target this pest with insecticides (Berlinger 1977). Kriegler (1954) and Whitehead (1957) recommended the

application of spot treatments with chemicals at high mealybug infestations.

However, they emphasized the integrated use of chemical and biological control.

## 1.11 CULTURAL CONTROL STRATEGIES

Bugg & Waddington (1994), Whitehead (1957), and Urban (1985) suggested that the preservation of surrounding vegetation was important for optimising conditions for natural enemies. Cover crops were effective only if they attracted Coccinellidae and Neuroptera (Bugg & Waddington, 1994). These authors also noted that common vetch (*Vicia sativa*) had stipular extra floral nectaries that attracted parasitic wasps.

Urban (1985) and Neuenschwander & Hagen (1980) showed that, by providing pollen, nectar, suitable habitats, sprays of sucrose or a yeast product plus sucrose, led to an increase in local populations of predatory coccinellids, chrysopids, and hemerobiids. These food sources increased the longevity not only of predators, but also adult encyrtid wasps and enhanced biocontrol of mealybugs in the field (Neuenschwander & Hagen 1980, Urban 1985).

Kriegler (1954) and Flaherty et al. (1982) found that leaf removal and correct summer pruning reduced the number of leaves which predators and parasitoids had to cover in search of prey, increasing their effectiveness. This also reduced mealybug populations by removing them with the surplus stems and leaves, and contributed to better aeration of vines. Road dust and inert carriers of fungicides

should be kept to a minimum as these adversely affected natural enemies (Searle 1965). Mealybugs overwintered on old wood under loose bark and readily infested bunches which later touched the woody parts of the vine. Bunches that hung free from old wood were less susceptible to cosmetic damage. Therefore, they should be thinned so as to avoid contact with old wood (Kriegler 1954, Flaherty *et al.* 1982). The use of chemical and sticky stem barriers to keep ants from the vine canopy could further aid in biological control of *P. ficus* (Whitehead 1957, Ueckermann 1999).

#### 1.12 INTEGRATED CONTROL

Whitehead (1957), Berlinger (1977) and Urban (1985) believed that an integrated approach should be followed. This would enhance biological control. In addition, ant exclusion by stem barriers was considered an important element of the integrated system (Whitehead 1957). If biological control was not adequate, limited chemical intervention using spot treatments of short residual pesticides, should be considered.

Presently, integrated production of wine (IPW) is encouraged by the wine industry in South Africa (Tromp & Marais 2000). This system includes sound integrated pest management strategies for suppressing pests such as *P. ficus*. Strategies include monitoring pest activity, pest control practices such as trunk barriers, optimised use of biological control, and limited use of chemicals during the growing season. In addition, an AgChem Environmental Work Group codes

all registered pesticides for acceptability in integrated production systems for use against insect pests, including those for *P. ficus*. This coding system is based on the environmental impact of products (Walton & Pringle 1999, Tromp & Marais 2000, Walton & Pringle 2001). Producers are encouraged to implement these guidelines (www.ipw.co.za) Random audits are conducted to test compliance with the guidelines.

## 1.13 CONCLUSIONS

The taxonomic status of *P. ficus* has been uncertain. In addition, the techniques used for preparing specimens for identification are difficult (De Lotto 1975, Ben-Dov 1994). Because of this and as a result of several discussions with Yair Ben-Dov, René Sforza and Ian Millar it was decided to have the specimens found during the surveys identified by Ian Millar who also has access to the necessary reference material deposited in the South African National Collection of Insects, Pretoria.

No recent information is available on the species composition of pseudococcids and related natural enemies in Western Cape vineyards. Several authors (paragraph 1.7) have indicated the importance of mealybugs as vectors of vine leafroll. Therefore, information on the species composition of mealybugs which is currently lacking is required for planning control measures in Western Cape vineyards. The identity and phenology of the most abundant natural enemies

must also be determined so that the effectiveness of biological control systems can be optimised.

The work by Kriegler (1954) on the developmental biology of *P. ficus* was detailed. However, controlled environmental conditions were not used, making it impossible to determine parameters such as lower and upper developmental temperatures for *P. ficus* and its important natural enemies. These parameters can be used to estimate the number of degree days required for both insects. Degree days may highlight susceptible developmental periods in which control actions would be most profitable. Developmental parameters could further be used to optimise mass rearing techniques.

Reliance on pesticides for *P. ficus* management necessitated the development of an alternative pest control tool such as mass releases of natural enemies. To implement this, natural enemies need to be produced. A survey of the published information on mass rearing parasitoids has been produced by Etzel & Legner (1999) but no literature was available on the mass rearing of *C. peregrinus* on *P. ficus* and this information should be submitted. Mass release methodology and effectiveness of natural enemies on *P. ficus* pest populations need investigation.

In order to correctly time control actions such as mass releases or chemical control of *P. ficus* pest populations, accurate information on field infestation levels are needed. Currently no monitoring system with known levels of error for

*P. ficus* infestation levels exist and this aspect should be addressed. With the above information, action thresholds could be determined and used as a powerful tool in *P. ficus* management.

Information gathered on the above aspects should be combined to construct a decision model for integrated *P. ficus* management. The decision model should be verified in field situations and appropriate adjustments made. Further, future work which is not included in this study are the use of *P. ficus* pheromone traps (Hinkens *et al.* 2001) as an added monitoring aid, as well as the use of pheromones for mating disruption.

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### **CHAPTER 2**

# MASS REARING OF *PLANOCOCCUS FICUS* AND *COCCIDOXENOIDES*PEREGRINUS; EXPERIMENTAL SITE LAYOUT AND SAMPLING METHODS

#### 2.1 INTRODUCTION

Many of the techniques employed in the study were used in more than one of the chapters. In addition, the same field study sites were used for more than one aspect of the work. Therefore, to avoid repetition this short chapter describing the methods and study sites common to more than one of the chapters has been included.

#### 2.2 MATERIAL AND METHODS

# 2.2.1 Mass rearing of P. ficus and C. peregrinus

Mass rearing of *P. ficus* and its parasite *C. peregrinus* was required for the developmental biology (Chapter 4) and biological control (Chapter 7) studies. Mass rearing of *P. citri* on butternut pumpkins, *Cucurbita moschata*, has been described by Krishnamoorthy and Singh (1987). However, no published information on mass rearing *P. ficus* could be found. A comprehensive survey of the published information on mass rearing parasitoids has been produced by Flaherty & Wilson (1999) and Elzen & King (1999). These literature reviews include mass production methods for pseudococcid parasitoids. However, no reference to methods for mass production of *C. peregrinus* could be found. This

parasitoid is being produced in a local insectary in South Africa for biological control of *P. citri*, but the techniques being used have not been published for commercial reasons. The methods of mass rearing and field release was therefore modified for experimental purposes and are described here.

Mealybug stock cultures were reared on butternuts in cages (500 mm x 300 mm x 300 mm) in rearing rooms at 23 to 26°C. The cages were covered with fine insect netting to prevent infestation by other parasitoids. For mass rearing, butternut-filled nylon sleeves were placed on and around these rearing cages to collect newly emerged *P. ficus* crawlers. The nylon sleeves, about 1.4 m in length, were then suspended at a height of 1.7 m from steel rails bolted onto the wall and the mealybugs were allowed to develop. As crawlers started to appear, new nylon sleeves containing butternuts were laid flat in wire containers (400 mm x 300 mm x 100 mm) below and resting on top of the suspended sleeves in order to collect them. The rate of emergence of crawlers was increased by raising the room temperatures from 25°C to 27°C for a maximum of three days. However, optimum mealybug production was achieved between 23°C and 26°C. Relative humidity was kept below 60 % to prevent fungal growth on the honeydew secreted by the mealybugs. The mealybug rearing rooms were washed at weekly intervals and spoiled butternuts were removed daily.

As soon as the new sleeves were adequately infested with newly emerged crawlers (after about 7 days), crawlers were transferred to a parasitoid mass

rearing cage. *C. peregrinus* stock cultures were maintained in a parasitoid rearing room in rearing cages similar to those used for vine mealybug stock cultures. Parasitoid populations from the stock cultures were maintained by continuously adding single butternuts infested with mealybug crawlers from the nylon sleeves.

Steel frame mass rearing cages (1800 mm x 500 mm x 1800 mm) with hinged doors, a solid base and covered with fine insect netting on the sides and top were constructed for mass rearing *C. peregrinus*. These mass rearing cages, containing a minimum of twenty sleeves of crawler infested butternuts, were wheeled directly from the mealybug culture room at the one end of the insectary to the parasitoid rearing room at the other end. A parasitoid stock cage with emerging parasitoids was placed inside the mass rearing cage and parasitoids (about 10 000) were allowed to oviposit in crawlers on the sleeved butternuts for the next seven to 10 days.

# 2.2.2 Harvesting, packaging and field release of C. peregrinus

After being parasitised, the mealybugs became restless and most of them dropped from the butternuts kept in the nylon sleeves. The mealybugs accumulated between layers of newspaper and shredded paper that had been placed on the floor of the mass-rearing cage. The parasitised mealybugs died shortly after dropping to the floor and formed *C. peregrinus* mummies two weeks after parasitism.

The shredded paper on top of the newspaper was used to reduce the amount of honeydew falling on the newspaper, thereby facilitating harvesting of the mummies. Most of the mummies containing parasitoid pupae were harvested one month after introduction of the parasitoids. The harvested mummies were sieved to remove paper clippings, and other waste products so as to ensure a clean parasitoid culture. About 2 % of the mummies were retained and put back into the parasitoid culture cages with crawler-infested butternuts. Mummies ready for field release were weighed (0.13 g ca. 1000 C. peregrinus mummies), and placed in paper distribution bags (40 mm x 70 mm), each containing approximately 1000 mummies. Adult parasitoids usually began to emerge one or two days after packaging. Emergence was delayed for up to two weeks by lowering the storage temperature of bagged mummies to 18°C.

# 2.2.3 Experimental blocks

2.2.3.1 Seasonal population studies (Chapter 6) of vine mealybug

One block of one hectare was regularly inspected in each of three grape growing areas, namely Stellenbosch (33°54'E, 18°52'S, alt. 146 m) (Merlot, planted in 1989), Hex River Valley (33°30'E, 19°33'S, alt. 370 m) (Dauphine, planted in 1985) and Robertson (33°49'E, 19°47'S, alt. 180 m) (Cabernet Sauvignon, planted in 1990). These blocks were at least 100 m away from the biological control study blocks (Chapter 7).

# 2.2.3.2 Biological control studies (Chapter 7)

The experimental site layout to evaluate the effect of mass releases of natural enemies in the field was the same as that used by Luck *et al.* (1988), Luck *et al.* (1999) and Elzen & King (1999). Three experimental vineyards were used in the Hex River Valley (Dauphine, planted in 1989 and two Barlinka vineyards, planted in 1985), a table grape area, and three in each of the wine grape areas of Stellenbosch (Merlot, planted in 1989; Cinsaut planted in 1960 and Chardonnay planted in 1993) and Robertson (Merlot, planted in 1990; Cabernet Sauvignon, planted in 1990 and Chardonnay, planted in 1992). 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. Therefore, a total of 30 ha were sampled in this study. This was made up of 3 ha for the seasonal population studies (Chapter 6) and 27 ha for the biological control studies (Chapter 7). All 30 ha were also used for developing a sampling system for monitoring *P. ficus* population levels in vineyards (Chapter 5).

# 2.2.4 Chemical control

In the blocks used for the phenological population study of vine mealybug and its natural enemies (Chapter 6), pesticide applications against mealybugs included two applications of chlorpyrifos EC at 200 ml/100l before bud break at an interval of two weeks in all blocks. Stem barrier treatments with alpha-cypermethrin SC at 20 ml/l for ant control were applied where necessary. All vines and trellis

systems were treated with 50 m² of this pesticide (Ueckermann 1998) in order to prevent ants from moving into the vine canopy.

In the blocks used for the biological control studies (Chapter 7), dormant IPM-compatible ant and mealybug treatments of chlorpyrifos two weeks apart before bud burst were applied in the buffer and control blocks(Table 2.1). In-season (from October to March) ant (alpha-cypermethrin) and mealybug (mevinphos) treatments were applied where necessary (Table 2.1) in the buffer and control blocks. These treatments were applied prior to the first parasitoid releases. The normal fungicide treatments were used in all blocks. All cover sprays of insecticides were omitted from the parasitoid release blocks (Table 2.1)

# 2.2.5 Sampling

# 2.2.5.1 Mealybugs

Sampling in the blocks used for the seasonal population study of vine mealybug and its natural enemies (Chapter 6) and sub-plots used for the biological control studies (Chapter 7) was conducted in twenty evenly spaced plots each consisting of five vines. Therefore, a central systematic sampling system was used. The lateral branches of each of these vines were inspected for *P. ficus* for a distance of 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 vines. 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.

**Table 2.1**. Insecticide treatments applied in the nine trial sites in Stellenbosch, Hex River Valley and Robertson where field trials were done.

Insect	Chemical	Treatment	Time of	Release	Buffer	Control
treated			treatment	area	area	area
Mealybug	Dursban EC (chlorpyrifos) 100 – 200 ml/100l	Dormant	2, and 1 weeks before budbreak (September)	None	Spot	Spot
Mealybug	Phosdrin EC (mevinphos) 150 ml/100l	Seasonal	One month before harvest (February – March)	None	Spot	Spot, and full cover
Ants (where necessary)	Fastac (alpha- cypermethrin) 20 ml/l	Seasonal	Early season (October)	Stem (full plot)	Stem (full plot)	Stem (full plot)

#### 2.2.5.2 Natural enemies

Yellow sticky traps have been used for trapping parasitoids (Samways 1988, Viggiani 1995) and predatory beetles (Heathcote 1978, Dowell & Cherry 1981, Neuenschwander 1982, Schultz 1985). In the present study yellow rectangular

Agribiol® (200 mm x 100 mm) sticky traps were used to sample adult parasitoids and predators. In addition, mealybug infested butternuts, each containing at least one hundred mealybugs at various stages of development were placed in polystyrene containers with entry holes smeared with petroleum jelly which effectively excluded ants. This was used as an additional method to monitor natural enemy populations as described by Urban (1985).

Two butternuts and two sticky traps were used; one on the edge and one in the middle of each trial block (Chapter 6, Seasonal population studies) and sub-plot (Chapter 7, Biological control of the vine mealybug). Both butternuts and yellow sticky traps were placed in the cordon area of the vines between 1.2 and 1.5 m above ground level. The butternuts and sticky traps were left in the field for one month, after which they were replaced. Butternuts were placed in emergence cages for between one and two months, after which natural enemies were identified and counted. Yellow sticky traps were taken to the laboratory, where identification and counting of predatory beetles and parasitoids was conducted using a stereoscopic microscope. Initial verification and comparison with reference material of the predatory beetle and parasitoid species was done in conjunction with V. B. Whitehead at the S.A. Museum in Cape Town, and G. L. Prinsloo at the ARC – Plant Protection Research Institute in Pretoria respectively. The predatory beetles and parasitoids found using these methods are listed in Chapter 3.

#### 2.2.6 Weather data

Daily minimum and maximum temperature data as well as average daily temperatures for the study period were obtained from the ARC Institute for Soil Climate and Water Agrimet in Stellenbosch for the three stations in Stellenbosch (33°54'E, 18°52'S, alt. 146 m) (Nietvoorbij), Hex River Valley (33°30'E, 19°33'S, alt. 370 m) (ARC experimental farm) and Robertson (33°49'E, 19°47'S, alt. 180 m) (Goree). These data were used for estimating the accumulated number of degree days (°D) in each area, enabling the estimation of the number of *P. ficus* and *C. peregrinus* generations in each area (Chapter 8).

## 2.3 CONCLUSIONS

Mass rearing methods of *P. ficus* and *C. peregrinus* were necessary to ensure sufficient quantities of *P. ficus* and *C. peregrinus* for developmental studies (Chapter 4) as well as sufficient quantities of *C. peregrinus* for biological control studies (Chapter 7). Experimental blocks were selected for seasonal population studies (Chapter 6) in each of the Stellenbosch, Hex River Valley and Robertson areas, and for evaluation of mass releases of natural enemies (Chapter 7) under different climatic conditions.

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### **CHAPTER 3**

# A SURVEY OF MEALYBUGS AND ASSOCIATED NATURAL ENEMIES IN VINEYARDS IN THE WESTERN CAPE PROVINCE, SOUTH AFRICA

#### 3.1 INTRODUCTION

The vine mealybug, *Planococcus ficus*, causes direct crop loss and progressive weakening of vines through early leaf drop. It is also a vector of the vine leafroll virus (Engelbrecht & Kasdorf 1990, Cabaleiro *et al.* 1999, Sforza *et al.* 2000). Nineteen other species of Pseudococcidae cause similar damage worldwide (Krishnamoorthy & Mani, 1989, Longo, Ben-Dov 1994, Mazzeo & Russo 1994, Williams 1998). It is possible that mealybug species other than *P. ficus* could have colonised vineyards in South Africa subsequent to a survey by Kriegler (1954). Therefore, updated information on the species complex of pseudococcids in South African vineyards is necessary as the most recent work done was the survey conducted by Kriegler (1954).

#### 3.2 MATERIAL AND METHODS

One random set of at least five samples of mealybugs was collected from vineyards in each of the districts of Stellenbosch (L'Avenir, 33°54'E, 18°52'S; alt. 146m), Malmesbury (Swartland wine cellar, 33°27'E, 18°44'S; alt. 210 m), Porterville (Lankgewag, 33°10'E, 19°01'S; alt. 866 m), Paarl (St. Pieters Roche, 33°45'E, 18°56'S; alt. 115 m), Hex River Valley (Werda, 33°26'E, 19°33'S; alt.

370 m), Robertson (Goree, 33°49'E,19°47'S; alt. 180 m), Vredendal (Houmoed, 31°66'E, 18°49'S; alt. 56 m), Montagu (Witklei, 33°79'E, 20°25'S; alt. 465 m), Mc Gregor (Steenbokslaagte, 33°54'E, 20°42'S; alt. 354 m), Barrydale (Lentelus, 33°57'E, 19°49'S; alt. 165 m), Ladismith (33°30'E, 21°16'S; alt. 531 m), Calitzdorp (33°32'E, 21°41'S; alt. 280 m) and De Rust (Doornkraal, 33°24'E, 22°33'S; alt. 593 m) during March of 2000. Samples were taken from bunches, leaves and the main stem in all the areas. Mealybug samples were also taken from vine roots to a depth of 60 cm and up to 60 cm from the main stem of vines in Stellenbosch, Robertson and Hex River Valley. Mealybugs were further collected from weeds growing in close proximity to the vines. They were sampled by examining the entire plant for their presence. All mealybug samples were preserved in 70 % alcohol and sent to I. Millar of the Plant Protection Research Institute (PPRI) in Pretoria for identification.

Sampling natural enemies was done on a monthly basis using mealybug infested butternuts and yellow sticky traps as described in Chapter 2.

#### 3.3 RESULTS AND DISCUSSION

Most vines were infested with *P. ficus* (Table 3.1), with the largest populations above ground throughout the season. This suggested that *P. ficus* was the

dominant mealybug infesting vines. However, *P. ficus* was also present on vine roots to a depth of 30 cm. In one case *P. ficus* was found surviving on roots of a vineyard that was pulled out 24 months earlier in the Mc Gregor area. Other mealybug species found on vines included *Pseudococcus longispinus* (Targioni) and *Ferrisia malvastra* (McDaniel).

Mealybugs on weeds were found mainly on the roots. Of the mealybugs found on weeds in vineyards, only *Pseudococcus viburni* (Maskell) was previously reported on grapevines in Chile (Gonzalez, Curkovic & Barria 1996), Australia (Williams 1985), New Zealand (Cox 1987), United States (Phillips & Sherk 1991) and Israel (Ben Dov 1994). However, during the present survey it was not recorded on grapevines.

**TABLE 3.1**. Mealybug species identified from different host plants in vineyards in various areas of the Western Cape Province, South Africa.

Host plant	Sample area	Mealybug species
Vitis vinifera	Barrydale, Calitzdorp, De	Planococcus ficus (Signoret)
L. (above	Rust, Hex River Valley,	
ground)	Ladismith, Malmesbury, Mc	
	Gregor, Montagu, Paarl,	
	Porterville, Robertson,	
	Vredendal	

Table 3.1 continued

Host plant	Sample area	Mealybug species		
Vitis vinifera L.	Hex River Valley,	Planococcus ficus (Signoret)		
(below ground)	Malmesbury, Mc			
	Gregor, Robertson,			
	Stellenbosch			
Conyzia	Stellenbosch	Vryburgia transvaalensis (Brain),		
bonariensis (L.)		Pseudococcus viburni (Maskell),		
Cronq (roots)		Phenacoccus solani Ferris		
Bidens pilosa L.	Stellenbosch	Phenacoccus solani Ferris,		
(roots)		Pseudococcus viburni (Maskell)		
Datura stramonium	Stellenbosch	Pseudococcus viburni (Maskell)		
L. (roots)				
Erodium	Stellenbosch	Pseudococcus viburni (Maskell)		
moshantum (L.)				
L'Herit ex Ait.(roots)				
Sonchus oleraceus	Stellenbosch	Pseudococcus viburni (Maskell)		
(L.) Hill. (roots)				

Predatory beetles recorded in Stellenbosch, Hex River Valley and Robertson included *Cryptolaemus montrouzieri* Mulsant, *Nephus angustus* (Casey), *N. quadrivittatus* (Mulsant), *N. binaevatus* (Mulsant), *Nephus* sp., *Hyperaspis felixi* (Mulsant), *Scymnus nubilis* Mulsant, *Cydonia lunata* F., a *Rhizobiellus* sp. and a *Hippodamia* sp., confirming work by Whitehead (1957). The only predatory beetle not previously recorded in South Africa prior to the survey, was *S. nubilis*. This species was recorded from all areas. The *Nephus* species were the most abundant species found during the survey. Other species of predatory beetles

were found only occasionally. The only predators found other than Coleoptera were *Chrysopa* spp..

Three primary parasitoids recorded in all three areas belonged to the Encyrtidae.

They were *Anagyrus* sp., *Leptomastix dactylopii* (Howard), and *Coccidoxenoides*peregrinus (Timberlake). A fourth encyrtid, *Chrysoplatecyrus splendens* Howard was found once in Robertson and Stellenbosch.

Possible hyperparasitoids of *P. ficus* found were *Chartocerus* spp.

(Hymenoptera: Signiphoridae), *Cheiloneurus* spp. (Hymenoptera: Encyrtidae) and *Pachyneuron* spp. (Hymenoptera: Pteromalidae). They were recorded in Stellenbosch, Hex River Valley and Robertson.

#### 3.4 SUMMARY

The dominant mealybug species in South African vineyards was *P. ficus*, which confirmed work by Whitehead (1957) with *Ps. longispinus* recorded occasionally. *Ps. longispinus* was an addition to the list of pseudococcid vine leafroll virus vectors in South Africa and should be included in future epidemiological work of the vine leafroll virus.

The fact that *P. ficus* could colonise roots to a depth of 60 cm has far reaching implications for the control of this virus vector. Chemical control of vine mealybug is designed to target the pest on above-ground parts of the vine. No

below-ground chemical or systemic chemical control measures are available. This suggests that current vector control practices in supposedly vine leafroll virus free propagation material blocks need revision. Work on the control of these below-ground *P. ficus* populations with systemic pesticides is therefore needed. Altered conventional pesticide spray protocols to control these populations should be investigated.

The range of natural enemies found during the study period was similar to that found in South Africa by Whitehead (1957) and Urban (1985). This indicated that no significant change regarding dominance of specific species has occurred since the 1950's. Care should however be taken to preserve these insects by limiting chemical sprays as outlined in Walton & Pringle (1999) and Walton & Pringle (2001). Future work on natural enemies should be focused on the importation of new species, possibly similar to those reported by Trjapitzyn & Trjapitzyn (1999) from Argentina.

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#### **CHAPTER 4**

DEVELOPMENTAL BIOLOGY OF VINE MEALYBUG, PLANOCOCCUS FICUS
(SIGNORET) (HOMOPTERA: PSEUDOCOCCIDAE), AND ITS PARASITOID

COCCIDOXENOIDES PEREGRINUS (TIMBERLAKE)(HYMENOPTERA:
ENCYRTIDAE).

## 4.1 INTRODUCTION

Vine mealybug is a key pest on grapevines in most grape growing areas in South Africa. The biology of *Planococcus ficus* was described in South Africa by Kriegler (1954). The only information available on the developmental biology of mealybug was from Kriegler (1954) who did developmental studies on *P. ficus* at fluctuating temperatures. The object of the current study was to compare the developmental biology of *P. ficus* at a range of temperatures with that of an important natural enemy, *Coccidoxenoides peregrinus* (Timberlake), on grapevines in the laboratory. This information was required as a first step in understanding the effect of temperature on the rate of development of the pest and its natural enemy.

## 4.2 MATERIALS AND METHODS

Cultures of *P. ficus* and *C. peregrinus* from the insectary colonies (Chapter 2) were used. The developmental times, fecundity and fertility of the two insects were determined at 18, 20, 25, 27 and 30°C for *P. ficus* and 18, 21, 25, 27 and

30°C for *C. peregrinus* using cooled incubators in which the humidity ranged from 60-90 %. A light: dark regime of L16:D8 was used for both insects.

Ovipositing adult *P. ficus* females were introduced onto potted grapevine seedlings (Waltham Cross) and left for 24 hours before being removed. A minimum of 25 eggs was retained on each of four plants per treatment. Barriers of petroleum jelly restricted mealybug movement. The development, adult longevity and fecundity of individual mealybugs were recorded daily. Mealybugs lost due to escape or injury were omitted from the analysis.

A minimum of twenty, one-day-old adult *C. peregrinus* was introduced into each of three ventilated plastic boxes containing butternuts heavily infested with first and second instar mealybugs. After 24 h all surviving parasitoids were removed and placed in similar holders for a further 24 h. This process was repeated until no more parasitoids were alive. Each of the boxes was monitored daily for the emergence of offspring of the parasitoids. Newly emerged *C. peregrinus* were removed daily and isolated in ventilated glass vials. They were provided with honeydew as a food source. As *C. peregrinus* is normally parthenogenic (Clausen 1962), no sexing was considered necessary. Mortality of *C. peregrinus* was recorded daily.

# 4.2.1 Life table calculations

 $L_X$ , the proportion of individuals alive on day x, and  $M_X$ , the mean number of female progeny produced on day x, were determined for the duration of the life span, of both *P. ficus* and *C. peregrinus*. The net reproduction rate ( $R_0$ ) was determined using  $\sum_{x=1}^{t} L_x M_x$ , where t = time in days. The mean generation time (T) was calculated using (Watson 1969; Price 1984),

$$T = \frac{\sum L_x M_x X}{\sum L_x M_x} \ .$$

These values were subsequently used to obtain an initial estimate of the intrinsic rate of of natural increase  $(r_m)$  using (Price 1984),

$$r_m = [\ln R_o)]/T$$
.

The estimate of  $r_m$  was then used in the first iteration to solve the equation (Watson 1964),

$$\sum_{x=1}^{t} (e)^{-r_m x} L_x M_x = 1, X = 1, 2, 3, \dots, t \text{ days.}$$

The iterations were continued until the left-hand side of the equation was within 0.0001 of the right hand side.

The minimum threshold temperature for development was determined by regressing 1/t on temperature for P. ficus and C. peregrinus and then solving the regression equation for 1/t = 0, where t =time in days. In instances where the rate of development decreased at temperatures higher than an optimum temperature a quadratic regression of the rate of development on temperature was used. The optimum temperature was estimated by setting the first derivative of the quadratic equation equal to zero.

#### 4.3 RESULTS AND DISCUSSION

# 4.3.1 Developmental times

The time for development from egg to oviposition of adult female mealybugs (egg to adult plus pre-oviposition period) decreased from 90.33 days at 18°C to 28.05 days at 25°C (Table 4.1). At 30°C it increased to 43.1 days. Fecundity was directly influenced by temperature (Table 4.1) and reached a maximum number of eggs/female at 20 to 25°C. This was similar to the fecundity of *P. ficus* reported by Kriegler (1954) who recorded 275 eggs/female at an average temperature of 19.3°C, 348 eggs/female at an average of 20.8°C and 395 eggs/female at an average of 23.5°C. Prinz (1923) and Bodenheimer (1929) found that the fecundity of *P. citri* was 12 eggs/female at 17°C and 180 eggs/female at 21°C.

**TABLE 4.1.** Developmental times in days (± S.E.) for eight developmental stages and fecundity of *P. ficus* on Waltham Cross grapevines at five temperatures (±0.5°C).

Developmental	Temperature					
stage	18° C	20° C	25° C	27° C	30° C	
Egg	11.70 (0.12)	10.60 (0.3)	7.31 (0.13)	6.96 (0.1)	6.37 (0.2)	
1 <sup>st</sup> . Nymphal	5.60 (0.1)	4.75 (0.2)	2.70 (0.2)	2.20 (0.1)	3.86 (0.3)	
2 <sup>nd</sup> . Nymphal	10.86 (0.4)	11.75 (0.4)	8.20 (0.6)	6.20 (0.4)	5.50 (0.4)	
3 <sup>rd</sup> . Nymphal	16.30 (1)	13.53 (0.7)	6.42 (0.9)	10.20 (0.3)	7.43 (0.4)	
Male prepupa	8.50 (0.7)	4.10 (0.5)	5.53 (0.8)	3.00 (0.3)	3.11 (0.2)	
Male pupa	5.94 (0.87)	2.95 (0.3)	4.13 (0.2)	3.78 (0.3)	2.47 (0.2)	
Adult male	3.33 (0.2)	2.60 (0.4)	1.53 (0.1)	1.33 (0.1)	1.07 (0.1)	
Adult female	45.71 (3.1)	41.00 (1.9)	19.05 (1.2)	17.61 (1.1)	22.82 (3.3)	
Egg to adult:	44.46 (0.4)	40.60 (0.4)	24.61 (0.5)	25.53 (0.2)	23.20 (0.3)	
Female						
Pre oviposition						
period	45.87 (1.1)	36.11 (0.8)	3.44 (1.5)	15.79 (1.2)	19.90 (0.9)	
Eggs per	75.0 (9.9)	316.0	297.0	148.0	78.8 (6.7)	
female		(12.9)	(14.7)	(13.8)		

**TABLE 4.2**. Developmental times in days (±S.E.) of two developmental stages of *Coccidoxenoides peregrinus* parasitising *Planococcus ficus* at five constant temperatures (±0.5°C).

Developmental	Temperature					
stage	18° C	21° C	25° C	27° C	30° C	
Oviposition to adult	82.29 (0.5)	48.67 (0.3)	31.19 (0.1)	29.86 (0.2)	27.98 (0.3)	
Adult longevity	2.36 (0.1)	1.74 (0.1)	1.12 (0.02)	1.01 (0.01)	1.01 (0.01)	

Table 4.1 continued

Developmental	Temperature					
stage	18° C	21° C	25° C	27° C	30° C	
Eggs per female	18.0 (0.1)	109.0 (1.2)	104.0 (1.5)	18.7 (0.3)	16.2 (0.1)	

The time for *C. peregrinus* to develop from egg to ovipositing adults decreased from 82.29 days at 18°C to 27.98 days at 30°C (Table 4.2). Fecundity increased from an average of 18 eggs per female at 18°C to 109 eggs per female at an average of 21°C. Thereafter, the fecundity decreased as the temperature increased to a minimum of 16.2 eggs per female at 30°C. The number of eggs/female at 25°C, (104 eggs/female) was similar to that reported by Gol'Berg (1985).

# 4.3.2 Life tables

The net replacement rate ( $R_o$ ) was higher for P. ficus than for C. peregrinus at all five temperatures.  $R_o$  for P. ficus reached a maximum at 21°C (308.9) (Table 4.3), and at 25°C for C. peregrinus (69.9) (Table 4.4). The generation times (T) of C. peregrinus were shorter (minimum of 28.5 at 30°C) than those of P. ficus (minimum of 38.0 at 25°C) at all five temperatures. Temperature had less of an effect on the T values of C. peregrinus, than on those of P. ficus. At the higher (30°C) and lower temperatures (18°C) values of T for P. ficus were higher than those for C. peregrinus, suggesting that C. peregrinus may have a reproductive advantage over P. ficus at low and high temperatures.

 $R_{\rm o}$  was greater than zero at all temperatures for both insects (Tables 4.3 & 4.4), indicating positive population growth. The maximum  $R_{\rm o}$  and  $r_{\rm m}$  for P. ficus occurred at 21°C and 25°C respectively (Table 4.3), while the maximum  $R_{\rm o}$  and  $r_{\rm m}$  were recorded at 25°C in the case of C. peregrinus.

**TABLE 4.3**. Life table parameters for *Planococcus ficus* at different temperatures (°C) on Waltham Cross grapevine plants.

Parameter	Temperature								
	18° C	20° C	25° C	27° C	30° C				
Ro	52.45	308.87	248.01	140.26	28.83				
r <sub>m</sub>	0.039	0.068	0.169	0.131	0.072				
T	112.79	96.00	38.0	42.31	57.0				
Sex ratio ♀:♂	2:5	3:5	3:5	4:5	2:5				

The ratio of P. ficus females declined at the extremes of the temperatures tested. The higher numbers of males at high and low temperatures may indicate higher stress levels. This phenomenon was previously recorded and may produce greater genetic variability, which in turn could increase the probability of survival (Castagnoli & Simoni 1991) under stressful conditions. The  $r_{\rm m}$  values for P. ficus were slightly higher than those for P. P0. P1. However, differences in the P1 values between the two insects were small.

**TABLE 4.4**. Life table parameters for *Coccidoxenoides peregrinus* at different temperatures (°C) on *Planococcus ficus*.

Parameter	Temperature								
	18° C	21° C	25° C	27° C	30° C				
Ro	12.33	49.24	69.94	11.72	9.36				
r <sub>m</sub>	0.032	0.093	0.149	0.083	0.081				
T	79.0	42.0	29.5	33.0	28.5				

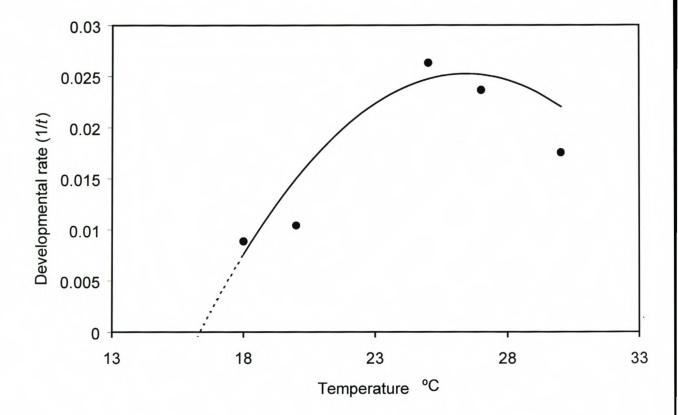
# 4.3.3 Minimum threshold temperature for development

The quadratic regression (Fig. 4.1) of 1/t on temperature for P. ficus was  $Y = -0.00025X^2 + 0.0132X - 0.149$  (F = 3.11; d.f. = 2, 2; P = 0.24;  $R^2 = 0.84$ ).

The estimated minimum and maximum threshold temperatures for development of *P. ficus* were 16.59 and 35.61°C respectively, while the optimum temperature for development was 27.84°C.

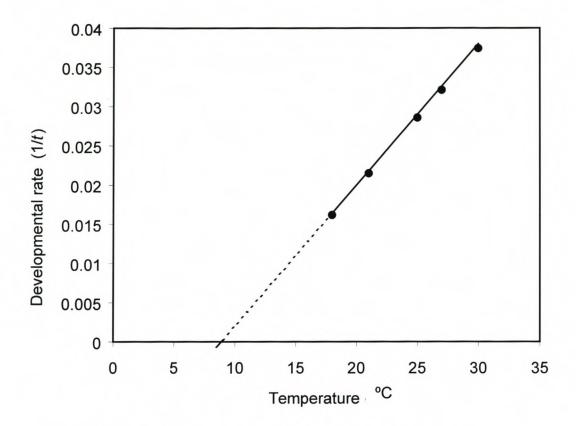
The linear regression of 1/t on temperature (Fig. 4.2) for *C. peregrinus* (F = 15.57; d.f. = 1, 3; P = 0.03; R<sup>2</sup> = 0.84) was Y= 0.0018X - 0.016.

The minimum threshold temperature for development estimated from the regression was 8.85°C. There was no turning point. Therefore, the optimum temperature for development could not be estimated.



**Fig. 4.1.** Developmental rate (1/t) of *Planococcus ficus* at a range of temperatures.

The minimum threshold temperature for development of *C. peregrinus* (8.85°C) was lower than that of *P. ficus* (16.59°C), indicating that the parasitoid should remain active until late winter (July/August). There could then be a decline in activity towards the beginning of the season (October/November) because of low host population levels (Price 1984).



**Fig. 4.2**. Developmental rate (1/t) of *Coccidoxenoides* peregrinus at a range of temperatures.

This could cause vine mealybugs to rapidly increase at the start of the season before that of the parasitoid population. Inundative releases of *C. peregrinus* should start from early November when *P. ficus* infestation levels were low (Kriegler 1954) and the temperatures were still low at that time. Therefore the generation time of *P. ficus* will be long relative to that of *C. peregrinus*. This could result in biological control being achieved early and at a low pest population level.

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### **CHAPTER 5**

THE DEVELOPMENT OF A SAMPLING SYSTEM FOR MONITORING
POPULATION LEVELS OF VINE MEALYBUG, *PLANOCOCCUS FICUS*(SIGNORET)(HOMOPTERA: PSEUDOCOCCIDAE)

#### 5.1 INTRODUCTION

Vine mealybug, *Planococcus ficus*, overwintered and fed underground on roots as well as under the bark and in crevices on the main stem of vines. During spring, crawlers (first instar nymphs) moved up the main stem to the new growth areas where colonies were formed. As the season progressed, these colonies dispersed to newly formed apical leaves. Later in the season colonisation of developing bunches took place reaching a maximum at harvest (Kriegler 1954; Berlinger 1977). In order to prevent bunch damage corrective sprays were often required.

During the season the most commonly used chemicals included chlorpyrifos, dichlorvos, formothion, and mevinphos. These were contact chemicals, and with the exception of chlorpyrifos, had a short residual action. Therefore, correct timing of sprays was important, as mealybug populations could appear at different times during a season. Their time of appearance could also be dependent on vine cultivar. The object of this study was to develop a sampling system for estimating *P. ficus* population levels in commercial vineyards with

known levels of error, enabling producers to decide on the necessity for and correct timing of intervention.

### 5.2 MATERIAL AND METHODS

The experimental sites described in Chapter 2 (2.2.3.1; 2.2.3.2) were used. The release, buffer and control plots were monitored separately by examining 20 plots (per ha) of 5 vines per plot. Sampling was conducted throughout the year for two seasons at intervals of one to four weeks depending on the time of year. The sampling units were stems, leaves and bunches described in Chapter 2.

Presence-absence cluster sampling (Binns *et al.* 2000) was used. The proportion of infested units, *p* (stems, leaves or bunches) was estimated using (Binns *et al.* 2000),

$$p = \frac{\sum_{i=1}^{n} \sum_{j=1}^{N} X_{ij}}{nN}$$
 (1);

for *N* plots (20 in this instance) and *n* stems, leaves or bunches. The binomial variance,  $S_B^2$ , was then estimated using (Binns *et al.* 2000),

$$Var(Bin) = S_B^2 = \frac{p(1-p)}{nN}$$
 (2).

This is only the case for constant *n* in each plot, or for stems and leaves in which case there were five vines in each plot. However, the number of bunches was

not the same in each plot. In such cases  $S_B^2$  was estimated using (Madden *et al.* 1995),

$$S_B^2 = \frac{p(1-p)}{\bar{n}} \quad (3),$$

where 
$$p = \sum_{i=1}^{n} \frac{X_i}{n_i}$$
,

 $\overline{n}$  = the average number of bunches per vine,

 $X_i$  = the number of infested bunches on the i th vine and  $n_i$  the number of bunches on the i <sup>th</sup> vine.

The observed variance,  $S_0^2$ , was estimated using (Binns et al. 2000),

$$Var(Obs) = S_O^2 = \sum_{j=1}^{N} \frac{(p_j - p)^2}{N - I}$$
 (4).

Again, this expression was only true for equal numbers of secondary units, n (stems and leaves in this instance) per primary unit N (plots in this instance). When this is not the case, as with bunches,  $S_0^2$  could be estimated using (Madden *et al.* 1995),

$$S_0^2 = \frac{\sum n_i^2 (y_i - p)^2}{\overline{n}^2 (N - 1)}$$
 (5),

when 
$$y_i = \frac{X_i}{n_i}$$
.

The regression (Binns et al. 2000),

$$ln(S_0^2) = ln(a) + (b)ln(S_B^2)$$
 (6)

was fitted. Taking the antilog of (6), an expression relating the observed variance to the binomial variance can be obtained,

$$S_0^2 = a(S_B^2)^b$$
 (7),

which was very similar to Taylor's power law (Taylor 1961, Binns *et al.* 2000). If infestations were random, the variance of infected units will conform to the binomial distribution, given in (3). A general index for estimating sampling error could be written as (Elliot 1979; Binns *et al.* 2000),

$$D = \frac{\sqrt{S_0^2 / N}}{p}$$
 (8),

where p was the average infestation. Substituting (7) into (8), an estimate of the sampling error can be obtained for any value of average infestation, p,

$$D = \frac{\sqrt{\frac{a}{N} \left(S_B^2\right)^b}}{p}.$$

From (3)

$$D = \frac{\sqrt{\frac{a}{N} I^{\frac{\rho(1-\rho)}{2n}} J^b}}{\rho} \quad \text{(9a),}$$

for equal numbers of secondary units (stems or leaves) and

 $D = \frac{\sqrt{\frac{a}{N} l \frac{p(1-p)}{n}} l^b}{p}$  (9b), for unequal numbers of secondary units (bunches), can be used to estimate the sampling error for any proportion of infested units, p.

Operating characteristic curves (OC curves) can be used to determine the probability of incorrectly deciding not to intervene (for example to apply a spray or to release parasitoids) when the infestation estimated by sampling is below a fixed economic threshold (Binns *et al.* 2000). Generally OC curves can be estimated using a range of values for the average infestation  $\bar{x}$  in,

$$Z = \frac{\overline{x} - ET}{\sqrt{S^2 / N}}$$

where *ET* is the economic threshold and *Z* was the cumulative normal probability function. In the case of the binomial distribution this can be written as (Binns *et al.* 2000),

$$Z = \frac{p - ET}{\sqrt{\frac{1}{N}(S_B^2)}} \quad (10).$$

For cluster sampling, (10) can be written as

$$Z = \frac{p - ET}{\sqrt{\frac{1}{N}(S_0^2)}}$$
 (11).

Substituting (7) into (11),

$$Z = \frac{p - ET}{\sqrt{\frac{a}{N} (S_B^2)^b}} \quad (12)$$

is obtained and substituting (3) into (12),

$$Z = \frac{p - ET}{\sqrt{\frac{a}{N} \left[\frac{p(I-p)}{\bar{n}}\right]^b}} \quad (13),$$

giving an expression for estimating the OC curve for a fixed value of the economic threshold, *ET*, and a range of values of *p*, or a range of estimates of infestation levels obtained from sampling. The corresponding probability levels

of *Z* can be obtained from right handed one-tailed normal probability tables. This provides estimates of the probability of correctly deciding not to apply control measures at a range of infestation levels estimated by sampling.

### 5.3 RESULTS AND DISCUSSION

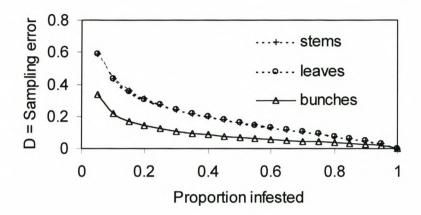
All the regressions of  $\ln(S_{\theta}^2)$  and  $\ln(S_{\theta}^2)$  were highly significant, with good correlations (Table 5.1). The regression constants were very similar for stems and leaves, indicating a similar degree of clustering of mealybugs on these plant parts (Table 5.1). However, the regression constants for bunches were lower than for the leaves and stems, suggesting that bunch infestation was more uniform (less clustered) than for leaves and stems.

**TABLE 5.1**. Regression constants for  $ln(S_{\theta}^{2})$  on  $ln(S_{\theta}^{2})$  for stems, leaves and bunches infested by vine mealybug, *Planococcus ficus*.

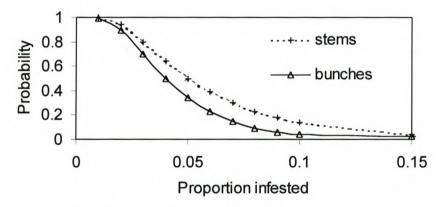
REGRESSION CONSTANTS							
а	Р	b	Р	R <sup>2</sup>			
4.9266	<0.001	1.213	<0.001	0.93			
4.9181	<0.001	1.214	<0.001	0.94			
0.489	0.085	0.87	<0.001	0.56			
	4.9266 4.9181	a P 4.9266 <0.001 4.9181 <0.001	a P b 4.9266 <0.001 1.213 4.9181 <0.001 1.214	a P b P 4.9266 <0.001 1.213 <0.001 4.9181 <0.001 1.214 <0.001			

These regression data were used in (9) to estimate the sampling error, D, for a range of infestation levels, *p*, of stems, leaves and bunches (Fig. 5.1). Similar regression constants for stem and leaf infestation resulted in similar estimates of

sampling error (Fig. 5.1). The lower regression constants for bunches resulted in lower estimates of sampling error than for stems and leaves (Fig. 5.1).



**Fig. 5.1.** Sampling error, D, plotted against proportion of stems, leaves and bunches infested with vine mealybug, *Planococcus ficus*.



**Fig 5.2**. Operating characteristic curve (OC) for sampling *Planococcus ficus* on stems and bunches using a economic threshold (ET) of 5 % infestation per block.

The OC curves for *P. ficus* infestation on stems and bunches were given in Fig. 5.2. An economic threshold (ET) of 5 % infestation per one hectare block was used for both stem and bunch infestations. At an ET of 5 % the decision not to intervene when 2 % of the stems were infested, will not lead to under reacting (exceeding the ET) in 95 % of the cases. For stems with infections of between 3 and 4 %, the reliability of a decision not to intervene will be reduced to between 82 and 65 % of the cases respectively.

In the case of bunches, when the ET is set at 5 % the decision not to intervene at a 2 % infestation level, will not lead to under reacting (exceeding the ET) in 98 % of the cases. For bunches with infestations of between 3 and 4 %, the reliability of decisions not to intervene will be reduced to between 70 and 88 % of the cases. The OC curves for bunches are, however, of little value in decision making as the damage is not reversible. In addition, *P. ficus* is a direct pest on vines with a low ET. At low infestation levels sampling errors are high (Fig. 5.1). If, however, stem infestations were used as an early warning for bunch infestations, this can be partly overcome. It is suggested that intervention should be planned at 2 % stem infestation. As will be shown later (Chapter 6) stem infestation precedes bunch infestation, facilitating forward planning for intervention such as parasitoid releases.

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#### **CHAPTER 6**

SEASONAL POPULATION STUDIES OF VINE MEALYBUG, PLANOCOCCUS
FICUS (SIGNORET), AND ITS NATURAL ENEMIES IN VINEYARDS IN THE
WESTERN CAPE PROVINCE, SOUTH AFRICA

#### 6.1 INTRODUCTION

Vine mealybug, *Planococcus ficus* (Signoret), is a key pest in vineyards worldwide (Whitehead 1957, Berlinger 1977, Urban 1985, Duso 1989, Trjapitzyn & Trjapitzyn 1999). The tendency of *P. ficus* to enter refugia, and to cluster beneath the bark as well as the fact that it excretes large amounts of wax make chemical control of this pest exceedingly difficult (Berlinger 1977). Natural enemies of *P. ficus* (Whitehead 1957, Berlinger 1977, Urban 1985, Duso 1989, Trjapitzyn & Trjapitzyn 1999) and temperature (Berlinger 1977, Copland 1983, Duso 1989) were the major factors affecting population development during the growing season. However, in South Africa there is little information on the phenological trends of *P. ficus* and its natural enemies. This chapter address this shortfall and focuses on when during the year the pest and its different natural enemy guilds are active. In addition the relative importance of the two major quilds (predators and parasitoids) was studied.

### 6.2 MATERIAL AND METHODS

Sampling of mealybugs and natural enemies and trial sites were described in Chapter 2 (2.2.3.1). Average daily temperatures (Chapter 2) were summed and the mean monthly temperatures calculated for each of the three grape growing areas.

Interaction between both groups of natural enemies (parasitoids and predators) and *P. ficus* were analysed by plotting natural enemy population levels on *P. ficus* population levels. These plots aided in the identification of density dependant relationships (May *et al.* 1981). An anti clockwise trend indicated a density dependent relationship. Clockwise and other trends indicated a density independent relationship (May *et al.* 1981).

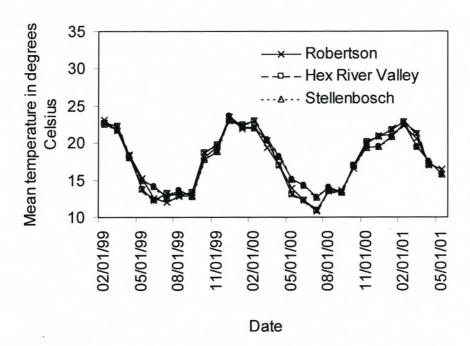
Percent parasitism (%PA) was estimated using (van Driesche 1983),

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

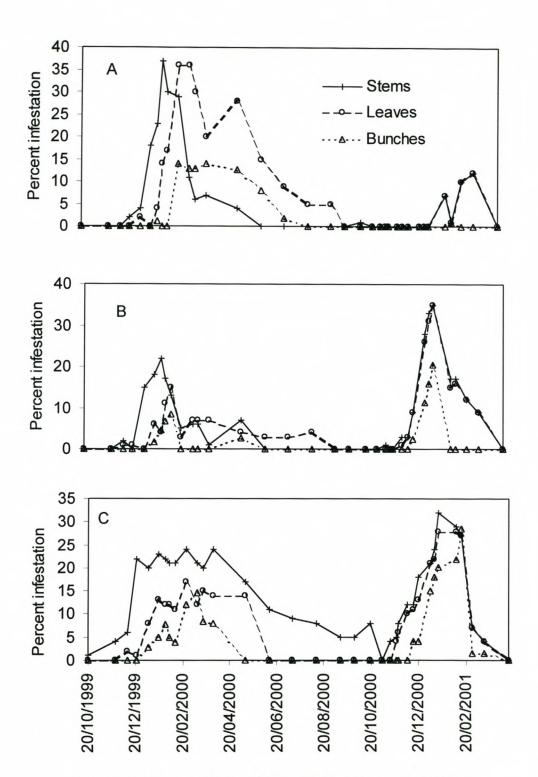
where EMP = emerged parasitoids, LP = all live parasitoids, and UMH = unparasitised mealybug hosts.

### 6.3 RESULTS AND DISCUSSION

P. ficus occurred on the vine trunk in all areas throughout the year. The lowest P. ficus population levels were recorded during the winter months. As temperatures started to increase during November (Fig. 6.1), mealybug colonies appeared on the new growth of the stems (Fig. 6.2 A, B, & C).



**Fig. 6.1.** Mean monthly temperatures for Robertson, Hex River Valley and Stellenbosch for the 1999/2000 and 2000/2001 seasons.



**Fig. 6.2.** Vine mealybug, *Planococcus ficus*, infestation levels on stems, leaves and bunches during two seasons in A, the Hex River Valley; B, Stellenbosch; C, Robertson.

The highest percentage stem infestation was recorded during February in the Hex River Valley (Fig. 6.2A) and Stellenbosch (Fig. 6.2B) and during January in Robertson (Fig. 6.2C). A successional trend of mealybug colonisation was observed on the stems, leaves and bunches (Fig. 6.2A, B, & C) in all three grape growing areas. Early in the season vine mealybugs colonised new growth on the stems, followed by the leaves and eventually bunches towards the end of the season (Fig. 6.2A, B, & C).

The highest percentage infestation of leaves and bunches was recorded during March in the Hex River Valley (1999/2000 season) (Fig. 6.2A), Stellenbosch (1999/2000 season)(Fig. 6.2B) and Robertson (both seasons) areas (Fig. 6.2C). This was followed by a rapid decline in infestation in most cases. Initial high stem infestations early in the season usually resulted in corresponding high bunch infestation levels at harvest (Fig. 6.2 A, B & C). Mealybug infestation of new growth on the stem early in the season can therefore be an early indication of potential bunch infestation and crop loss towards the end of the season.

In most cases the highest numbers of predatory beetles were recorded during early December (Fig. 6.3 A, B & C). *Nephus* spp. were the most abundant, supporting the findings of Whitehead (1957) and Berlinger (1977). Peak population levels of *P. ficus* (February) (Fig. 6.2 A, B & C) occurred after those of the predatory beetles (December), suggesting that predatory beetles did not have a major effect on reducing vine mealybug population levels. The parasitoid

population peak in most cases was during March, about one month after the population peak of their host. Data from yellow sticky traps (Fig. 6.4 A, B &C) in all three areas indicated that *Coccidoxenoides peregrinus* (Timberlake) and the *Anagyrus* sp. were the dominant parasitoids, followed by *Leptomastix dactylopii* (Howard). The former two species could therefore be seen as the major contributors to biological control during the season. The unexpected increase in parasitoid numbers during the 2000/2001 season in the Hex River Valley and Robertson (Figs. 6.4 A & C) areas could be ascribed to more efficient ant control (Tumminelli 1997; Addison & Samways 2000). These higher parasitoid numbers, together with the relatively low mealybug infestation levels towards the end of the 2000/2001 season, suggested that there might have been an increase in the efficiency of biological control.

By plotting parasitoid numbers on their host numbers, a density dependent relationship was evident in all areas and during both seasons (Fig. 6.5 A - F). This further supported the notion that parasitoids were the main biological control agents for *P. ficus*. *L. dactylopii* numbers increased later in the season in all three areas (Fig. 6.4 A, B & C), but were in the minority during this period, suggesting that they played a minor role in the biological control of *P. ficus*.

Plots of predator numbers on the numbers of their prey (Fig. 6.6) showed a clockwise trend, suggesting that there was not a density dependent relationship between the predators and their prey. This supported the contention that they

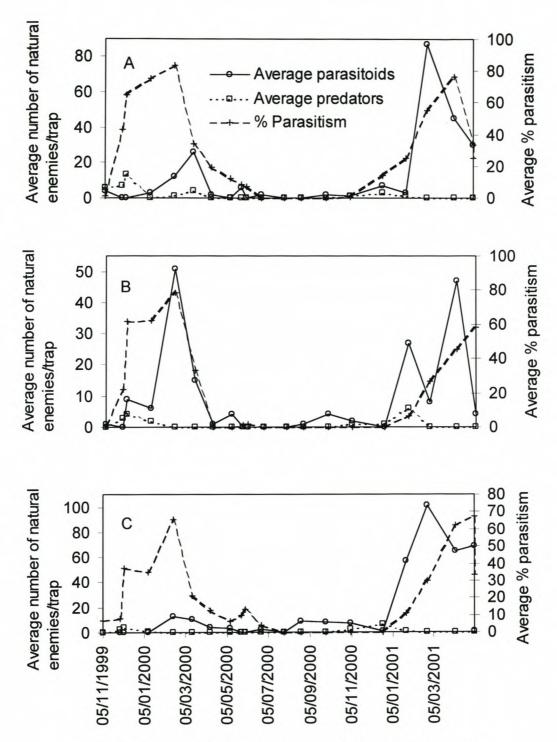
were not as important as the parasitioids (Berlinger 1977) in the regulation of *P. ficus* populations, contrary to the conclusions made by Whitehead (1957).

Mealybug population levels declined from February until the end of each season in each of the three areas despite suitable temperatures (Fig. 6.1). The major mortality factor of *P. ficus* at this time of the season may have been the high parasitoid populations which resulted in high percentage parasitism (Fig. 6.3 A, B & C).

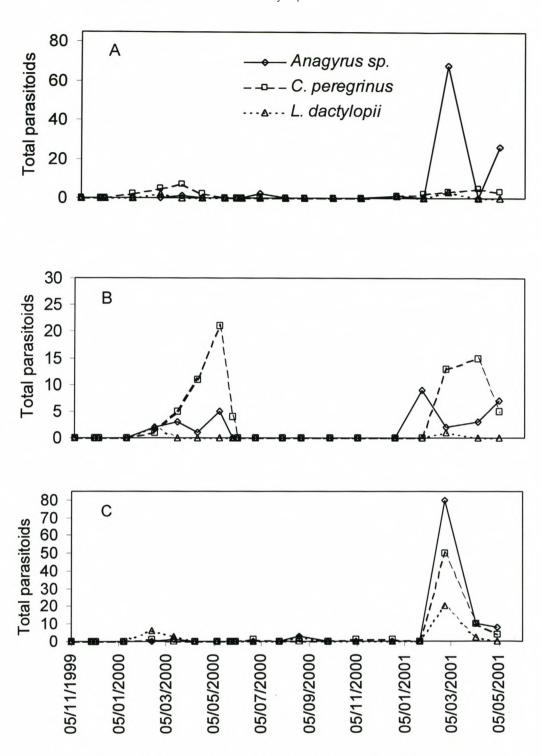
### 6.4 SUMMARY

A successional trend of mealybug colonisation was observed between the different positions on vines in all three grapegrowing areas. Vine mealybugs colonised new growth on the stems early in the season, followed by colonisation on the leaves and eventually bunches towards the end of the season. Initial high stem infestations early in the season usually resulted in correspondingly high bunch infestation levels at harvest. Mealybug infestation of new growth on the stem early in the season can therefore be an early indication of potential bunch infestation and crop loss toward the end of the season.

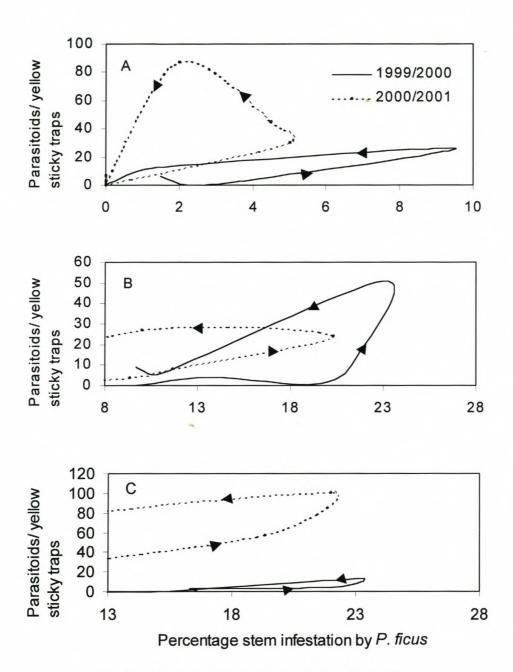
Predatory beetles did not play an important role in the biological control of *P. ficus* pest populations. The hymenopteran parasitoids, *C. peregrinus* and *Anagyrus* sp., however, played a major role in biological control of *P. ficus*. Biological control was however not effective as it was only achieved towards the end of the season and when damage to the crop had already been done.



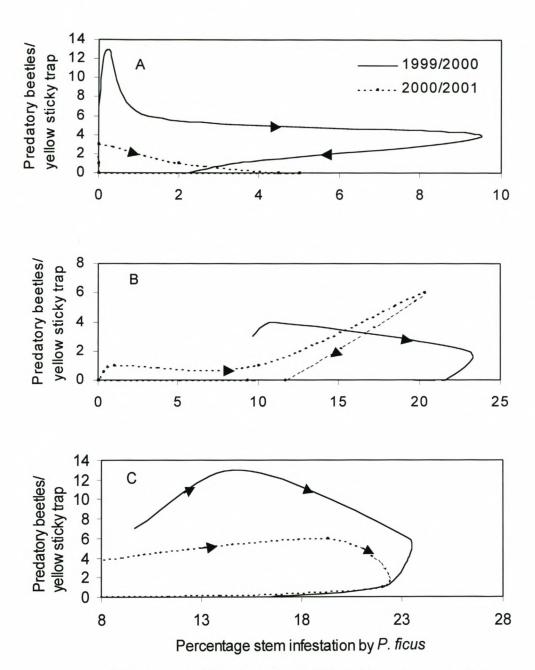
**Figure 6.3**. Average number of coccinellid predators and hymenopteran parasitoids on yellow sticky traps and % parasitism of *Planococcus ficus* in vineyards during 1999/2000 and 2000/2001 in A, The Hex River Valley; B, Stellenbosch; C, Robertson.



**Fig. 6 4.** Hymenopteran parasitoids caught on yellow sticky traps in vineyards during the 1999/2000 and 2000/2001 seasons in A, the Hex River Valley; B, Stellenbosch; C, Robertson.



**Fig. 6.5.** Density dependant relationship between parasitiods and *Planococcus ficus* during the 1999/2000 and 2000/2001 seasons with A, Hex River Valley; B, Stellenbosch and C, Robertson.



**Fig. 6.6.** Density independant relationship between predatory beetles and *Planococcus ficus* during the 1999/2000 and 2000/2001 seasons with A, Hex River Valley; B, Stellenbosch and C, Robertson.

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### **CHAPTER 7**

BIOLOGICAL CONTROL OF THE VINE MEALYBUG, PLANOCOCCUS FICUS (SIGNORET), THROUGH MASS RELEASES OF THE PARASITOID COCCIDOXENOIDES PEREGRINUS (TIMBERLAKE) (HYMENOPTERA: ENCYRTIDAE)

### 7.1 INTRODUCTION

Chemical applications are currently used to control *Planococcus ficus* (Signoret). Several attempts of classical biological control have been made with the importation and release of *Chrysoplatycerus splendens* (Joubert 1943), *Cryptolaemus montrouzieri* (Greathead 1971), *Scymnus guttulatus* and *S. sordidus* (Joubert 1943), *Pseudaphycus angelicus*, *Zarophagus corvinus* (Joubert 1943) and *Anagyrus pseudococci* (Girault) from Israel (Urban 1985). In a study of natural enemies associated with vine mealybug (Chapter 6), 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 the parasitoids played an important role in the biological control of *P. ficus*.

Biological control by mass releases of natural enemies has contributed to the control of several pseudococcid pests (Mineo 1977, Longo 1982, Summy 1986, Smith *et al.* 1988, Smith 1991, Nagarkatti 1992, Reddy & Bhat 1993, Fronteddu *et al.* 1996, Smith *et al.* 1996, Raciti *et al.* 1997). *C. 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). The present study was conducted to investigate the effectiveness of control using mass releases of C. peregrinus as an alternative to chemical control of P. ficus.

### 7.2 MATERIAL AND METHODS

The layout of trial sites and pesticide treatments are described in Chapter 2 (2.2.3.2). Parasitoids were reared as described in Chapter 2.2.1 and distributed in the field in paper distribution bags (Chapter 2.2.2) by stapling one bag in the crown of the vine. One bag containing approximately 1000 parasitoids was stapled to one of the vines in each of the 20 plots. Therefore, the release rate was  $\pm$  20 000 parasitoids/ha. Six and five releases were made at monthly intervals starting during November in 1999/2000 and November 2000/2001 giving a total of 120 000 and 100 000 parasitoids released per site during the 1999/2000 and 2000/2001 seasons respectively. Parasitoids usually emerged over a period of one month, after which another release was made (Table 7.1).

**Table 7.1**. Monthly *Coccidoxenoides peregrinus* releases in the nine trial sites during the 1999/2000, and 2000/2001 seasons.

Date	Number released	Date	Number released
	per site		per site
5/11/1999	20 000	1/11/2000	20 000
7/12/1999	20 000	27/12/2000	20 000
4/01/2000	20 000	31/01/2001	20 000
3/02/2000	20 000	28/02/2001	20 000
8/03/2000	20 000	30/03/2001	20 000
6/04/2000	20 000		
Total released per	120 000		100 000
season			

# 7.2.1 Evaluation of parasitoid releases

Evaluation of the effectiveness of released parasitoids was done by determining:

- Vine mealybug stem infestation levels by regular monitoring using the sampling system described in Chapter 2.2.5.1.
- C. peregrinus counts on yellow sticky traps were used as described in Chapter 2.2.5.2.
- Percentage parasitism was determined as described in Chapter 6. The average percentage parasitism was calculated for the whole season for each of the treatments.

 Crop loss due to vine mealybug infestation was determined by sampling bunches as described in Chapter 2.2.5.1. The three assessments closest to harvest were summed and averaged as an estimate of crop loss for the season.

## 7.2.2 Data analysis

Stem infestation, percent parasitism and trap catch data collected during the season were summarised by converting them to insect days (Ruppel 1985) by averaging the data from two consecutive sampling dates and multiplying by the number of days between these dates. These were summed to give the total number of insect days (Ruppel 1985). These data were used in a split plot analysis with the three areas as the main plots. Treatments (release, buffer and control plots) and years were the main effects in the sub-plots. Prior to the analysis the data were log transformed to stabilise the variances. The split plot experimental design was also used to analyse data pertaining to percentage crop loss.

### 7.3 RESULTS

#### 7.3.1 Stem infestation

There were differences in stem infestation between areas (p<0.001; Table 7.2), with lower levels of stem infestation in the Hex River Valley than in Stellenbosch and Robertson (Fig. 7.1 A, B & C).

**TABLE 7.2**. Split plot analysis of stem infestation in three areas (main plots) during two seasons (1999/2000, 2000/2001) and in three treatments (release, buffer and control plots).

Source	SS	Degrees	MS	F	Р	
		of				
		freedom				
Area	21.67	2	10.83	69.15	< 0.001	
Error	0.63	4	0.16			
Season	0.77	1	0.77	2.1	0.16	
Treatment	1.09	2	0.54	1.49	0.24	
Area*season	1.07	2	0.54	1.46	0.25	
Area*treatment	1.15	4	0.29	0.78	0.54	
Season*treatment	1.1	2	0.55	1.5	0.24	
Error	13.2	36	0.37			

## 7.3.2 Yellow sticky traps

There was no difference in the number of *C. peregrinus* caught on yellow sticky traps between the three areas or between the treatments (Table 7.3). There were differences between seasons (Table 7.3). More parasitoids were caught on the yellow sticky traps during the second season than during the first (Fig. 7.2). The differences were not as marked in the Hex River Valley as in the Stellenbosch and Robertson areas (Table 7.4). This discrepancy resulted in interactions between area and season (Table 7.3).

**TABLE 7.3.** Split plot analysis of cumulative insect days of *Coccidoxenoides* peregrinus caught on yellow sticky traps in three areas (main plots) during two seasons (1999/2000, 2000/2001) and in three treatments (release, buffer and control plots).

Source	SS	Degrees	MS	F	р
		of			
		freedom			
Area	1.78	2	0.89	2.41	0.21
Error	1.47	4	0.37		
Season	17.04	1	17.04	58.41	< 0.001
Treatment	1	2	0.5	1.72	0.19
Area*season	5.21	2	2.6	8.93	0.001
Area*treatment	0.69	4	0.17	0.59	0.67
Season*treatment	0.01	2	0.01	0.02	0.98
Error	10.5	36	0.29		

**TABLE 7.4**. Cumulative insect days of *Coccidoxenoides peregrinus* caught on yellow sticky traps in three areas (main plots) during two seasons (1999/2000, 2000/2001) and in three treatments (release, buffer and control plots).

Area	Control		Bu	Buffer		ase
Season	1999/	2000/	1999/	2000/	1999/	2000/
	2000	2001	2000	2001	2000	2001
Hex River Valley	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

## 7.3.3 Percentage parasitism

There were no differences in percent parasitism between areas or treatments (Table 7.5). There was a difference between seasons (Table 7.5), with a slightly higher average percent parasitism during the first season than during the second (Tables 7.5, 7.6; Fig. 7.3 A, B & C).

**TABLE 7.5.** Split plot analysis of average percentage parasitism of *Planococcus* ficus by *Coccidoxenoides peregrinus* in three areas (main plots) during two seasons (1999/2000, 2000/2001) and in three treatments (release, buffer and control plots).

Source	SS	Degrees	MS	F	р
		of			
		freedom			
Area	0.29	2	0.15	0.71	0.55
Error	0.83	4	0.21		
Season	11.75	1	11.75	56.66	<0.001
Treatment	0.45	2	0.23	1.1	0.35
Area*season	0.46	2	0.23	1.1	0.34
Area*treatment	1.44	4	0.36	1.73	0.16
Season*treatment	0.47	2	0.23	1.13	0.34
Error	7.47	36	0.21		

**TABLE 7.6**. Average percentage *P. ficus* parasitism in three areas (main plots) during two seasons (1999/2000, 2000/2001) and in three treatments (release, buffer and control plots).

Area	Control		Buffer		Release	
Season	1999/2	2000/	1999/	2000/	1999/	2000/
	000	2001	2000	2001	2000	2001
Hex River	35.44	25.44	42.44	32	44.33	32.66
Valley						
Robertson	28.77	19.22	37.22	31.22	37.55	32.67
Stellenbosch	20.77	22.55	25.22	37.88	26.22	38.44
Total	28.33	22.41	34.96	33.7	36.03	34.59

# 7.3.4 Infestation at harvest (crop loss)

There were significant differences in *P. ficus* bunch infestations (Table 7.7) between treatments. There were also differences between areas (Table 7.7) with less bunch infestation due to *P. ficus* infestations in the Hex River Valley than in Stellenbosch and Robertson (Table 7.8). There were also interactions between areas and treatments (Table 7.7). This was because there was less bunch infestation in the release and buffer treatments than in the control in the Hex River Valley and in Stellenbosch but not in Robertson (Table 7.8). The highest bunch infestation during both seasons was in the control treatments, while the bunch infestation in the buffer and release treatments was similar, but lower than in the control (Table 7.8).

**TABLE 7.7.** Split plot analysis of bunch infestation data in three areas (main plots) during two seasons (1999/2000, 2000/2001) and in three treatments (release, buffer and control plots).

Source	SS	Degrees of	MS	F	р
		freedom			
Area	568.89	2	284.45	492.66	<0.001
Error	46.29	36	1.29		
Season	4.76	1	4.76	3.7	0.06
Treatment	21.19	2	10.6	8.24	0.001
Area*season	2.87	2	1.43	1.11	0.34
Area*treatment	28.1	4	7.03	5.46	0.001
Season*treatment	2.69	2	1.34	1.04	0.36
Area*season*treatment	2.31	4	0.58	0.45	0.77

**TABLE 7.8.** Mean percentage bunch infestation in release, buffer and control plots due to vine mealybug (*Planococcus ficus*) infestation at harvest in three grape growing areas during two seasons.

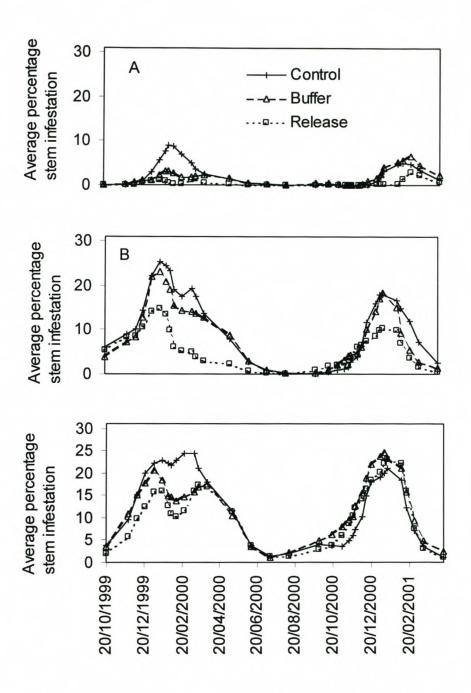
Grapegrowing	Mean % crop loss							
area	Control		Buffer		Release			
	1999/	2000/	1999/	2000/	1999/	2000/		
	2000	2001	2000	2001	2000	2001		
Hex River Valley	2.3	0.03	1.11	0.03	0.05	0		
Stellenbosch	8.6	7.3	4.05	3.9	6.5	5.8		
Robertson	8.5	8.2	8.01	8.5	6.9	8.3		
Average	6.47	5.18	4.39	4.14	4.48	4.7		

#### 7.4 DISCUSSION

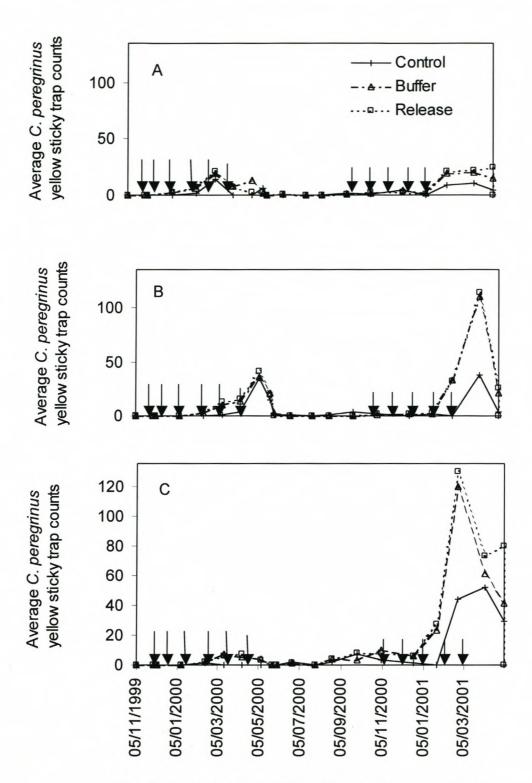
No differences were detected in the percentage *P. ficus* stem and bunch infestation, *C. peregrinus* numbers on yellow sticky traps and percentage parasitism between the release, buffer and control blocks. The large plots made it logistically difficult to increase the number of replicates, which would 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.

P. ficus stem infestation in the Hex River Valley was lower than in Robertson and Stellenbosch. The lower stem infestations in the Hex River Valley did not influence the number of parasitoids caught or the 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. 7.1 A, B & C), although this was not reflected in the formal analysis (Table 7.3). This may indicate that the released C. peregrinus could also have improved biological control in the buffer blocks.

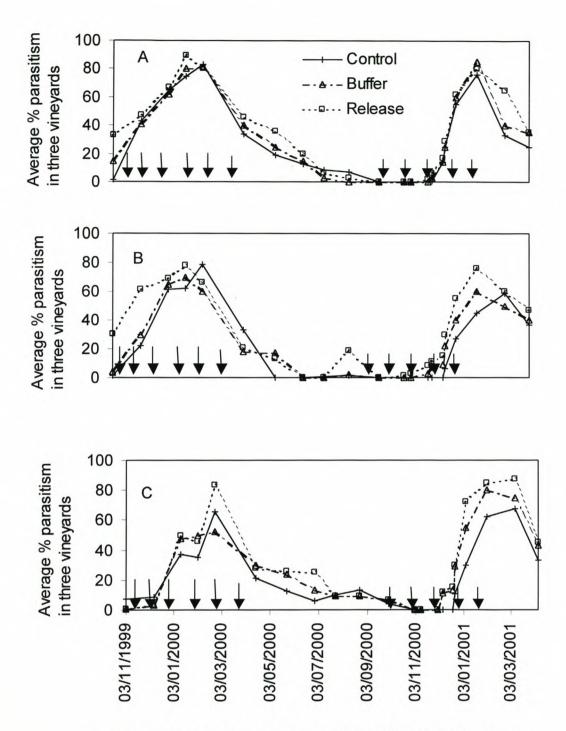
P. ficus bunch infestations at harvest (Table 7.8) were similar in the release, buffer and control treatments in the Robertson area. However, they were lower in the buffer and release treatments than in the control in the Stellenbosch and Hex River Valley areas. Therefore, it appeared as if the releases successfully supplemented naturally occurring *C. peregrinus* populations in these areas.



**Fig. 7.1.** Average stem infestations during two seasons by *Planococcus ficus* in blocks into which *Coccidoxenoides peregrinus* was released, in buffer and control blocks in three vineyards in A, Hex River Valley; B, Stellenbosch; C, Robertson.



**Fig. 7.2.** Average number of *Coccidoxenoides peregrinus* caught on yellow sticky traps during two seasons in blocks in which *C. peregrinus* was released and in buffer and control blocks in three vineyards in A, Hex River Valley; B, Stellenbosch; C, Robertson. Arrows indicate the release of 20 000 *C. peregrinus*/ha.



**Fig. 7.3**. Average percentage parasitism of *Planococcus ficus* during two seasons in blocks in which *Coccidoxenoides* peregrinus was released and in buffer and control blocks in three vineyards in A, Hex River Valley; B, Stellenbosch; C, Robertson. Arrows indicate a release of 20 000 *C. peregrinus /*ha.

#### 7.5 SUMMARY

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 suppressing of high mealybug population levels through the use of dormant chemical treatments.

Augmentative releases were 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. *C. peregrinus* is commercially available and can be used by producers as an alternative to chemical control. 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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#### **CHAPTER 8**

# THE USE OF DEGREE-DAY ESTIMATION AND MODELING IN AN INTEGRATED VINE MEALYBUG MANAGEMENT SYSTEM

#### 8.1 INTRODUCTION

Heat accumulation is widely used by economic entomologists to predict the outbreak of pest populations. It is expressed in degree-days (°D), and is determined by the rate of development of the insect at different temperatures. Information resulting from the use of °D models can be used as additional inputs in a pest management system for a key pest such as *Planococcus ficus* (Signoret). Degree-days were estimated for two seasons in three different vine growing areas, and correlated with known pest infestation levels in these areas.

In addition, this information and information from previous chapters were used to construct a simple decision model for managing *P. ficus* in South African vineyards. An expert system model similar to those described by Norton & Mumford (1993) was used. In the past, management of *P. ficus* pest populations relied on the application of chemicals. Information gathered in the current study can contribute to the development of a model making use of ecological and biological information resulting in increased efficiency of *P. ficus* control, and a reduction in chemical applications.

The model contains discussed in this chapter three of the four steps in the development of an expert system (Norton & Mumford 1993). These three steps included problem structuring, knowledge acquisition and knowledge engineering and encoding (Chapter 11, Norton & Mumford 1993). The final step of verification, validation and testing need to be investigated in future field work.

#### 8.2 MATERIAL AND METHODS

#### 8.2.1 Degree day estimation

Daily weather data were used to estimate the accumulated number of °D for both *P. ficus* and *C. peregrinus* in Stellenbosch, Robertson and the Hex River Valley using the methods described by Baskerville & Emin (1969). The number of degree days required for *P. ficus* to complete one generation was 235 °D and for *Coccidoxenoides peregrinus* 500 °D (Chapter 4).

The lower threshold for development of *P. ficus* was 16.59 °C (case A, B and C, Baskerville & Emin 1969) (Chapter 4), while the upper threshold was 35.61 °C (case C, Baskerville & Emin 1969) (Chapter 4). These values were used for estimates of the °D development for *P. ficus*. The lower threshold for development of *C. peregrinus* was 8.85 °C. This was used for estimates of the °D development for the parasitic wasp in cases A and B in Baskerville & Emin (1969). No estimates of the upper threshold for development of *C. peregrinus* were available. Therefore, case C (Baskerville & Emin 1969) was not used (Chapter 4).

The estimation of accumulated °D for the 1999/2000 and 2000/2001 seasons was started from the beginning of September as temperatures started to increase at this stage (Chapter 6). The number of generations completed from the beginning of September during each season for each insect and area was also estimated.

#### 8.2.2 Correlation studies

The operating characteristic curves (OC curves) (Chapter 5) for *P. ficus* infestations on stems suggested that the best time for intervention against *P. ficus* was when more than 2 % of the stems were infested. °D were correlated with both stem and bunch infestations (Chapters 6 and 7) in the three vine growing areas of Stellenbosch, Robertson and the Hex River Valley. Correlations were estimated using the averages of stem and bunch infestations obtained from the control, buffer and release blocks combined.

#### 8.3 RESULTS

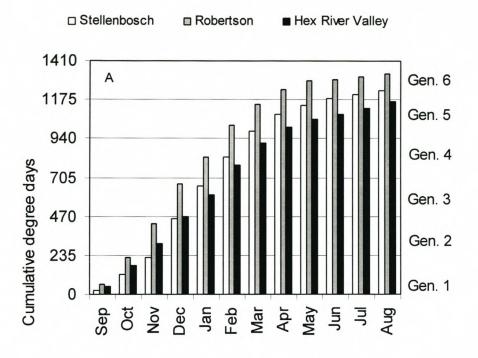
### 8.3.1 Degree day estimation for *Planococcus ficus*

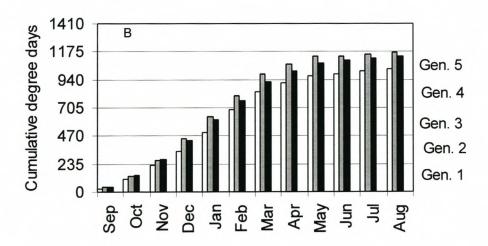
The number of °D for the development of *P. ficus* accumulated rapidly from October to April in all areas (Fig. 8.1). This was also the period during which *P. ficus* populations increased rapidly (Fig. 6.2 A, B & C; 7.1 A, B & C). Increases in °D accumulation ceased after April, and remained at very low levels in all three

areas until September (Fig. 8.1A) or October (Fig. 8.1B), which coincided with very low mealybug population levels (Fig. 6.2 A, B & C; 7.1 A, B & C).

The estimated number of generations was higher during the 1999/2000 season than during the 2000/2001 season in all three areas (Fig. 8.1). In addition, infestation levels during the 1999/2000 season were higher than during the 2000/2001 season (Figs. 6.2 A, B & C; 7.1 A, B & C).

Robertson had the highest accumulated °D for *P. ficus* during both seasons (Fig. 8.1), and *P. ficus* infestation levels were also higher in this area than in the other two areas (Figs. 6.2 & 7.1). Therefore, it appeared as if temperatures were more suitable for *P. ficus* population development in Robertson than in Stellenbosch and the Hex River Valley. Differences in accumulated °D between the three areas were less pronounced (Fig. 8.1) during both seasons than differences between *P. ficus* infestation levels (Figs. 6.2 & 7.1). Relatively low *P. ficus* infestation levels on the stems were recorded in the Hex River Valley (< 10%) compared to Robertson (> 15 %) and Stellenbosch (> 15 %). The spray programme used in the Hex River Valley was similar to those used in the other areas (Table 7.1) and could therefore not have influenced *P. ficus* populations differently than in the other areas.





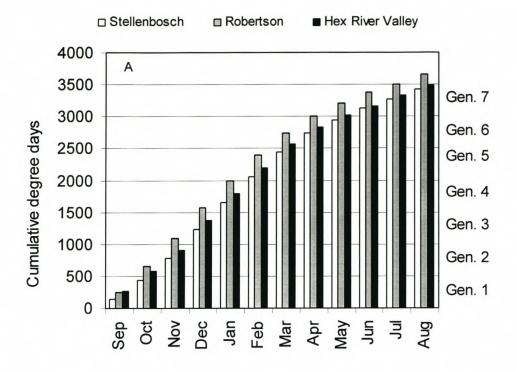
**Fig 8.1**. Estimated cumulative degree days for *Planococcus ficus* and number of generations (right axis) in the three areas during A, 1999/2000 and B, 2000/2001 seasons.

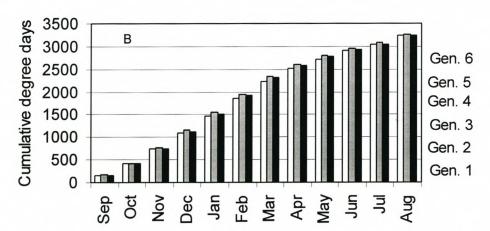
#### 8.3.2 Degree day estimation for Coccidoxenoides peregrinus

Degree day accumulation for *C. peregrinus* was constant throughout the year, including the cooler months (Fig. 8.2). *C. peregrinus* development was not negatively influenced by winter temperatures. Parasitoid numbers remained relatively high until May during both seasons (Figs. 6.3 & 6.4), particularly in the Stellenbosch area. More *C. peregrinus* generations were estimated during the 1999/2000 season than during the 2000/2001 season in all three areas (Fig. 8.2). Temperatures may therefore have been more suitable for *C. peregrinus* population development during the 1999/2000 than during the 2000/2001 season. However, parasitoid numbers did not reflect this (Figs. 6.4 & 7.2), probably because of the reduced mealybug populations.

#### 8.3.3 Correlation between P. ficus infestations and °D

Correlation coefficients between cumulative °D and stem infestation (Table 8.1) and cumulative °D and bunch infestation (Table 8.2) were not consistent, and in some cases the correlations were poor. It was not possible to calculate correlation coefficients between bunch infestation and cumulative °D in Hex River Valley during 2000/2001 as there was no bunch infestation.





**Fig 8.2**. Estimated cumulative degree days for *Coccidoxenoides peregrinus* and number of generations (right axis) in the three areas during A the 1999/2000 and B, the 2000/2001 seasons.

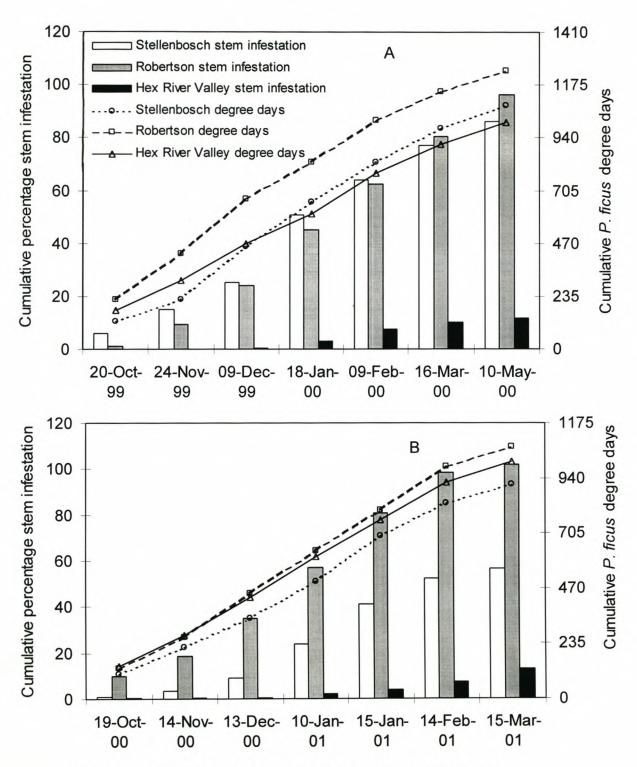
**TABLE 8.1.** Correlation (r) between cumulative degree days and percentage stem infestation in the three grape growing areas during two seasons.

Grapegrowing	Block 1		Block 2		Block 3	
area	1999/	2000/	1999/	2000/	1999/	2000/
	2000	2001	2000	2001	2000	2001
Stellenbosch	0.51	0.72	0.81	0.38	0.54	0.34
Robertson	0.43	0.24	0.49	0.22	0.31	0.25
Hex River Valley	0.68	0.92	0.85	0.92	0.89	0.82

**TABLE 8.2.** Correlation (r) between cumulative degree days and percentage bunch infestation in the three grape growing areas during two seasons.

Grapegrowing	Block 1		Block 2		Block 3	
area	1999/	2000/	1999/	2000/	1999/	2000/
	2000	2001	2000	2001	2000	2001
Stellenbosch	0.55	0.52	0.55	0.49	0.46	0.40
Robertson	0.91	0.49	0.94	0.49	0.92	0.56
Hex River Valley	0.43	-	0.43	-	0.25	-

P. ficus stem infestation levels in the Hex River Valley were low (Figs. 8.3 A & B) compared to those in Stellenbosch and Robertson, despite little differences in the cumulative degree days between the three areas.



**Fig. 8.3.** Cumulative degree days (lines) and percentage stem infestation (bars) in three areas during A, 1999/2000 and B, 2000/2001.

There was a rapid increase in *P. ficus* stem infestation during both seasons in the Stellenbosch and Robertson areas (Fig. 8.4 A & B). In the Hex River Valley, however, this increase started much later during both seasons. This may indicate that monitoring *P. ficus* infestations using assessments of stem infestations must start earlier in Stellenbosch and Robertson, than in the Hex River Valley.

#### 8.3.4 Modeling

A time driven decision chart is presented in Table 8.3. Decisions are divided into three management periods, winter (Table 8.3a); spring and early summer (Table 8.3b); late summer and autumn (Table 8.3c). These periods can each be seen as an initial pathway to start decision-making.

Due to the relationship between ants and mealybugs during the spring to autumn periods, two types of monitoring actions are needed. Monitoring activity of ants (group 1)(Ueckermann, 1998) and mealybugs (group 2) are specified. These are done separately (Table 8.3b). Monitoring ant activity can be done by classifying presence or absence of ants on individual vines (Ueckermann, 1998). The vines used can be the same used for monitoring mealybugs.

From these monitoring actions sub-pathways are defined in terms of seasons (Norton & Mumford, 1993). These sub-pathways include:

Winter: Less than 2 % P. ficus infestation during the previous season

More than 2 % P. ficus infestation during the previous season

Spring and early summer. group 1) No ant activity

Ant activity

group 2) Less than 2 % P. ficus infestation during the

current season

More than 2 % P. ficus infestation during the

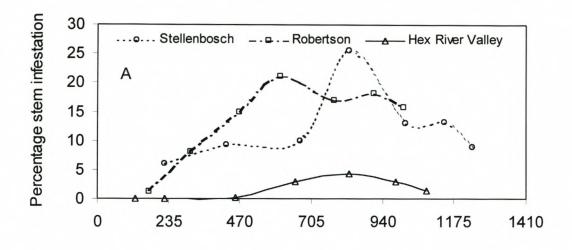
current season

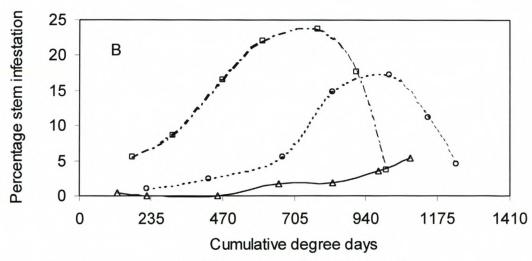
Late summer and autumn: P. ficus infestation less than 2 %

P. ficus infestation more than 2 %.

For each sub-pathway there is a choice of management actions or recommendations. Therefore, each of the initial sub-pathways leads to the specification of a problem typical to that time of the season, which, in turn, leads to management choices best suited to that time and area.

Different management actions are possible for each of the three time periods. During the *winter* (June – August), temperatures were sub-optimal for *P. ficus* development. The *P. ficus* populations overwintered on the main stem and roots (Chapter 6). This was the best time to spray against this pest if infestation levels exceeded 2 % during the previous season (IPW, 2000). Targeting the pest at this stage should be easier, as new shoot growth and leaves were absent.





**Fig. 8.4**. Percentage *Planococcus ficus* stem infestation plotted against cumulative degree days in three grapegrowing areas during A, 1999/2000 and B, 2000/2001.

During the *spring and early summer* season (September – December) temperatures were suitable for *P. ficus* and ant activity (Chapters 6 & 7; Ueckermann 1998). Therefore ant control was important during this period (Chapter 5). Chemical control and biological control, using mass releases of *C. peregrinus*, should be done when *P. ficus* infestation levels exceed 2 % (Chapters 5 and 7).

During the *late summer and autumn* period (January – May)(Chapter 6 & 7) biological control played an important role (Chapter 6), which suggested that chemical sprays should be limited to vines weakened by severe *P. ficus* infestations (Chapter 5). By spraying only marked vines the detrimental effect of pesticides on beneficials such as *C. peregrinus* (Walton & Pringle 1999) and *Nephus 'boschianus'* (Walton & Pringle 2001) will be limited to those specific areas. Mass releases of *C. peregrinus* should continue in table gape blocks with low *P. ficus* tolerance as a control measure for this pest.

This decision chart can be presented as a simple decision model similar to those described by Norton & Mumford (1993). Management actions for Table 8.3a (*winter*) include:

Action 1: IF *P. ficus* infestation during the previous summer period did not exceed 2 % THEN no action.

Action 2: IF *P. ficus* infestation during the previous summer period did exceed 2 % THEN spray all marked and infested vines as well as two vines on either side at a fortnightly interval before budbreak.

Management actions summarised in Table 8.3b (spring and early summer) include:

Action 1: IF there is no ant activity THEN do not apply stem barrier treatments

Action 2: IF there is ant activity THEN spray stem barrier treatments

Action 3a: IF in Stellenbosch and Robertson THEN start monitoring at the beginning of October AND if *P. ficus* is less than 2 % THEN do not spray/release *C. peregrinus*.

Action 3b: IF in Stellenbosch and Robertson THEN start monitoring at the beginning of October AND if *P. ficus* is more than 2 % THEN spray all marked and infested vines as well as two vines on either side/release *C. peregrinus*.

Action 4a: IF in the Hex River Valley THEN start monitoring at the beginning of November AND if *P. ficus* is less than 2 % THEN do not spray/release *C. peregrinus*.

Action 4b: IF in the Hex River Valley THEN start monitoring at the beginning of November AND if *P. ficus* is less than 2 % THEN spray all marked and infested vines as well as two vines on either side /release *C. peregrinus*.

Management actions summarised in *late summer and autumn* (Table 8.3c) include:

Action 1: IF P. ficus stem infestation is less than 2 % THEN do not spray.

Action 2: IF *P. ficus* stem infestation more than 2 % THEN spot spray highly infested/weakened vines.

**Table 8.3**. Decision chart for integrated *P. ficus* management during three seasonal periods.

## a) Winter period (June – August)

Monitoring/ action	Qualifier and value	Management action or recommendation
P. ficus infestation during previous	Less than 2 % infestations	Do not spray
summer period	More than 2 % infestations	Spray all marked and infested vines as well as two vines either side at a fortnightly interval before budbreak

# b) Spring and early summer (September – December)

Monitoring/ action	Qualifier and value	Management action or recommendation
1) Ant activity	a. No Activity	a. Do not spray ant stem barrier
monitoring from the		treatments
beginning of	b. Activity	b. Spray ant stem barrier treatments
September		

Table 8.3 b continued

Monitoring/ action	Qualifier and	Management action or
	value	recommendation
2a) Mealybug stem	a. P. ficus	a. Do not spray
monitoring in	infestation less than	
Robertson and	2% infestations	
Stellenbosch starting	b. P. ficus	b. Spray all marked and infested
at the beginning of	infestation more	vines as well as two vines either side
October	than 2%	OR release C. peregrinus
	infestations	
2b) Mealybug stem	a. P. ficus	a. Do not spray
monitoring in the Hex	infestation less than	
River Valley starting at	2% infestations	
the beginning of	b. P. ficus	b. Spray all marked and infested
November	infestation more	vines as well as two vines either side
	than 2%	OR release C. peregrinus
	infestations	

# c) Late summer and autumn (January – May)

Monitoring/ action	Qualifier and value	Management action or recommendation
P. ficus infestation monitoring	a. Less than 2% infestations	<ul> <li>a. Do not spray (biological control is at its peak)</li> </ul>
, and the second	b. More than 2% infestations	b. Spot spray highly infested/weakened vines (biological control is at its peak and natural enemies should be allowed refuge sites)

#### 8.4 DISCUSSION

Differences in accumulated °D between areas were greater for *P. ficus* (Fig. 8.1) than for C. peregrinus (Fig. 8.2), particularly during the 1999/2000 season. The number of °D accumulated for P. ficus was noticeably lower in the Hex River Valley than in the other areas, while the number of °D accumulated for C. peregrinus was similar in the three areas. Therefore, the rate of population development of P. ficus relative to that of C. peregrinus in the Hex River Valley may have been more favourable for biological control than in the other two areas. This was supported by average percentage parasitism (Table 7.6) during 1999/2000 where the highest average percentage parasitism was recorded in the Hex River Valley. However, other factors may also be of importance in regulating mealybug infestations. Vine architecture in table grape blocks (Hex River Valley) differs considerably from that in the wine grape blocks (Stellenbosch and Robertson). The main stem, new growth areas and bunches are more exposed to chemical sprays in table grape blocks compared to wine grape blocks. Penetration of chemicals may therefore be better in table grapes than in wine grapes, which may have resulted in more efficient chemical control in the Hex River Valley. Vines in wine grape vineyards are more closely spaced at 3300 vines ha<sup>-1</sup> compared to 1800 vines ha<sup>-1</sup> in table grape growing areas. This may lead to the creation of more refuge sites for mealybugs because of the presence of more leaves, stems and bunches per unit area. Wine grape berries in bunches are also more tightly packed than in table grape bunches. Loosely packed berries in table grape bunches may contribute to easier penetration of chemicals during spraying.

There was poor correlation between cumulative °D and infestation levels of both stems and bunches. However, there appeared to be an indirect qualitative relationship between bunch infestation and cumulative °D. During the 1999/2000 season more °D were accumulated than during the 2000/2001 season. During the former season there were also more generations and higher levels of bunch infestation than during the latter season. In addition in the Robertson area more °D were accumulated, and there were more generations. The levels of infestation of bunches were higher in this area than in the other two areas.

The estimated number of vine mealybug generations of between five and six per year estimated in this study (Fig. 8.1) supported work done by Kriegler (1954) who recorded 6 generations per year on sprouting potatoes in an outdoor insectary. Duso (1990) recorded three annual generations in Italy.

#### 8.5 SUMMARY

Favourable temperatures for *P. ficus* development occurred from October to the end of April. The Robertson area appeared to have the most suitable temperatures for *P. ficus* development. Lower temperatures did not have the same negative effect on the rate of population development of *C. peregrinus* development as on that of *P. ficus*. Therefore, the relatively cooler summer

periods in Stellenbosch and Hex River Valley may have aided biological control by *C. peregrinus*. Low winter temperatures did not slow the accumulation of *C. peregrinus* degree days as much as that of *P. ficus*.

Stem infestation levels started to increase rapidly earlier in the season in Stellenbosch and Robertson than in the Hex River Valley. This may indicate that monitoring stem infestation should start earlier in the two former regions. The percentage of stems infested with *P. ficus* started decreasing when temperatures were still suitable for population development, indicating that some mortality factor, such as parasitism, was important later in the season. Infestation levels appeared to be affected by a number of factors including temperature, ant activity, architecture of the vine and tightness of the berries in the bunch.

The simple decision chart could also be computer based. It should be useful to growers as it is a summary of the knowledge acquired during the current study. The final step of four in the development of an expert system, namely verification, validation and testing needs to be carried out under a range of field conditions.

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#### **CHAPTER 9**

#### SUMMARY OF RESEARCH RESULTS

A survey of the species of mealybugs occurring in vineyards in the Western Cape Province indicated that *Planococcus ficus* (Signoret) was the dominant mealybug. The largest populations were above ground. However, the first records of *P. ficus* on roots of grapevines were obtained during this study. To date control of *P. ficus* has focused on the above ground plant parts, but if leafroll virus is to be effectively controlled, measures for controlling the underground populations will also have to be developed. In addition information on the biology of these populations is required.

Pseudococcus longispinus (Targioni) was found on vines to a limited extent and is also a vine leafroll virus vector. Pseudococcus viburnii (Maskell), another vine leafroll vector, was found on roots of certain weeds growing in vineyards. The latter is polyphagous and has been recorded on vines in other parts of the world. The fact that these species were detected in the survey suggests that regular surveys of pseudococcids on vines should be conducted as it is possible that the pest status of these currently more minor polyphagous mealybugs could change.

The survey of natural enemies indicated that the *Nephus* spp. beetles were the dominant specific predators occurring early in the season. Three parasitoids,

Anagyrus sp., Coccidoxenoides peregrinus (Timberlake) and Leptomastix dactylopii (Howard) were dominant in the areas sampled. A density dependent relationship was found between these parasitoids and *P. ficus*, illustrating the importance of these parasitoids in biological control of *P. ficus*.

Developmental studies on *P. ficus* and *C. peregrinus* indicated that both insects were well adapted to temperatures in the Western Cape Province. The parasitoid was better adapted to high and low temperatures, indicating that it was active in a wider temperature range than the pest. This makes it a good candidate for biological control of *P. ficus* throughout the season. *P. ficus* activity can start as early as October. However, population levels remain low until the middle to the end of November. Bunch infestations start during January. Therefore, preventative releases of *C. peregrinus* should commence early in November. The necessity for *C. peregrinus* releases can be determined using the systematic presence-absence sampling system with known levels of experimental error. Stems were the most suitable plant part to sample as this provided an early warning for pending bunch infestations. An OC curve for stem sampling suggested that a decision to intervene at a 2 % infestation level of stems would not result in under intervention in 94 % of the cases. Field work is required to verify these findings.

The main limiting factor in population development during the beginning of the season (September) appeared to be temperature. However, towards the end of

the season the presence of natural enemies was the main limiting factor.

Anagyrus sp., C. peregrinus and L. dactylopii were the major natural enemies of vine mealybug involved. Biological control was severely hampered by ants in the Robertson area. Therefore, attention should be given to ant control in areas such as Robertson.

Mass releases of the parasitoid, *C. peregrinus*, can be used to augment biological control of *P. ficus* pest populations. This technique succeeded in suppressing *P. ficus* infestation levels to below the economic injury level of 5 % in the Hex River Valley. Although the parasitoid also suppressed *P. ficus* in the Stellenbosch and Robertson areas, infestation levels could not be kept below the economic threshold level of 5 %. However, the level of control using augmentative releases was at least as good as chemical control in these areas.

C. peregrinus is commercially available and augmentative releases can be used as an alternative strategy for managing P. ficus pest populations. Future work needed to improve this technique includes refining packaging and transport systems from the insectary to producers to increase survival. The use of cold storage to manipulate adult emergence should also be investigated, as this could improve the shelf life of the parasitoids. Further, the quality of insectary reared parasitoids has been stressed by Luck et al. (1999). Parameters which have been taken into account in addressing quality of consignments of insectary reared parasitoids include percentage survival, fecundity and searching ability.

Quality control procedures should be built into the augmentative release system, as this will increase the reliability of biological control.

The use of augmentative releases of parasitoids is a new technology for producers. Inadvertent incorrect use of this technology can also be seen as a risk. This risk can be avoided by intensive training.

A simple decision chart was developed for the integrated management of *P. ficus*. This decision chart can be used to optimise control actions against *P. ficus* as the season progresses. Three of the four steps in the development of an expert system have been completed. These are problem structuring, knowledge acquisition and knowledge encoding (Chapter 11, Norton & Mumford 1993). The final step of verification, validation and testing needs to be done under different field conditions.

Recently pheromone traps for monitoring *P. ficus* activity have been developed. These should be investigated as an additional monitoring tool, which can also provide information that can be incorporated into the decision chart. The isolation of this pheromone provides the possibility of using mating disruption to suppress *P. ficus* population levels. This, together with mass releases of natural enemies against *P. ficus* may in future become more attractive especially with tighter regulatory issues against the use of insecticides.

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