Chemical Stem Barriers for the Control of Ants (Hymenoptera: Formicidae) in Vineyards

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Ants significantly reduce the efficacy of biological control of the mealybug *Planococcus ficus* (Signoret) in vines. Two trials were conducted to find a cost-effective method for ant control that is environmentally friendly, practicable and acceptable in an integrated pest management programme. Thirteen chemical stem barriers were assessed for the control of two ant species, *Linepithema humile* (Mayr) and *Anoplolepis custodiens* (Smith), in two field trials during two seasons. Four of the treatments that showed high efficacy in the field trials were also evaluated in two simulated field trials for the control of *L. humile* and *Anoplolepis steingroeveri* (Forel) due to high variability in pre-treatment counts that occurred in the field trials. Treatments showing the highest efficacy against *L. humile* and *A. custodiens* in field trials were the chlorpyrifos-impregnated band and the terbufos slow-release band. Alphacypermethrin SC at 10 mL/L was effective against *L. humile* and has subsequently been registered as a chemical stem barrier on vines. The treatment showing the highest efficacy against *A. steingroeveri* in the simulated field trial was alphacypermethrin SC at 20 mL/L. In the simulated field trial, a decline in ant infestation was observed five to six weeks after application of treatments. The most likely explanation is that chemical stem barriers result in ant mortality, although other reasons for this decline are discussed. It is recommended that suitable bioassay techniques, which expose ants to the treated substrate for a limited period, thereby simulating field conditions, be developed in order to determine if chemical stem barriers result in ant mortality.

Certain ants significantly reduce the efficacy of biological control of the mealybug *Planococcus ficus* in vines by coccinellid predators and parasitic Hymenoptera (Kriegler & Whitehead, 1962; Myburgh *et al.*, 1973 and Urban & Mynhardt, 1983). These ants, while feeding on honeydew excreted by the mealybug, deter natural enemies from controlling mealybug populations. The ants that are most abundant in vineyards in South Africa are *Anoplolepis custodiens*, *A. steingroeveri, Linepithema humile* (all epigaeic or ground-nesting species) and *Crematogaster peringueyi* (Emery), an arboreal or vine-nesting species (Whitehead, 1957; Urban & Bradley, 1982; Addison & Samways, 2000). With the introduction of the Scheme for Integrated Production of Wine (promulgated under the Act on Liquor Products [Act 60 of 1998]), it was necessary to find a cost-effective method for ant control that is environmentally friendly, practicable and acceptable in an integrated pest management programme. This study was conducted to assess various chemical stem treatments against three epigaeic ant species, the Argentine ant *L. humile*, the black pugnacious ant *A. steingroeveri* and the common pugnacious ant *A. custodiens*. Chemical stem barriers have been found to be effective against various ant pests, including *L. humile* and *A. custodiens*, in citrus orchards and are considered to be a suitable method of ant control as ants are left to forage on the orchard floor, where they fulfil important ecological functions such as feeding on other pest insects (Samways & Tate, 1984; Moreno *et al.*, 1987; Stevens *et al.*, 1995; James *et al.*, 1998).

MATERIALS AND METHODS

Field trials

Treatments

Concentrations and application methods are listed in Table 1. Two synthetic pyrethroids and one organophosphate were tested as direct stem sprays. These were applied above the irrigation pipes (approximately 40 cm above the ground) as a 10 cm wide band using a ring spray attached to a knapsack spray pump (Fig. 1). A dosage of 50 mL/vine of the spray mixture was applied to each stem. Slow-release chlorpyrifos-impregnated bands were fastened around stems by stapling them onto vines above the irrigation pipes. A slow-release terbufos band, also an organophosphate, consisted of a cotton material stocking filled with granulated chemical. The fruit on vines exposed to the latter treatment were analysed for organophosphorous residues by the Department of Health (Forensic Chemistry Laboratory, Cape Town) shortly before harvest, due to the volatility [vapour pressure: 34.6 mPa (25°C)] of this chemical (Tomlin 1997).

One insect and three plant extracts, intended for use by resource-limited producers, were also tested. Using methods described by Elwell & Maas (1995), the leaves and fruit of the syringa tree (*Melia azedarach* Linnaeus), the leaves and stems of the tomato plant (*Lycopersicon esculentum* Linnaeus), five moderate-sized garlic cloves (*Allium sativum* Linnaeus) and ants, the same species as the pest ant species, were crushed and soaked in 5 mL liquid soap (Sunlight Liquid) and 1 L water for 12 h. The ants were col-

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lected during December during both years and consisted mainly of workers. The solutions were strained through filter paper (milk fileters, 191 mm in diameter) and used as direct stem sprays. The terbufos slow-release band, crushed garlic, syringa and tomato extracts were each tested during one season only.

Sites

Two field trials, one to assess the control of *A. custodiens* in Robertson (33°50'S 19°56'E) and one to assess the control of *L. humile* in Simondium (33°10'S 18°55'E), were carried out during the 1997/98 and 1998/99 seasons. The same vineyards were not re-used for trials during the following season. All trials were carried out in established, trellised, wine-grape vineyards. The vineyards were micro-irrigated and weeds were trimmed where necessary to prevent ants from gaining alternative access to vines. All vineyards were naturally infested with mealybug.

Trial layout

Robertson – Anoplolepis custodiens

Five replicates of eleven treatments and one untreated control (Table 2) were randomised in a complete block design. Each replicate consisted of the five to six vines occurring between two adjacent trellis poles. Pre-treatment ant counts were made on 4 December 1997 and on 18 November 1998. Treatments were applied on 9 December 1997 and on 3 December 1998, respec-

TABLE 1

Treatments and application method used in Robertson against *Anoplolepis custodiens* and in Simondium against *Linepithema humile* during 1997 and 1998.

Active ingredient and concentration	Concentration in mL/L (unless otherwise stated)	Application method
(1)Alphacypermethrin SC (100 g/L)	10	Stem spray
Alphacypermethrin SC	20	Stem spray
Alphacypermethrin EC (100 g/L)	10	Stem spray
Alphacypermethrin EC	20	Stem spray
(2)Betacyfluthrin EC (50 g/L)	20	Stem spray
Betacyfluthrin EC	30	Stem spray
(3)Chlorpyrifos EC (480 g/L)	41	Stem spray
Crushed ant extract	5 g/L	Stem spray
Crushed garlic extract	15 g/L	Stem spray
Syringa plant extracts	225 g/L	Stem spray
Tomato plant extracts	140 g/L	Stem spray
*Chlorpyrifos-impregnated band	–	Stem band
**Terbufos slow-release band	–	Stem band

(1)Fastac (Cyanamid), (2)Bulldock (Bayer), (3)Dursban (Efekto), * Suscon Blue, ** Donor (Quest Developments).

FIGURE 1

Ring spray attachment with four nozzles fastened onto a knapsack spray pump (not shown here), which was used to apply chemical stem treatments around vine stems.

tively. Trials were evaluated until after harvest when ant activity decreased, 115 and 139 days after application of treatments in 1998 and 1999, respectively.

Simondium – Linepithema humile

Five (1997) and four (1998) replicates of eleven treatments and one untreated control (Table 2) were randomised in a complete block design. Pre-treatment counts were made on 27 November 1997 and on 25 November 1998. Treatments were applied on 11 December 1997 and on 17 December 1998, respectively. Trials were evaluated until after harvest, 135 and 117 days after application of treatments in 1998 and 1999, respectively.

Method of evaluation

Vines were classified as infested if one or more ants were seen moving up or down the entire length of vine stems during a 5 to 10 sec stem observation. All the treated vines per replicate were used to collect data. Stem observations were done weekly during 1997/98 and every second week during 1998/99 until ant activity decreased. The number of days after which 25% or more of the vines per plot were infested with ants was used to measure the efficacy of treatments. This is the guideline that is currently used by producers as an action threshold to determine if ant control is necessary. A *two-way* ANOVA, LSD was performed on the data. Data were analysed separately for each season and for each trial.

Simulated field trials

Due to the uneven distribution of ants in vineyards resulting in some plots having low or zero ant counts in pre-treatment evaluations, a simulated field trial was conducted in order to achieve even and high pre-treatment ant counts for testing chemical stem barriers. Five treatments were evaluated, alphacypermethrin SC at 20 mL/L, betacyfluthrin EC at 30 mL/L, chlorpyrifos EC at 41 mL/L, the chlorpyrifos-impregnated band and an untreated control. Two vineyards in the Stellenbosch area, one infested with *L. humile* and one infested with *A. steingroeveri* were used. Twenty-five cm lengths of old vine stems were attached to dowel sticks at one end and feeding trays at the other end (Fig. 2). The dowel sticks were used to secure stems vertically in the ground. Stems were distributed in ten groups of five each (ten replicates, five treatments), throughout each vineyard directly in front of planted vines where ant activity was noticed. The five stems in each group were placed 30 – 40 cm apart. The feeding trays were filled with syrup daily until all trays were infested with ants, resulting in a 100% pre-treatment infestation. The stems were then dipped into the chemicals and the bands were fastened onto the stems, which were replaced in the same locations. Feeding continued throughout the trial. Stems were treated in February 1999 against *L. humile* and in March 1999 against *A. steingroeveri.* Feeding trays were inspected at weekly or fortnightly intervals and classified as infested or uninfested with one or more ants during 5 sec observations. A 9 x 1 Chi-squared test was used to test for treatment differences in the trial assessed for *L. humile* and a 6 x 1 Chi-squared test was used for the same purpose in the trial assessed for *A. steingroeveri* (Snedecor & Cochran, 1967).

RESULTS

Field trials

Ant activity in the pre-treatment counts was variable between treatments and between seasons (Table 2). Treatments were regarded as effective if they succeeded in keeping 75% or more vines free of ants for 90 days, the approximate time between the start of ant activity in vines and harvest.

TABLE 2

Pre-treatment ant counts of *Anoplolepis custodiens* in Robertson and of *Linepithema humile* in Simondium during 1997 and 1998.

	Pre-treatment counts (% infested vines)*			
Active ingredient and Concentration	***Anoplolepis custodiens***		***Linepithema humile***	
	1997	**1998**	**1997**	**1998**
Control	8 ab	70 c	14 a	75 ab
Alphacypermethrin SC (10 mL/L)	15 ab	88 abc	8 a	84 a
Alphacypermethrin SC (20 mL/L)	10 ab	100 a	16 a	67 ab
Alphacypermethrin EC (10 mL/L)	28 a	81 abc	12 a	73 ab
Alphacypermethrin EC (20 mL/L)	15 ab	71 c	12 a	60 b
Betacyfluthrin EC (20 mL/L)	8 ab	88 abc	12 a	62 ab
Betacyfluthrin EC (30 mL/L)	10 ab	80 bc	13 a	72 ab
Chlorpyrifos EC (41 mL/L)	17 ab	74 bc	17 a	71 ab
Crushed ant extract	7 b	75 bc	12 a	57 b
Crushed garlic extract	–	92 ab	–	71 ab
Syringa plant extracts	20 ab	–	12 a	–
Tomato plant extracts	22 ab	–	8 a	–
Chlorpyrifos-impregnated band	5 b	87 abc	10 a	77 ab
Terbufos slow-release band	–	88 abc	–	66 ab

*Numbers followed by the same letter in a column do not differ significantly ($p \leq 0.05$).

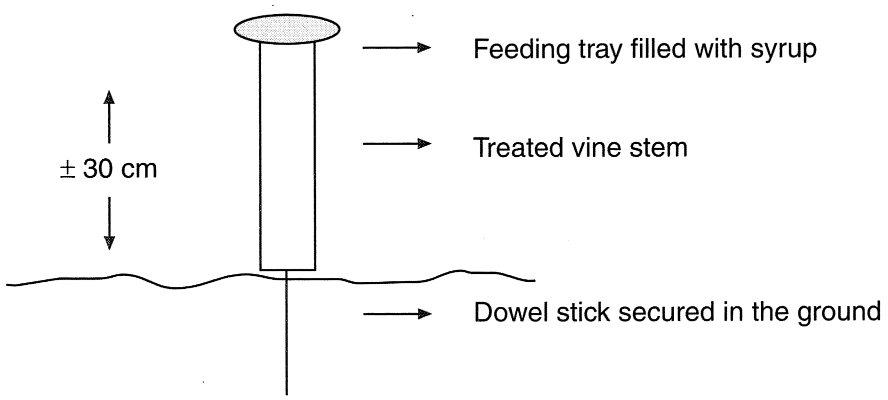

FIGURE 2

Schematic respresentation of an old vine stem used in the simulated field trial to test chemical stem barriers against ants.

Robertson

Anoplolepis custodiens was not present in high numbers in vines treated with chlorpyrifos EC, alphacypermethrin EC and SC at 20 mL/L, both concentrations of betacyfluthrin EC and the chlorpyrifos-impregnated band during the first season (Table 2). These treatments did not differ significantly from the control as a result of low ant infestations and, therefore, their effectiveness cannot be evaluated using these results (Fig. 3A). During the second season only the chlorpyrifos-impregnated band and the terbufos slow-release band resulted in acceptable control. No organophosphate residues were found on grapes from the terbufos-banded vines. These bands could have been effective for a longer period, but as evaluations ceased 139 days after application of treatments, this could not be determined.

Simondium

During the first season *L. humile* was excluded from vines by chlorpyrifos EC, alphacypermethrin EC and SC at both concentrations, betacyfluthrin at 20 mL/L, the chlorpyrifos-impregnated band and the crushed ant extract (Fig. 3B). Alphacypermethrin EC at 20 mL/L was still effective at the last sampling date during the first season. Alphacypermethrin SC at 10 mL/L, betacyfluthrin EC at 20 mL/L, the chlorpyrifos-impregnated band and the terbufos slow-release band were effective during the second season, indicating that these treatments are effective against high ant infestations. Although the higher concentrations of alphacypermethrin EC and SC were less effective than the lower concentrations during the second season, these differences were not significant. However, the higher concentrations of betacyfluthrin EC were significantly less effective than the lower concentrations during both seasons. The chlorpyrifos-impregnated band was still effective at the last sampling date during 1998/99.

Simulated field trials

Linepithema humile

All treatments differed significantly from the control and from each other (Table 3A). Figure 4A shows the percentage infestation of *L. humile* for each observation.

Anoplolepis steingroeveri

All treatments differed significantly from the control at the last sampling date and all treatments, except chlorpyrifos EC and the chlorpyrifos-impregnated band, differed significantly from each other (Table 3B). Figure 4B shows the percentage infestation of *A. steingroeveri* for each observation.

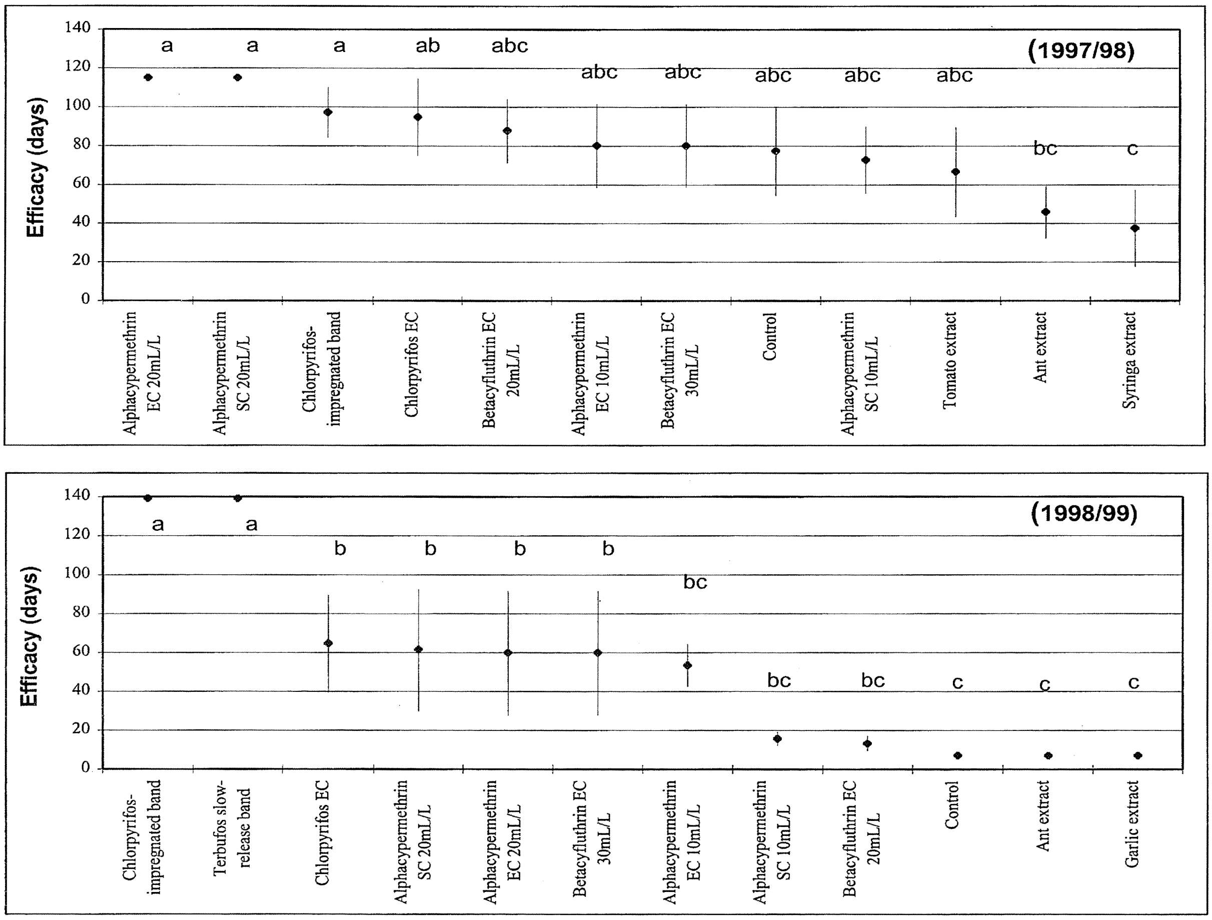

FIGURE 3A

Efficacy (number of days after application of treatments until ants infest 25% or more vines) assessed during two years for *Anoplolepis custodiens* in Robertson. Error lines represent plus/minus standard errors. Treatments not designated by the same letter are significantly different ($p \leq 0.05$).

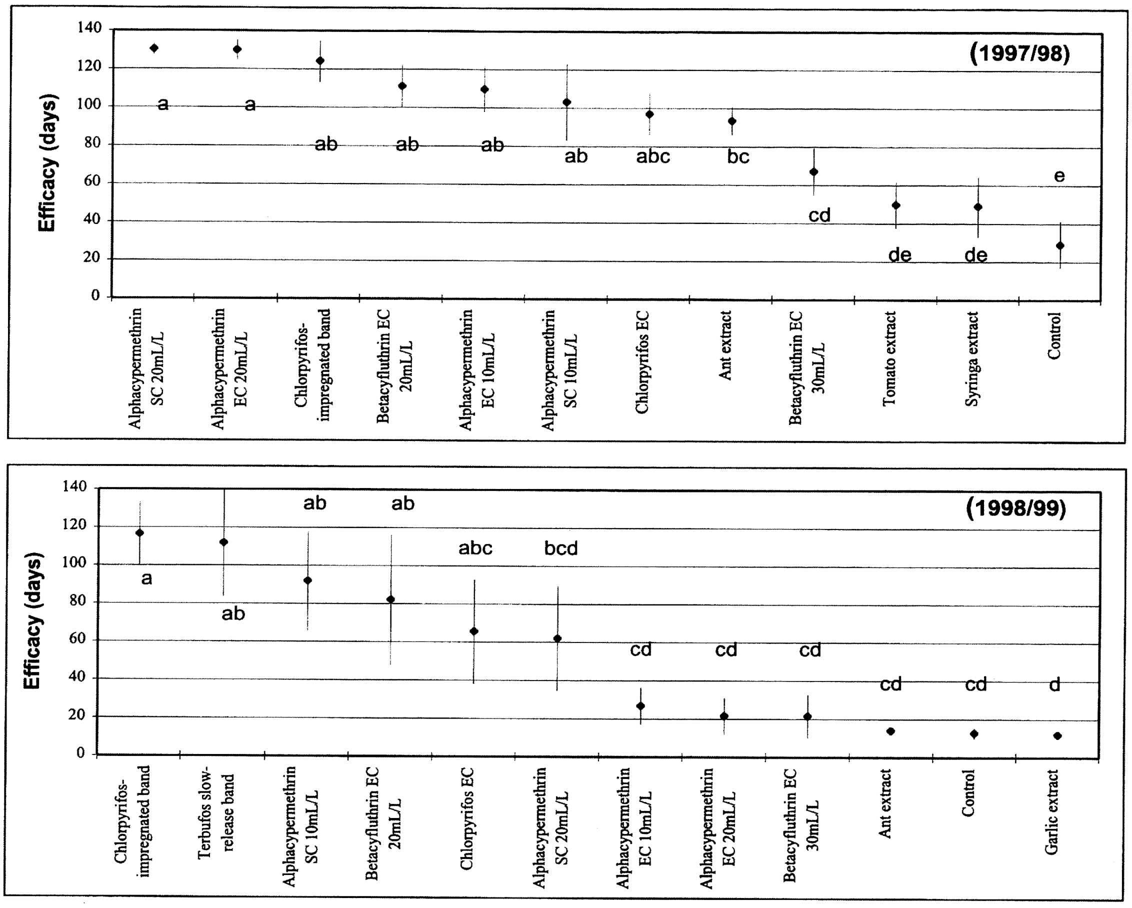

FIGURE 3B

Efficacy (number of days after application of treatments until ants infest 25% or more vines) assessed during two years for *Linepithema humile* in Simondium. Error lines represent plus/minus standard errors. Treatments not designated by the same letter are significantly different ($p \leq 0.05$).

TABLE 3A

Average percentage infestation during ten sampling dates with Chi-square values (df = 9) of treatments tested against *Linepithema humile* in a simulated field trial in Stellenbosch during 1999.

Treatments		**Control**	**Chlorpyrifos EC**	**Alphacypermethrin SC**	**Betacyfluthrin EC**
	% Infestation	**81**	**20**	**30**	**40**
Chlorpyrifos EC	20	94.6 *	–	–	–
Alphacypermethrin SC	30	57.6 *	54.2 *	–	–
Betacyfluthrin EC	40	65.8 *	124.7 *	54.6 *	–
Chlorpyrifos band	6	92.0 *	34.9 *	60.0 *	56.4 *

* Indicates significant differences between treatments ($p \leq 0.05$).

TABLE 3B

Average percentage infestation during seven sampling dates with Chi-square values (df = 6) of treatments tested against *Anoplolepis steingroeveri* in a simulated field trial in Stellenbosch during 1999.

Treatments		**Control**	**Chlorpyrifos EC**	**Alphacypermethrin SC**	**Betacyfluthrin EC**
	% Infestation	**86**	**77**	**42**	**50**
Chlorpyrifos EC	77	42.6 *	–	–	–
Alphacypermethrin SC	42	115.3 *	58.9 *	–	–
Betacyfluthrin EC	50	134.2 *	69.1 *	21.23 *	–
Chlorpyrifos band	79	52.6 *	11.7	80.4 *	96.0 *

* Indicates significant differences between treatments ($p \leq 0.05$).

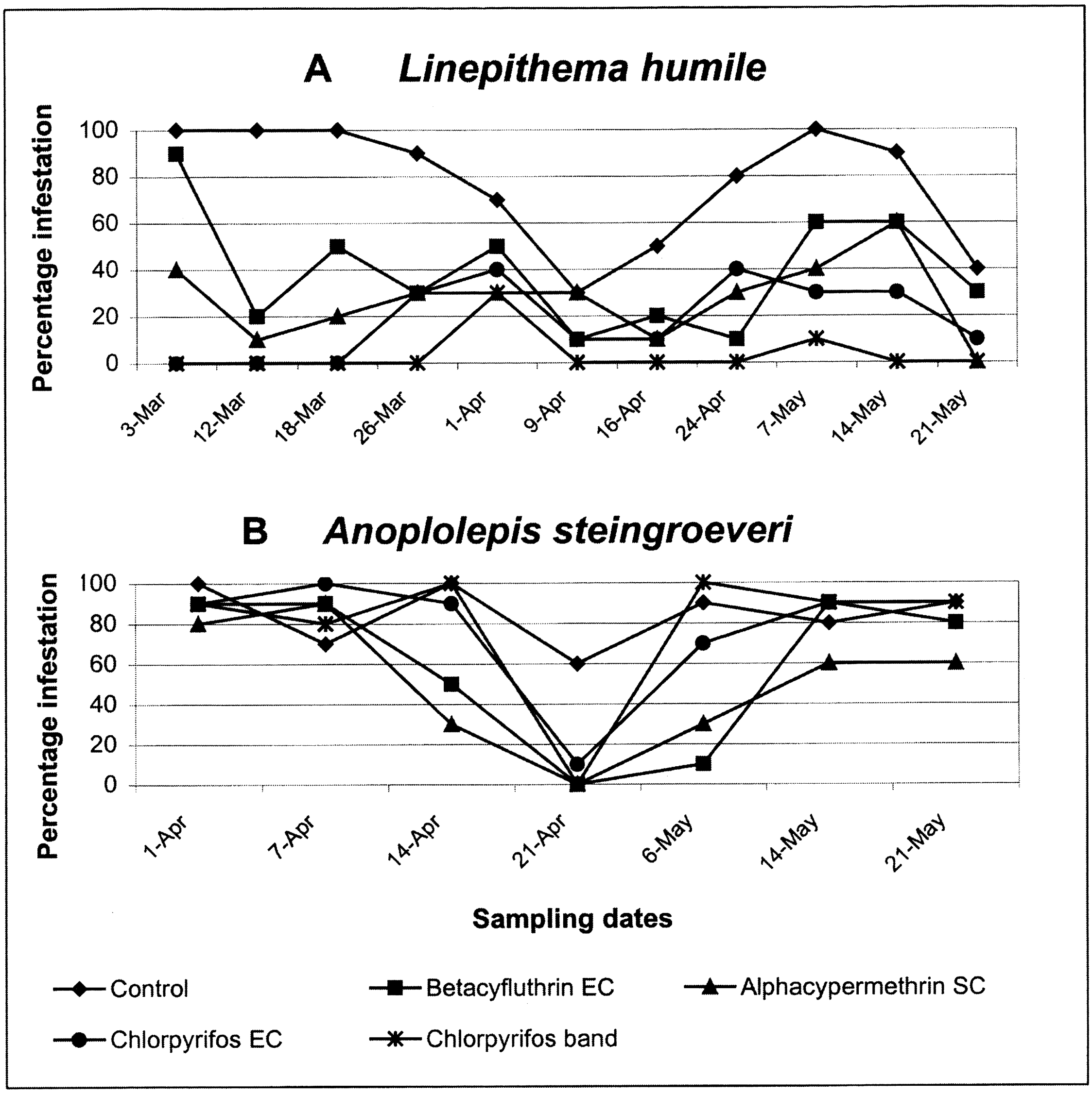

FIGURE 4

Percentage infestation by *Linepithema humile*and *Anoplolepis steingroeveri* during eleven and seven weeks, respectively, in a simulated field trial where five treatments were tested in Stellenbosch during 1999.

DISCUSSION

From the field trials and simulated field trials it can be seen that *L. humile* is easier to control than the *Anoplolepis* spp. One possible reason for this could be the larger size and longer legs of the *Anoplolepis* spp. compared to the smaller *L. humile* (Arnold 1915), resulting in less contact with the treated area when walking up and down the vine stem.

Field trials

The variability in ant infestations during the pre-treatment counts can possibly be ascribed to climatic conditions favouring mealybugs and/or ants during the second season. More treatments were effective in excluding *L. humile* than *A. custodiens*. In vineyards with high ant activity the chlorpyrifos-impregnated band, the terbufos slow-release band and alphacypermethrin SC at 10 mL/L were consistently effective in excluding ants for three months or longer. The latter was only effective against *L. humile*. Some unusual trends were observed during the second season of testing against *L. humile*, where betacyfluthrin EC at 20 mL/L was consistently less effective than the higher concentration of 30 mL/L. Results also indicated that alphacypermethrin SC at 10 mL/L was more effective than the lower concentration. Although this difference was not significant, the higher concentration does not meet with the requirement of a minimum of 90 days ant exclusion. No explanation can be given for this result.

Simulated field trials

None of the treatments met with the requirements of a 25% infestation or less for *A. steingroeveri*. The percentage infestation for all treatments often reached over 80% for *A. steingroeveri*, but always remained at 60% or below for *L. humile* for all treatments (Fig. 4). Alphacypermethrin SC at 20 mL/L gave the best control out of the four treatments for this ant and was significantly better than the chlorpyrifos-impregnated band, which significantly outperformed all treatments in the second season (high ant infestation) of field trials. This difference in results can possibly be explained by observations where an established trail of ants was never seen moving over bands in the field trial, only an occasional single ant. In the simulated field trial such established trails were more common, possibly due to the easily accessible food source. The same trend was not observed for *L. humile,* where the chlorpyrifos-impregnated band always gave acceptable control in field trials and simulated field trials, even with high ant infestations. A decrease in ant infestation was noticed five to six weeks after application of treatments for all five treatments, including the control, in the simulated field trials for both ant species. A possible reason for this could be that treatments were killing ants and that the effect was only being noticed at this time. In Australian field trials testing controlled-release chlorpyrifos stem bands against *Iridomyrmex rufoniger* gp. spp. in citrus trees, a decline in ant activity was also observed and ascribed to the bands killing ants (James *et al.*, 1998). From laboratory bioassays conducted in the same study, it was shown that these bands can result in ant mortality after exposure for 16 h, and it is therefore possible that other stem treatments also cause ant mortality. Since the reduction in numbers also occurred in the untreated control, it is assumed that ants from one colony may have fed on vines with different treatments. The subsequent increase in numbers can be explained by a new invasion of ants from other nests or a recovery of the ant populations. From visual observations it was noticed that the nest density of *A. steingroeveri* was much higher than that of *L. humile,* which could explain why the population reduction was shorter-lived for *A. steingroeveri* than for *L. humile*. Another possible reason for the decline in numbers could be that feeding trays were lacking syrup for an extended period between sampling dates. This occasionally occurred as a result of bees feeding on the syrup. Bees were particularly abundant in the vineyard where *A. steingroeveri* was being monitored. However, this would not explain the gradual decrease and subsequent gradual increase in numbers over three to five sampling dates. The first reduction in numbers was probably not due to unfavourable weather conditions as temperatures were more or less constant on each sampling date and average rainfall was low for that month (5 mm).

The simulated field trial has several advantages over a conventional field trial. A pre-treatment infestation of 100% can be obtained for all treatments. Assessments are less time consuming and more accurate. High ant activity can be maintained throughout the evaluation period by the continuous availability of an energy-rich, easily obtainable food source, which allows for very thorough testing of treatments. A simulated field trial can give quick results under field conditions and does not require the establishment and maintenance of laboratory colonies. Finally, standardisation is easy and it is possible to test treatments that have been variably weathered, thereby allowing their efficacy over time to be assessed simultaneously. Weathering of treatments will take place at the same rate as in the field as the substrate (vine stems) used is the same. However, it is recommended that the same treatments be grouped together instead of replicates in order to prevent cross-infestation of ants between treatments from taking place. This is important if treatments result in ant mortality. The feeding trays should be screened to prevent bees from feeding, but still allow the ants access to the food source.

CONCLUSIONS

More treatments effectively control *L. humile* than *A. custodiens* or *A. steingroeveri* for the required period of approximately three months. Subsequent to the start of these trials alphacypermethrin SC at 10 mL has been registered as a direct stem treatment in vineyards against *L. humile*, while alphacypermethrin SC at 20 mL has been registered against the *Anoplolepis* spp.. The two treatments that provided the best control against all three ant species, the chlorpyrifos-impregnated band and the terbufos slow-release band, may be impractical and expensive for use in commercial vineyards due to high planting density (±2000 vines/ha). However, if they are effective for more than one season, their use on farms may be reconsidered and their continued efficacy for *L. humile* and *Anoplolepis* spp. control in vineyards needs to be determined. Alternatively, more practical methods should be investigated, such as the use of toxic baits with low mammalian toxicity which are dispensed in ant-specific feeding containers, for example, boric acid in a sugar solution (Klotz *et al.*, 1998). Although toxic baits can control *L. humile* effectively, it is more difficult to find a suitable bait that controls the *Anoplolepis* spp. Future research is also needed to establish suitable and practical bioassay techniques which expose ants to the treated substrate for a limited period thereby simulating field conditions, and more chemical stem treatments need to be assessed for ant mortality after exposure.

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