The integrated management of Kelly's citrus thrips

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Improving the Management of Kelly's Citrus Thrips (KCT)

Horticulture Australia Limited

Final Report CT00015 (June 2004)

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Project CT00015

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This report details the research and extension delivery undertaken in Project CT00015 on the integrated management of Kelly's citrus thrips (KCT), *Pezothrips kellyanus* (Bagnall), in inland Australian citrus crops. Main findings, industry outcomes and recommendations to industry along with suggested areas of future research are discussed.

November 2004

Coverphotos: Adult KCT and associated feeding scars on lemon flowers (L); the soil-dwelling predatory mite, *Protogamasellopsis* spp. (Acarina: Rhodacaridae), a predator of KCT pupae (R).

ACKNOWLEDGMENTS

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MEDIA SUMMARY

Kelly's citrus thrips (KCT) is the key pest of citrus in the Riverland-Sunraysia (R-S) region. The scurfing and rind bleaching that results from KCT feeding reduces fruit quality, thereby reducing the packout of export quality fruit and rendering some fruit unsaleable.

The management of KCT is presently restricted to the use of foliar insecticides. Building on the foundations laid in project CT97007, this new project aimed to expand the insecticidal and biological control options available for KCT control, and to start to integrate and optimise these control options.

The key outcomes were:

- Substantial levels of organophosphate resistance were shown to occur in KCT populations in R-S citrus.
- Baseline susceptibility levels and 'discriminating doses', that allow quick diagnosis of any shifts in resistance, were calculated for three existing and four new candidate insecticides for KCT control.
- Two new foliar sprays (Actara™ and Success™) were effective for KCT control, but each has 'off-target' impacts on a key beneficial insect of citrus.
- A complex of soil-dwelling mites is predacious on KCT in R-S orchards, and factors that appear to influence the abundance of these predators have been identified.
    - Together, these findings provides for the first time the basis for an effective biological alternative to the insecticidal control of KCT.
- The fungal insect pathogen, *Metarhizium*, is an ineffective tactic for KCT control.
- Ground application of the insecticide bifenthrin can reduce KCT emergence from treated soil. This may be an effective alternative treatment if targeted at lemons (an important KCT breeding source).
- The phenomenon whereby exposure of an insect to sublethal pesticide doses increases its egg production is unlikely to have contributed to the 1990's rise of KCT as a serious new pest.
- There is no large-scale regional movement of KCT in the R-S.
- KCT in this region is largely a 'self-contained' population cycling within citrus.
- Growers' understanding and practice of correct thrips identification and effective monitoring and spray control, and their awareness of the resistance threat and the biocontrol research developments were all improved.

Future R&D is required to develop an effective insecticide resistance management (IRM) strategy, to field trial several new insecticidal controls, and to enhance KCT biocontrol in citrus orchards.

We recommend that citrus growers should ensure correct thrips identification, frequent monitoring from petal fall to Christmas, accurate spray timing and good spray coverage to get good control of KCT. If poor spray efficacy occurs, and coverage is deemed to have been good, a sample of KCT should be tested for insecticide resistance. We recommend that the citrus industry should encourage Syngenta and Dow Agrosciences to swiftly advance the registration applications of new chemistry for KCT control, devise and implement an IRM 'rotation' strategy, and support endeavours to further enhance KCT biocontrol.

TECHNICAL SUMMARY

The Problem

Kelly's citrus thrips (KCT), *Pezothrips kellyanus* (Bagnall), is a serious pest of navel and Valencia oranges, grapefruit and lemons in the Riverland-Sunraysia region, and to a lesser extent in Western Australia and the Riverina. The feeding of KCT on young and mature fruit causes scurfing (or halo) marking and rind bleaching, respectively. These blemishes reduce fruit quality, thereby reducing the packout of export quality fruit and rendering some fruit unsaleable. A 2003 survey reveals that Riverland navel orange producers alone lose around $9+ million per annum from KCT.

Given the lack of biological information about this new pest, KCT management has been limited to the use of several insecticides, which often provide poor results. This project has built on findings of CT97007 to improve KCT insecticidal control options for citrus growers, and, in light of the CT97007 finding that KCT pupate in the soil, has set about to determine whether a sustainable IPM system based upon the biological control of KCT in citrus orchard soils is achievable.

The Project Science

The research focused in a number of key areas:

- Studies to determine the significance of resistance in Riverland populations of KCT to insecticides commonly used for their control.
- Efficacy trials to assess several new candidate insecticides for KCT control, and to generate data to assist in the registration of the effective compounds.
- Assessment of the impact of the commonly used and promising new KCT insecticides on citrus beneficials.
- Biological control studies to determine the potential impact of soil predators on KCT abundance in citrus orchards, and the orchard management factors that favour or harm these beneficials.
- Assessment of a strain of *Metarhizium* fungus as a mycoinsecticide for KCT control.
- Studies of KCT population movement (important for improving pest and resistance management strategies), and of the role of colour in attracting KCT to flowers (basic knowledge needed for the design of a mass trapping system).

The Key Research Findings, Extension Highlights and Industry Outcomes

- A survey has revealed a 30% increase in KCT sprays per Riverland citrus orchard over the past five years, and that an increasing number of growers are now substituting methidathion for chlorpyrifos when controlling KCT.
    - These changes reflect the increasing difficulty experienced by more and more growers to adequately control KCT.
- Insecticide bioassays have confirmed that some populations of KCT in Riverland-Sunraysia citrus have substantial levels of chlorpyrifos (and to a lesser extent methidathion) resistance. The higher levels of chlorpyrifos resistance that were recorded would almost certainly be causing field control failures.
- Baseline susceptibility levels and 'discriminating doses' for chlorpyrifos, methidathion, methomyl and several new candidate insecticides for KCT control have been established using a susceptible strain of KCT collected from Adelaide.
    - This allows quick diagnosis of any shifts in susceptibility, and judgement whether field control failures are caused by resistance.
- In a laboratory study exposure of adult female KCT to sublethal doses of chlorpyrifos had no effect on their fecundity.
    - Based on this finding it seems unlikely that hormoligosis (the phenomenon whereby exposure of an insect to sublethal doses of a pesticide causes an

increase in its fecundity) is a cause of the 1990's emergence of KCT as a serious new pest.

- Insecticide trials have demonstrated the efficacy of foliar spraying of Actara™ and Success™ (and the benefit of mixing Success™ with the oil products such as Brella™) for KCT control. The results have:
    - stimulated the manufacturers of these two insecticides to develop them for KCT control in Australian citrus, and
    - been provided to the manufacturers to assist with APVMA registration.
- Studies with the main red scale parasitoid *Aphytis melinus* have revealed a similar degree of residual contact toxicity from exposure to weathered residues of chlorpyrifos (Lorsban™) and spinosad (Success™), but a significantly greater persistence of the toxic impact of thiamethoxam (Actara™).
    - That is, frequent use of either of these new candidate insecticides for KCT control would be expected to disrupt the citrus IPM system.
- A low rate of bifenthrin applied to the ground in the drip-line area significantly reduced the emergence of adult KCT in a Riverland field trail.
    - The strategic application of bifenthrin to the soil of lemon orchards (important KCT breeding source) may be a cost effective, less disruptive (at the district-wide level) option for KCT control.
- The search for biocontrol agents has identified soil-dwelling mite populations that are predacious on KCT in Riverland-Sunraysia citrus orchards.
    - Negative correlations between the abundance of these soil predators and the survival of soil-dwelling KCT suggest an important causal link between predatory mites and low thrips numbers.
    - Several factors have been identified that appear to influence the abundance of these soil predators (soil organic carbon and run-off from chlorpyrifos sprays, and possibly the prevalence of grasses amongst the ground-cover).
    - This provides for the first time the basis for an effective biological alternative to the insecticidal control of KCT.
- The fungal insect pathogen, *Metarhizium anisopliae* var. *anisopliae* was investigated as an alternative KCT control strategy, but proved to be ineffective in field tests.
- The results of a suction trapping study in the Riverland suggest that there is no large-scale regional movement of KCT in this region during the spring. Instead, KCT are moving between citrus orchards at the local (settlement) level. It is likely that the same applies in the Sunraysia region.
    - Based on this evidence, and the CT97007 observation that there are few non-citrus hosts of KCT in the Riverland-Sunraysia region, it seems that KCT in this region are largely a 'self-contained' population cycling within citrus.
    - These findings have important implications for IRM (insecticide resistance management) and the landscape management of KCT.
        - They suggest that the only source of susceptible KCT available to dilute resistance will be citrus orchards that are left unsprayed.
- A study of the role of colour in attracting adult KCT to host flowers suggests that odour is a more significant cue than colour.
    - If the chemical components of these attractant odours could be identified, their use in a trap may significantly improve its effectiveness.
- Significant resources were directed into the delivery of field day displays, Cittgroup presentations and print media information throughout the project.
    - Growers' understanding and practice of correct thrips identification, crop monitoring and spray control practices were improved. Grower and industry awareness of the threat of insecticide resistance and the "chemical treadmill", and the importance of embracing an IPM system centred on the effective boosting of KCT natural enemies, have been significantly raised.

Recommendations

We recommend that the citrus industry:

1. Encourage Syngenta and Dow Agrosciences to swiftly advance the registration applications for the use of thiamethoxam and spinosad respectively for control of KCT in citrus.
2. Implement an insecticide resistance management (IRM) strategy as soon as new chemistry is registered for KCT control.
3. Given that the singular reliance on insecticidal control of KCT will be unsustainable even if an IRM strategy is adopted, the citrus industry should support efforts to enhance the contribution of biocontrol agents to KCT control.
4. Appraise the potential benefits to KCT biocontrol, and to citrus IPM generally, of multi-fan (lower-spray volume) spray technology.

We recommend that further research be undertaken in four key areas:

1. Insecticide resistance studies.
2. Insecticide field trials.
3. Enhancement of KCT biocontrol.
4. KCT management by irrigation modification.

INTRODUCTION

Historical background to project CT00015

Kelly's citrus thrips (KCT), *Pezothrips kellyanus* (Bagnall) (Thysanptera: Thripidae) emerged in the early 1990's as a serious citrus pest in the Riverland and Sunraysia regions, and more recently has been reported causing economic damage in the Riverina and Western Australia. The feeding of KCT on young and mature fruit respectively causes scurfing (or halo) marking and rind bleaching. Both forms of blemish downgrade fruit quality, thereby reducing the packout of export quality fruit and rendering some fruit unsaleable.

Lemons, navel and Valencia oranges and grapefruit are the most affected varieties. In unfavourable years, despite the application of 1-3 thrips sprays, an average of 20-40% of the fruit of these varieties can be rendered unsaleable for quality fresh markets. A recent survey reveals that Riverland navel orange producers alone lose around $9+ million per annum as a result of KCT damage and control costs.

Because KCT has no prior history as a pest, neither in Australia nor overseas, there was no published information available on its biology or management when project CT97007 commenced in 1997. As a result the only strategy available to the industry to manage KCT has been insecticidal control, of which foliar spraying of chlorpyrifos has been the main practice. Such treatments invariably disrupt natural enemies, and select for insecticide resistance in KCT. Less commonly, exposure to sublethal insecticide doses can actually increase the fecundity of the pest (as has been documented for the Californian citrus thrips), and help cause the pest problem in the first place.

Project CT97007 determined the optimal timing of insecticidal sprays for KCT control, and provided evidence that some KCT populations in Riverland and Sunraysia citrus may have reduced susceptibility to chlorpyrifos. It also elucidated important aspects of KCT biology, including the site of pupation (the soil), host plants, development rates, nutritional requirements, mating behaviour, and the scarcity of KCT natural enemies in most citrus canopies. Taken together, the findings of CT97007 heightened concerns about the insecticide resistance and "chemical treadmill" risks inherent in the current single-tactic approach to KCT management, but for the first time provided the biological knowledge base needed to develop a broader-based IPM system for the long-term management of KCT.

Concurrent with the progress made in CT97007 there was another significant development. Several new insecticides for thrips control became available in Australia in the late 1990's. Importantly from a resistance management standpoint, these compounds have different modes of action and metabolism from each other and from the long-established organophosphates (eg. chlorpyrifos, methidathion) and carbamates (eg, methomyl).

Aims

The aims of this project were to test the suitability of the several newly available insecticides for their efficacy against KCT and impact on citrus beneficials. Secondly, to identify KCT biocontrol agents, and to devise orchard management methods that enhance the contribution of these agents to KCT control. Thirdly, to identify other factors in the biology and/or behaviour of KCT, or in the environment that may be able to be exploited or modified to reduce KCT abundance and crop damage. Fourthly, to explore the use of an under-canopy *Metarhizium* (fungal) spray as an alternative control for KCT.

Significance for industry

Findings from these areas of investigation would be used to develop sound IPM and IRM tactics for the control of KCT.

ESTIMATION OF THE COST OF KCT TO THE AUSTRALIAN CITRUS INDUSTRY: A 2002-03 RIVERLAND SURVEY

INTRODUCTION

Kelly's citrus thrips has been a major pest of Riverland and Sunraysia citrus for the past 10 years (Baker *et al*. 2000), and has recently caused crop losses to Riverina and Western Australian citrus (Jianhua Mo and Sonya Broughton, *pers. comm*.). The feeding of KCT on young and mature fruit respectively causes scurfing (or halo) marking and rind bleaching. Both forms of blemish downgrade fruit quality, thereby reducing the packout of export quality fruit. Management of KCT requires the application of insecticides, often 2-3 times per season.

In 1998 a survey of three packing sheds was conducted to quantify the industry losses in pack-out due to KCT damage. A packing shed was selected from Renmark, Waikerie and Mildura areas. Juice Grade fruit from six to eight growers from each packing shed was inspected for KCT damage. This survey demonstrated that the incidence of KCT damage varies substantially between growers. However, it also demonstrated that KCT is a serious economic pest, with up to 58% of a grower's fruit downgraded to Juice as a result of KCT damage.

In June 2003, a second survey was conducted to estimate a monetary cost of KCT to Riverland growers. The results of this survey are reported below.

METHOD

The survey was conducted at two major packing sheds, Yandilla Park (Renmark) and Nippy's Waikerie Producers (Waikerie) in June 2003. For any particular grower, 100 fruit from both the 2nd Grade line and the Juice Line were assessed for KCT damage only. Fruit with KCT damage plus some other form of damage was disregarded in this survey as it can be difficult to determine whether the fruit was downgraded due to the KCT damage, the other form of damage or both combined.

At Yandilla Park packing shed fruit from 10 orchards were assessed for KCT damage only. At Nippy's Waikerie Producers packing shed fruit from 7 orchards were assessed for KCT damage only.

In addition to the cost of fruit rejection or downgrading in the packing shed, growers incur additional field control costs for KCT infestations. Therefore, each grower was asked to provide details on the number of insecticide sprays they applied specifically for KCT control.

RESULTS

There was, as expected, a large variation in KCT pest status between the orchards surveyed at these two Riverland packing sheds. On average 6.9% of the fruit sampled in this 2003 survey was downgraded from export to domestic grade, and a further 4.7% was downgraded from domestic to juicing grade, all due to KCT damage alone (Table 1). These losses occurred despite more than 40% of the orchards having been treated with 2-3 KCT insecticide applications, and despite the fact that KCT pressure in the Riverland in 2002-03 was only light to moderate.

Although this is a relatively small survey, and only representative of one year, it provides a valuable insight into the scale of economic losses that KCT causes.

Table 1. The percentage of 2nd Grade and Juice Line fruit with KCT damage only in samples from 17 Riverland orchards examined at Yandilla Park (Renmark) and Nippy's Waikerie Producers packing sheds, June 2003, and the corresponding KCT spray treatment details.

Grower	% of 2nd Grade fruit	% of Juice Line fruit	General KCT management details
Packingshed: Yandilla Park (Renmark) 11-12 June 2003			
YP1*	4	1	2-3 methidathion† or chlorpyrifos† applications, methomyl on mature fruit
YP2*	0	1	No KCT insecticides applied
YP3*	4	4	3-4 methidathion or chlorpyrifos applications, methomyl† on mature fruit
YP4*	21	4	As for YP3
YP5	15	4	4-5 chlorpyrifos + oil applications to cover all pests
YP6	2	0	No KCT insecticides applied
YP7	1	0	1 insecticide application
YP8	10	4	No KCT insecticides applied
YP9	4	2	Approx. 2 insecticide applications
YP10	2	1	No KCT insecticides applied
Packingshed: Nippy's Waikerie Producers, 17th June 2003.			
NWP 1	10	10	2-3 methidathion or chlorpyrifos applications
NWP 2	32	45	Temik™ and 1 chlorpyrifos application
NWP 3	0	1	No KCT insecticides applied
NWP 4	0	0	1 methomyl application
NWP 5	3	3	No KCT insecticides applied
NWP 6	6	0	No KCT insecticides applied
NWP 7	0	0	No KCT insecticides applied

* Leng Navels. All other fruit were Washington Navels.
† Methidathion is Supracide™ and similar products, chlorpyrifos is Lorsban™ and similar products, and methomyl is Lannate™ and similar products.

Estimated monetary cost of KCT in 2002-03
The approximate cost of a single insecticide application is $300/ha (Kym Thiel, *pers. comm*. 2004). As shown in Table 1, multiple applications of insecticide for the control of KCT are not unusual.

Based on 2003 harvest figures, the average returns for citrus are approximately:
Grade 1: $750/tonne
Grade 2: $150/tonne
Juice: $50/tonne

The mean cost per hectare of KCT, based on an industry production average of 35 tonne/ha of navel oranges (Kym Thiel, *pers. comm.*) and the mean crop loss and insecticide usage figures of the 17 surveyed properties, is $2600/ha in lost income due to fruit downgraded from Grade 1, plus $388/ha in insecticides costs, totalling $2988/ha. It should be noted that this estimate does not take account of the cost of KCT crop monitoring.

With approximately 3000 hectares of navel oranges in the Riverland, the economic cost of KCT for Riverland citrus growers in 2002-03, based on this $2988/ha cost estimate, is $8,964,000 per annum for the navel crop alone.

DISCUSSION

Based on the findings of this limited survey, KCT is estimated to have cost the Riverland navel orange industry in 2002-03 in excess of $8.9 million, despite this being a light to moderate pressure season for KCT attack. When the additional losses to grapefruit, lemon and Valencia orange crops and the Sunraysia region are taken into account, the true scale of the estimated direct cost of KCT to the Australian citrus industry would substantially exceed $10 million in 2002-03.

Aside from the monetary cost, there is also the biological cost to consider with KCT control. Repeated applications of insecticides not only have a detrimental impact on beneficial insects but will also hasten the development of KCT resistance to commonly used insecticides. As the threat of OP insecticide resistance in KCT populations spreads across the region, the cost of KCT control and damage will continue to rise, as growers struggle to control infestations and produce Grade 1 fruit. It is therefore essential that alternative insecticide groups are made available for KCT management, that they are incorporated into an insecticide resistance management rotation strategy, and that cultural and biological methods of KCT management be developed to assist the long-term effectiveness of all control options.

KCT INSECTICIDE RESISTANCE SURVEY OF RIVERLAND CITRUS GROWERS

INTRODUCTION

Bioassay research conducted during this project has shown that several KCT populations collected from commercial Riverland citrus orchards have evolved significant tolerance to chlorpyrifos and methidathion (respectively 252 and 11 times less susceptible than KCT populations sourced from several locations around Adelaide). This indicates a potential for organophosphate resistance in populations of KCT in commercial orchards to develop to levels where field control will be compromised.

Anecdotal evidence indicated that, over the past several years, significant numbers of Riverland citrus growers had started to increase their frequency of KCT spraying and/or increase their reliance on methidathion as the efficacy of chlorpyrifos for KCT control has diminished. This survey was undertaken to assess the extent to which Riverland growers have experienced greater difficulty in controlling KCT and responded by changing their insecticidal practices (insecticide type and frequency).

METHOD

Over 700 survey questionnaires (Appendix A) were mailed to South Australian citrus growers in August 2003, with reply paid envelopes. 130 surveys were completed and returned.

RESULTS & DISCUSSION

The results of the survey are summarized in Table 1 (at the end of this Section). Over the past five years, there has been an increase of approximately 30% in the average number of insecticide sprays applied per Riverland orchard for the control of KCT (Fig. 1). This is an alarming increase, which has occurred without being fuelled by any changes in market quality requirements, and without any apparent annual trend towards higher KCT abundance.

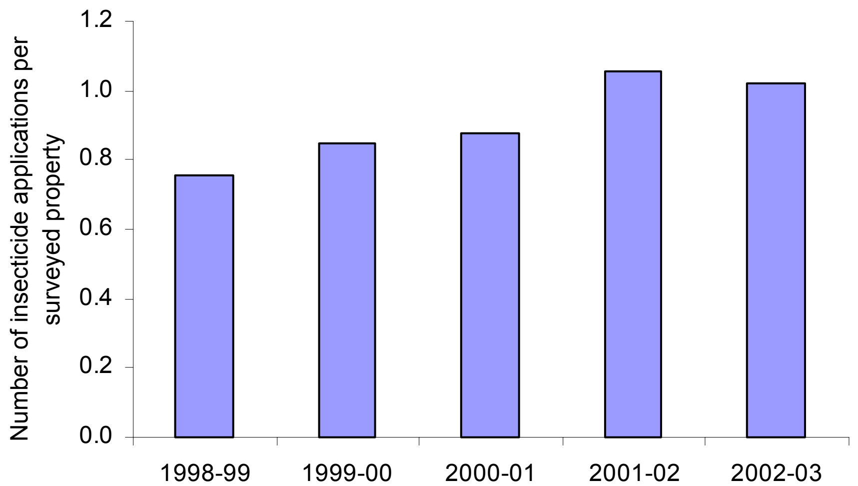

Fig. 1. The number of insecticides applied annually for the control of Kelly's citrus thrips per surveyed Riverland orchard, 1998-2003.

It seems that this increase in KCT spray frequency is due to the increasing difficulty experienced by an expanding number of growers to adequately control the pest with a single organophosphate (OP) spray application. The cause of this difficultly is the evolution of OP insecticide resistance, which the Project team have documented in a number of Riverland populations of KCT.

Another measure of this phenomenon is the recent substitution of methidathion for chlorpyrifos by an increasing number of growers when controlling KCT (Figs. 2 and 3). Chlorpyrifos usage, as a percentage of total KCT insecticide usage, was significantly lower in 2002-03 compared to the previous four years (Contingency table analysis, $\chi 2$=8.9, P=0.0029). Conversely, methidathion usage, as a percentage of total KCT insecticide usage, was significantly higher in 2002-03 compared to the previous four years (Contingency table analysis, $\chi 2$=26.4, P=0.0000). Further, the percentage of growers that use methidathion for KCT control has significantly increased over these five years from 5.4% to 28.5% (Contingency table analysis, $\chi 2$=23.0, P=0.0000).

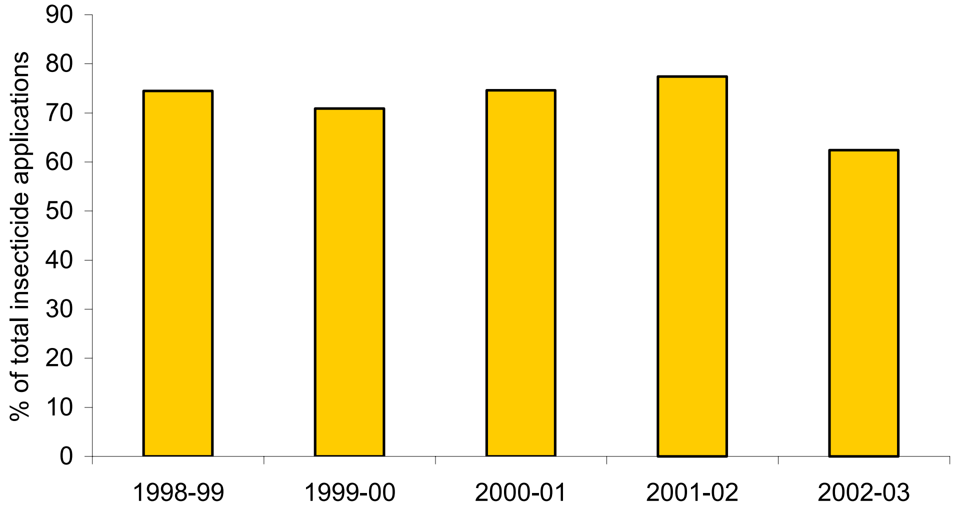

Fig. 2. Chorpyrifos usage expressed as a percentage of total insecticide applications for the control of Kelly's citrus thrips in Riverland orchards, 1998-2003.

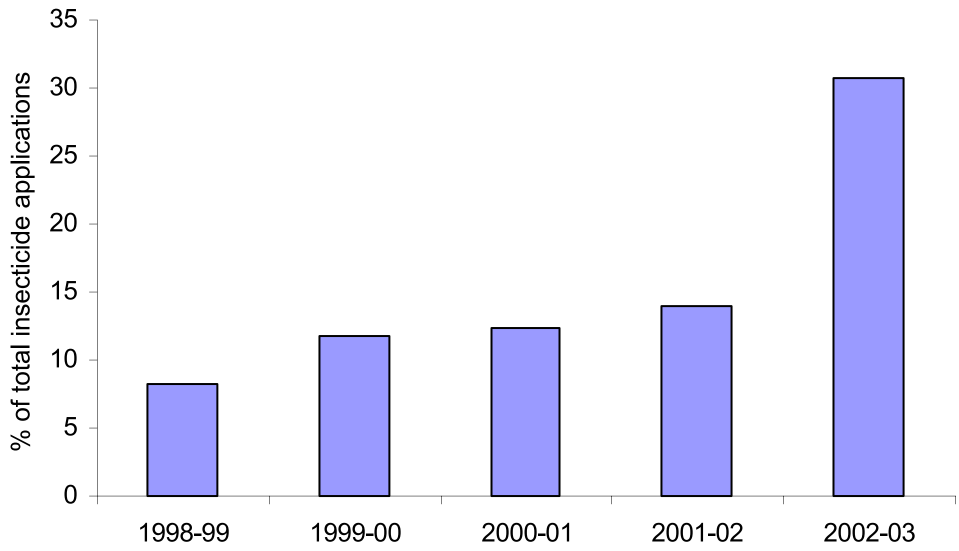

Fig. 3. Methidathion usage expressed as a percentage of total insecticide applications for the control of Kelly's citrus thrips in Riverland orchards, 1998-2003.

Methidathion is a more toxic insecticide to KCT than chlorpyrifos. With adults of a susceptible KCT strain the LC99 (the concentration that kills 99% of insects tested in a laboratory bioassay) for methidathion is approximately one third of the chlorpyrifos LC99 (Table 2). To be certain of achieving good field control many entomologists and chemical companies consider that the ratio of the field use rate to the LC99 should be in the 30 to 40:1 range or greater. Referral to Table 2 reveals that with susceptible KCT this ratio for chlorpyrifos and methidathion is respectively 161 and 454, well above this threshold value. However, for the 'resistant' strain this ratio for chlorpyrifos and methidathion is respectively 0.64 and 42. This indicates that although methidathion is still likely to be reasonably effective against this 'resistant' strain, chlorpyrifos is likely to be very ineffective when used at the recommended field-use rate. And increasing the chlorpyrifos rate, even a doubling or quadrupling, would not be expected to provide any significant benefit. The observed increase in methidathion usage for KCT control is understandable in the light of these bioassay results.

Table 2. Summary statistics for the relative toxicity of chlorpyrifos and methidathion to susceptible and 'resistant' KCT strains, based on data presented in Section "Resistance studies, including KCT resistance to chlorpyrifos, methidathion and methomyl and baseline susceptibility to new chemistries" of this report.

Insecticide	Field-use rate (g a.i./L) (A)	Susceptible Adelaide KCT strain LC99 (g a.i./L) (B)	A/B	'Resistant' Riverland KCT strain LC99 (g a.i./L) (C)	A/C
Chlorpyrifos	0.5	0.0031	161	0.78	0.64
Methidathion	0.5	0.0011	454	0.012	42

In conclusion, the trends highlighted by this survey are consistent with the results of the insecticide bioassay study, in which significant declines in susceptibility to chlorpyrifos, and to a lesser extent methidathion, have been detected in some Riverland KCT populations. With OPs used for 80-90% of the KCT insecticide applications in the Riverland-Sunraysia, and the KCT population in Riverland-Sunraysia citrus appearing to be largely "self-contained" within citrus (with minimal opportunity for dilution of resistance by susceptible immigrants from unsprayed alternative hosts or other regions), it is not surprising that KCT is developing OP resistance. The trend towards more frequent spray applications and greater resort to methidathion are likely to continue until new insecticides with no cross-resistance to the OPs become available.

Research conducted within this project should result in at least one new insecticide (Actara™) taken through the registration process for KCT control in Australian citrus, with real potential for a second to follow (Success™). Both of these compounds have unique modes of action that differ from each other and from that of OPs and carbamates.

For effective, long-term insecticidal control of KCT, it will be necessary for the Riverland-Sunraysia citrus industry to devise and implement a resistance management strategy. This should involve the strategic timing of applications to increase efficacy and limit repeat applications, and the alternation of insecticide groups.

INSERT LANDSCAPE TABLE FROM SHERIDAN's PAPER

RESISTANCE STUDIES, INCLUDING KCT RESISTANCE TO CHLORPYRIFOS, METHIDATHION AND METHOMYL AND BASELINE SUSCEPTIBILITY TO NEW CHEMISTRIES

INTRODUCTION

Due to the increasing demand over the past decade for export-quality, blemish-free, fresh fruit, the frequency of synthetic insecticide usage in inland Australian citrus has substantially increased. Much of this insecticide usage has been organophosphate sprays (primarily chlorpyrifos) targeted to control KCT. The industry is more or less solely reliant on insecticidal control to manage KCT; no alternative controls are currently available and the capacity for effective biological control of KCT is still being investigated. Further, the choice of registered chemicals available for KCT control is limited to one carbamate and two organophosphate insecticides. Anecdotal evidence from growers and the results of a preliminary study in Project CT97007 (Baker *et al*. 2000) strongly suggested that the susceptibility of some Riverland-Sunraysia populations of KCT to chlorpyrifos had declined.

This study was undertaken with four objectives, namely:

1. to generate baseline dose-response data for a number of 'susceptible' KCT strains collected from urban and peri-urban gardens around Adelaide and the three current KCT insecticides (chlorpyrifos, methidathion and methomyl), and, based on these data, to calculate discriminating doses for each of these insecticides,
2. to assess the variability in susceptibility of these 'susceptible' KCT strains,
3. to compare the response to the three insecticides of a number of KCT strains sourced from commercial Riverland citrus orchards with that of the 'susceptible' strains, and
4. to generate baseline dose-response data for a susceptible strain of KCT to four candidate KCT insecticides (thiamethoxam, spinosad, abamectin and emamectin benzoate).

MATERIALS AND METHODS

KCT 'susceptible' strains

-Waite strain sourced from a variety of untreated host plants around the Waite campus grounds at Urrbrae (~7 km SE of Adelaide GPO), 1998-99.
-Gawler strain collected 28/04/2001 from untreated navel orange trees out of a home garden in Gawler (~40km NE of Adelaide GPO). Strain lost due to culture room failure 09/2001.
-Daw Park strain collected 10/10/2002 from untreated lemon trees out of a home garden in Daw Park (~7 km SW of Adelaide GPO).
-Flagstaff Hill strain collected 17/12/2002 from untreated lemon trees out of a home garden in Flagstaff Hill (~15 km S of Adelaide GPO).

KCT Riverland strains

KCT strains were collected from a lemon orchard at Renmark, and from navel orange orchards at Waikerie, Sunlands and Loxton.

All eight of these KCT strains were reared in laboratories and rearing rooms at the Waite campus using the rearing arena and methods described by Baker *et al*. (2000).

Bioassay method

Bioassays are performed using a similar technique as used for testing Western Flower Thrips (Herron *et al*., 1996). Fresh young lemon leaves are collected then washed and blotted dry using paper towel. Then using a 25mm cutter, discs are cut from the lemon leaves, avoiding any ribbed area formed by the centre vein of the leaf, to provide a flat leaf disc. Agar (800 gel Ace Chemical Company) is prepared at a rate of 1g/100ml, and while still molten 3mls of agar is pipetted out into 35mm diameter 10 mm high plastic petri dishes. The agar is cooled to just before setting, and then the leaf discs are embedded with the underside of the leaf facing up, and left to set. Insecticides are prepared to specific concentrations. Adult female KCT are collected via a pooter into a glass vial (20-25 individuals per vial), and then capped with a ventilated lid. They are anaesthetised via the ventilated lid with CO2 and placed on to the prepared leaf disc. Insecticide is then applied to both insect and leaf disc via a potter spray tower (Burkard, Rickmansworth Hertfordshire, England) at a deposit rate of 1.67 □ 0.079 mg cm^{-2} with a 2ml aliquot. The treated dish is then covered with plastic cling film (Cling Wrap, Clorox, Aust. Pty Ltd). Holes (100–150) are placed in the plastic film using a headless micro-needle (0.01mm x 10mm). The dishes are placed in an incubator with the dish inverted as to simulate the underside of a leaf and exposed at a constant temperature of 22 ^{0}C for 48 hrs with 14:10 L:D periods.

The bioassay is assessed visually using a stereomicroscope. Thrips are recorded as alive if observed to move, and dead if no motor response is made when gently prodded with a micro needle. Bioassays in which the control mortality exceeded 10% were not included in the analyses.

Analysis

The full log-dose data sets were analysed using probit regression analysis (Finney 1971) by an in house statistical package called Pri-Probit version 1.6 (Masayuki Sakuma 1998) on an IBM compatible computer (Protech, Australasia). Percentage mortality data produced from a discriminating dose trial or response to log doses of serial dilutions of a chemical were corrected for control mortality (Abbott 1925).

The Tested Chemicals

The product and formulation details of the seven chemicals that were tested in this study are provided in Table 1.

Table 1. The product and formulation details of the three chemicals tested in this bioassay study

Active Name	Trade Name	Chemical Group	Form	Concentration of a.i.[1]	Product R.R.[2]	Active E.R.[3]	Supplier
Chlorpyrifos	Lorsban	Organophosphate	EC	500g L^{-1}	100ml/100L	0.5 g L^{-1}	Dow Agrosciences
Methidathion	Supracide	Organophosphate	EC	400g L^{-1}	125ml/100L	0.5 g L^{-1}	Ciba-Geigy
Methomyl	Lannate	Carbamate	LC	225g L^{-1}	200ml/100L	0.45 g L^{-1}	Crop Care
Thiamethoxam	Actara	Neonicotinoid	WG	250g kg^{-1}	30g/100L	0.075 g L^{-1}	Syngenta
Spinosad	Success	Spinosyn	LC	120g L^{-1}	40ml/100L	0.048 g L^{-1}	Dow Agrosciences
Abamectin	Vertimec	Avermectin	EC	18g L^{-1}	25/100L	0.0045 g L^{-1}	Syngenta
Ema. benzoate	Proclaim	Avermectin	SG	44g L^{-1}	30/100L	0.0132 g L^{-1}	Syngenta

EC, emulsifiable concentrate; LC, liquid concentrate; WG, wettable granule; SG, soluble granule.
[1] active ingredient. [2] Recommended rate. [3] Equivalent rate.

RESULTS

Baseline dose-responses of the 'susceptible' KCT strains, and the derivation of discriminating doses (DD)

Full dose response assays with chlorpyrifos, methidathion and methomyl were successfully conducted on the four 'susceptible' strains collected from the greater Adelaide area (Table 2 and Figs. 1-3).

The LC99 value for the Waite strain, which appeared to be the most susceptible of these strains, was initially selected as the discriminating dose (DD) for chlorpyrifos and methidathion. However, there were some survivors when the other susceptible strains were tested against this DD. As a result, these two DDs were recalculated based on the LC99 values for the Gawler strain. No 'susceptibles' survived at these rates. Unfortunately the Gawler strain died out in culture before the methomyl bioassay could be run. Hence the LC99 of the Daw Park strain was chosen as the DD for methomyl.

Table 2. Dose-response data of four 'susceptible' KCT strains and one Riverland KCT strain to test chemicals.

Chemical	Strain	n[1]	Slope (±s.e.[2])	LC50 (g ai L^{-1}) (95% F.L.[3])	R.F.[4]	LC99 (g ai L^{-1}) (95% F.L.)	R.F.[5]
Chlorpyrifos	Waite	624	4.0 (0.38)	0.00082 (0.00074 – 0.00090)	-	0.0031 (0.0025 – 0.0042)	-
	Gawler	900	3.5 (0.31)	0.0015 (0.0014 – 0.0017)	1.8	0.0071 (0.0054 – 0.011)	2.3
	Daw Park	155	3.9 (0.76)	0.0023 (0.0019 – 0.0027)	2.8	0.0091 (0.0062 – 0.021)	2.9
	Flagstaff Hill	285	3.4 (0.40)	0.00099 (0.00085 – 0.0011)	1.2	0.0047 (0.0034 – 0.0078)	1.5
	Renmark	288	1.9 (0.29)	0.049 (0.038 – 0.062)	59.8	0.78 (0.42 – 2.4)	251.6
Methidathion	Waite	683	3.6 (0.43)	0.00024 (0.00020 – 0.00027)	-	0.0011 (0.00079 – 0.0017)	-
	Gawler	856	2.5 (0.42)	0.00031 (0.00025 – 0.00038)	1.3	0.0027 (0.0016 – 0.0078)	2.5
	Daw Park	134	4.2 (0.95)	0.00042 (0.00033 – 0.00051)	1.8	0.0015 (0.001 – 0.004)	1.4
	Flagstaff Hill	303	2.6 (0.35)	0.00033 (0.00028 – 0.00039)	1.4	0.0025 (0.0016 – 0.0052)	2.3
	Renmark	178	3.2 (0.71)	0.0022 (0.0017 – 0.0028)	9.2	0.012 (0.0073 – 0.036)	10.9
Methomyl	Waite	756	2.6 (0.21)	0.00044 (0.00039 – 0.00049)	-	0.0034 (0.0025 – 0.0049)	-
	Gawler	*		*		*	
	Daw Park	176	2.0 (0.36)	0.00030 (0.00025 – 0.00040)	0.7	0.0046 (0.0021 – 0.023)	1.4
	Flagstaff Hill		2.8 (0.35)	0.00028 (0.00025 – 0.00033)	0.6	0.0018 (0.0013 – 0.0033)	0.5
	Renmark	369	2.5 (0.33)	0.00086 (0.00071 – 0.0010)	2.0	0.0071 (0.0046 – 0.015)	2.1

[1] Number tested [2] Standard Error
[3] 95% Fiducial Limits [4] Resistance Factor 50 – LC 50 of strain/ LC 50 of reference strain
[5] Resistance Factor 99 – LC 99 of strain/ LC 99 of reference strain

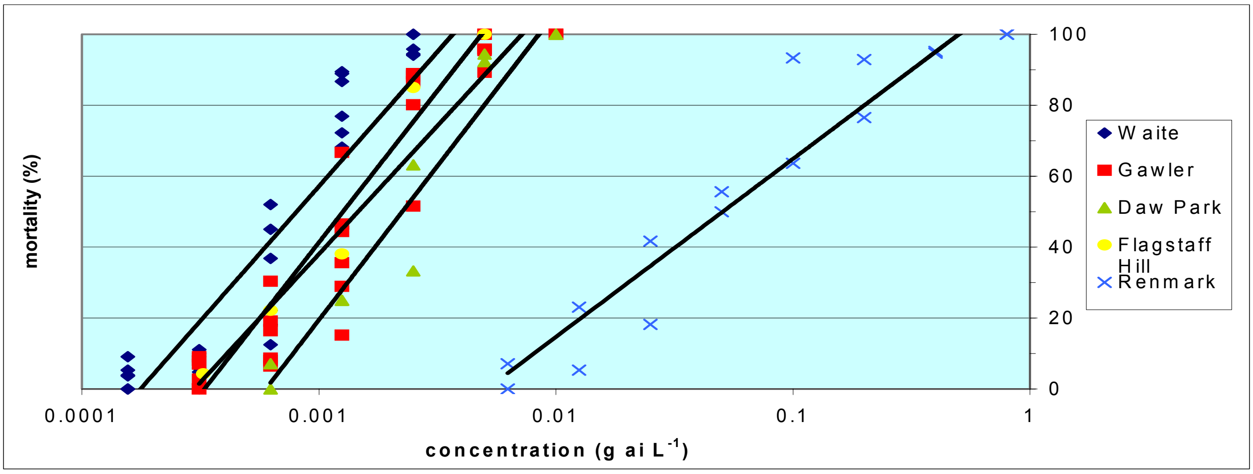

Fig. 1. Response to chlorpyrifos of Renmark field strain compared to the 'susceptible' strains.

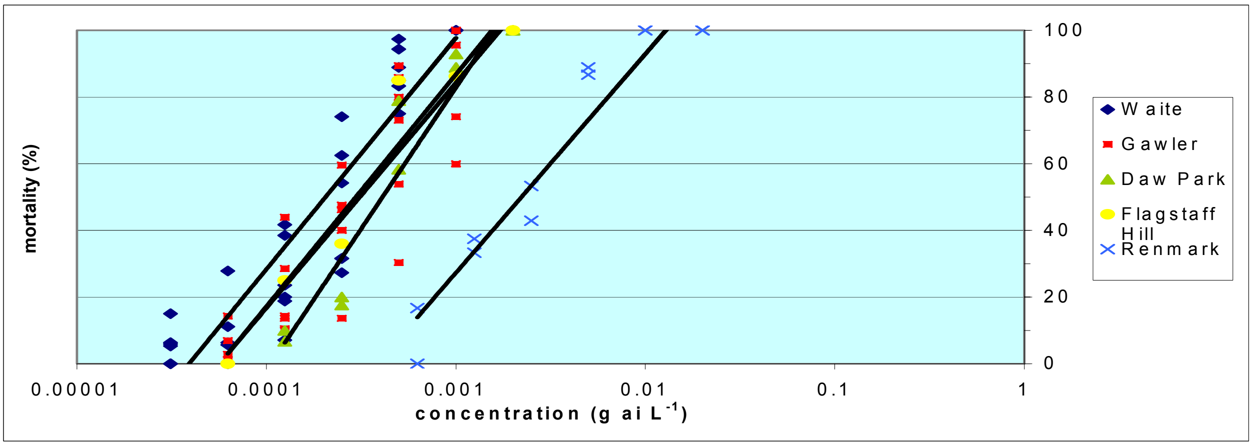

Fig. 2. Response to methidathion of Renmark field strain compared to the 'susceptible' strains.

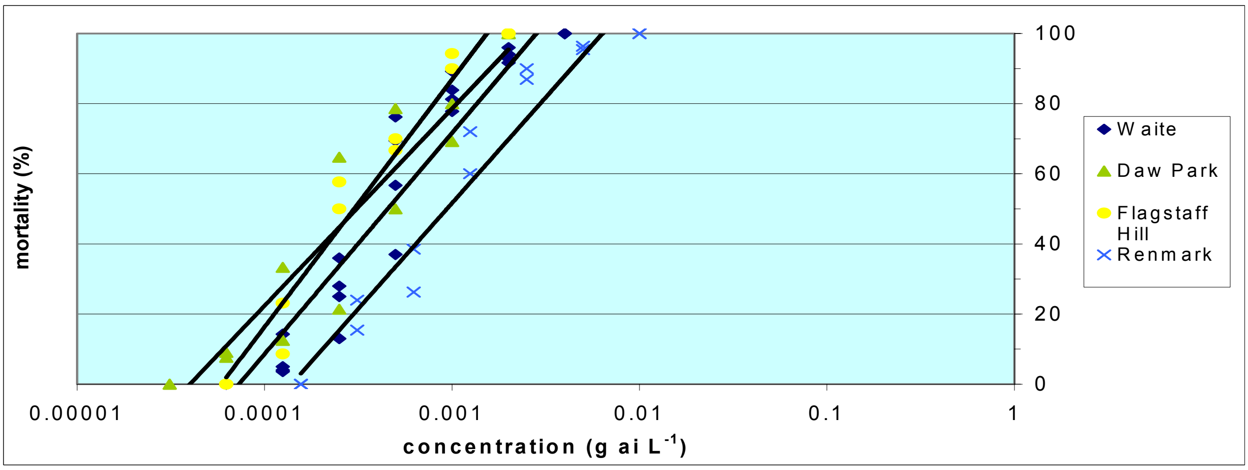

Fig. 3. Response to methomyl of Renmark field strain compared to the 'susceptible' strains.

The variability in susceptibility of the 'susceptible' KCT strains

The variability in susceptibility of the four 'susceptible' strains was quite limited; the resistance factors for chlorpyrifos, methidathion and methomyl at the LC50 were no greater than 2.8, 1.8 and 0.7 respectively, and at the LC99 were no greater than 2.9, 2.5 and 1.4 (Table 2). This suggests that the natural or unselected variability in susceptibility of KCT to these organophosphate and carbamate insecticides is minimal.

Comparison of Riverland-sourced KCT vs. 'susceptible' KCT strains

The results of screening the Riverland-sourced and 'susceptible' strains against the chlorpyrifos, methidathion and methomyl DDs are presented in Table 3. All of the 'susceptible' strains tested returned 100% mortality when challenged with these three DDs. By contrast, each of the four Riverland strains returned a low mortality when tested against the chlorpyrifos DD. The least result occurred with the Renmark strain, for which only 1.4% of tested individuals died. Significant survivorship occurred in three of the four Riverland strains when challenged with the methidathion DD, with the Sunlands strain having the greatest survivorship of 30%. All of the strains tested returned 100% mortality for methomyl.

Table 3. Percent mortality observed at the discriminating dose (DD), and the number of KCT tested.

Strain	Chlorpyrifos[1]		Methidathion[2]		Methomyl[3]	
	n[4]	% mortality	*n*	% mortality	n	% mort.
Waite[a]	59	100	56	100	75	100
Daw Park[a]	59	100	59	100	80	100
Flagstaff Hill[a]	64	100	45	100	60	100
Renmark[b]	73	1.4	74	81.1	57	100
Waikerie[b]	26	23.1	19	100	*	
Sunlands[b]	45	17.8	50	70	59	100
Loxton[b]	77	18.6	91	78.8	94	100

[1] Discriminating dose of Chlorpyrifos - 0.0075 g ai L^{-1}
[2] Discriminating dose of Methidathion – 0.003 g ai L^{-1}
[3] Discriminating dose of Methomyl – 0.005 g ai L^{-1}
[4] Number of adult female KCT tested.
* Not tested
[a] Sourced from untreated popns. in Adelaide region.
[b] Sourced from commercially sprayed Riverland orchards.

These results suggest that, compared to susceptible KCT populations that have not been exposed to insecticidal programs, at least some Riverland populations of KCT have a substantial proportion of individuals with increased fitness to chlorpyrifos and a lesser proportion of individuals with increased fitness to methidathion. By contrast, Riverland populations of KCT with an increased fitness to methomyl are probably uncommon.

These results confirm that KCT has the potential to become resistant to organophosphate insecticides such as chlorpyrifos and methidathion. Whether the reduction in susceptibility to methidathion observed in three of these Riverland strains of KCT is a response to methidathion usage, or a cross-resistance response to chlorpyrifos usage, or a combination of these, is uncertain from these data. Finally, these results provide no evidence of chlorpyrifos-methomyl cross-resistance.

The Renmark strain, which had the greatest tolerance to chlorpyrifos at the DD, was assayed with a full range of doses, and the results are presented in Table 2. Compared to the most susceptible strain (Waite), the Renmark strain has resistance factors of 59.8 times at the LC50 and 251.6 times at the LC99 for chlorpyrifos; 9.2 times at the LC50 and 10.9 times at the LC99 for methidathion; and 2.0 times at the LC50 and 2.1 times at the LC99 for methomyl. The very high resistance factor values for chlorpyrifos would be expected to result in field control failure of this Renmark strain.

To be certain of achieving good field control many entomologists and chemical companies consider that the ratio of the field use rate to the LC99 should be in the 30 to 40:1 range or greater. Referral to Table 4 reveals that this ratio for the Waite susceptible and Renmark strains is 161 and 0.6 respectively with chlorpyrifos, 455 and 42 with methidathion, and 132 and 63 with methomyl.

Table 4. The LC99 and insecticide field use rate:LC99 ratio for chlorpyrifos and methidathion on two KCT strains.

Strain	Chlorpyrifos		Methidathion		Methomyl	
	LC99 (g a.i. L^{-1})	Field use rate†:LC99 ratio	LC99 (g a.i. L^{-1})	Field use rate†:LC99 ratio	LC99 (g a.i. L^{-1})	Field use rate†:LC99 ratio
Waite	0.0031	161	0.0011	455	0.0034	132
Renmark	0.78	0.6	0.012	42	0.0071	63

†The chlorpyrifos, methidathion and methomyl field use rates are 0.5 g a.i. L^{-1}, 0.5 g a.i. L^{-1} and 0.45 g a.i. L^{-1} respectively.

Based on these chlorpyrifos data, the Waite strain would be expected to be effectively controlled by the chlorpyrifos field rate, but the Renmark strain, irrespective of good spray timing and coverage, would likely be poorly controlled or even unaffected by this chlorpyrifos rate. And increasing the chlorpyrifos rate, even a doubling or quadrupling, would not have any significant benefit. In summary, the efficacy of chlorpyrifos for KCT control has been seriously compromised against KCT strains such as the Renmark strain that have evolved high levels of chlorpyrifos resistance due to frequent exposure to this pesticide. The methidathion data suggest that the field rate of this insecticide would be highly effective on susceptible strains of KCT, and even against the Renmark strain would likely provide effective KCT control. Similarly the field rate of methomyl is likely to provide effective control of susceptible KCT strains and the Renmark strain.

Base-line dose-response data for four candidate KCT insecticides

The probit statistics for the dose-response of the Waite ‘susceptible’ strain to thiamethoxam, spinosad, abamectin and emamectin benzoate are presented in Table 5. Thiamethoxam and spinosad are presently in development for registration in Australian citrus (see “Foliar Insecticide Field Trials 2000-04” section of this report). Emamectin benzoate was field trialled for KCT control in 1999 as part of Project CT97007, where it provided a level of control that was comparable to that of chlorpyrifos. Abamectin was included in the same trial, but the control of KCT that was achieved was poor. However, abamectin is being used in New Zealand for KCT control (Dr David Steven, *pers. comm.*). These data are a valuable resource for the Australian citrus industry, when or if these candidate insecticides are adopted by the industry for KCT control. They provide the base-line reference needed to track changes over time in the susceptibility of KCT populations to these compounds, and to be able to ascertain whether cross-resistance between these insecticides and other insecticide groups used for KCT control is likely. The latter information is essential for the design of a robust insecticide resistance management strategy.

Table 5. Dose-response data of Waite "susceptible" KCT strain tested against four candidate KCT insecticides.

Chemical	n[1]	Slope (±s.e. [2])	LC50 (g ai L^{-1}) (95% F.L. [3])	LC99 (g ai L^{-1}) (95% F.L.)
Abamectin	350	2.99 (0.60)	0.0019 (0.0013 – 0.0027)	0.011 (0.0062 – 0.048)
Emamectin benzoate	430	2.23 (0.23)	0.0090 (0.0075 – 0.011)	0.099 (0.065 – 0.18)
Spinosad	273	3.49 (0.39)	0.00059 (0.00051 – 0.00069)	0.0027 (0.002 – 0.0043)
Thiamethoxam	328	2.65 (0.40)	0.00047 (0. 00038 – 0.00057)	0.0036 (0.0023 – 0.0079)

[1] Number tested [2] Standard Error [3] 95% Fiducial Limits

DISCUSSION

The results of this study confirm earlier reports of a decline in susceptibility of some Riverland-Sunraysia populations of KCT to chlorpyrifos, and demonstrate that in some instances this decline is of sufficient magnitude that chlorpyrifos field spraying of KCT would be ineffective. Further, this study documents a moderate decline in susceptibility of some KCT populations to methidathion. No evidence of lesser susceptibility to methomyl was detected in the study's limited screening of Riverland KCT populations.

Results of a 2003 survey (reported in "KCT Insecticide Resistance Survey of Riverland Citrus Growers" section of this report) reveal that insecticidal control of KCT in the Riverland currently averages 1.0-1.1 spray applications per orange orchard. Across the Riverland 62.4%, 30.8% and 3.0% of these applications consist of chlorpyrifos, methidathion and methomyl respectively. This survey documents that over the past five years the frequency of spraying for KCT control has increased, and that growers are increasingly substituting methidathion in place of chlorpyrifos. These changes in spray practices are an understandable response to the decline in chlorpyrifos performance, which is occurring as a result of the evolution of chlorpyrifos resistance in KCT populations in citrus across the Riverland-Sunraysia region.

A number of important questions remain unanswered.

Firstly, what are the cross-resistance patterns of insecticides used for KCT control? This knowledge is critical in choosing alternative chemicals and in designing an insecticide resistance management (IRM) rotation strategy. Is methidathion-chlorpyrifos cross-resistance contributing to the decline in methidathion susceptibility? Does cross-resistance between the carbamate methomyl and either of these OP insecticides occur? Resistance to both OPs and carbamates appears to often involve acetyl-cholinesterase insensitivity, and cross-resistance between certain OPs and carbamates is known (Immaraju *et al.* 1989, Sparks 1998, Sun 1990). Therefore, despite the lack of evidence for OP-methomyl cross-resistance in this small-sample study, the possibility of such cross-resistance in KCT should not discounted. Is cross-resistance likely between these established KCT insecticides and the new chemistry under development? Encouragingly the modes of action of the neonicotinoid thiamethoxam, the spinosyn spinosad and the macrocylic lactones abamectin and emamectin benzoate are each different from each other, and in turn different from OPs and carbamates (Sparks 1998). However, little is known about the mechanisms by which these new insecticides are metabolised in insects. Cross-resistance may occur if they are metabolised by similar enzyme systems.

Secondly, what is the stability of the resistance to chlorpyrifos and methidathion in KCT populations? Does it decline in a reasonable time period in the absence of OP usage due to reduced fitness of resistant individuals? This information is important for the design of an IRM rotation strategy.

Thirdly, how much KCT population flow is occurring between neighbouring citrus orchards? If sufficient amounts of population (gene) exchange are occurring, then a 'refuge' strategy whereby insecticides that are used for KCT control are not used in citrus blocks that are at low risk from KCT damage (eg. Valencia oranges, mandarins), would slow the development of resistance in nearby high-risk blocks (eg. navel oranges, mandarins, lemons). This strategy would involve using alternative controls for other key pests (eg. *Bacillus thuringiensis* or Isomate™ pheromone disruption for lightbrown apple moth, oil sprays for scale pests, Applaud™ for mealybugs) in those blocks at low risk from KCT. (Please refer to the Summary at the end of the "Foliar Insecticide Field Trials 2000-04" Section of this report for a discussion of the essential features of an effective "window" IRM strategy for KCT in Riverland-Sunraysia citrus.)

In conclusion, this study has importantly established baseline susceptibility levels for the three main insecticides currently used for KCT in the Riverland-Sunraysia region, and for four new candidate insecticides. It has established discriminating doses for these insecticides that allow cost-effective, quick diagnosis of any shifts in resistance, and allow judgement whether field control failures were due to the development of resistance or due to faulty spray application or timing. Finally, sound resistance management will be the key to effective, long-term insecticidal control of KCT. For this reason an IRM rotation strategy, based on an understanding of cross-resistance relationships between the KCT insecticides, must be adopted by the industry when the new KCT insecticides become available.

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LITERATURE CITED

Abbott W.S. 1925. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* **18**: 265-267.

Baker, G., Jackman, D. J., Keller, M., MacGregor, A., & Purvis, S. 2000. Development of an Integrated Pest Management system for thrips in Citrus (Adelaide, SARDI).

Finney, D.J. 1971. Probit analysis. 3rd Edition. Cambridge University Press: Cambridge.

Herron, G.A., Rophail, J. and Gullick, G.C. 1996. Laboratory-based, insecticide efficacy studies on field-collected *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) and implications for its management in Australia. *Australian Journal of Entomology* **35**: 161–164.

Immaraju, J.A., Morse, J.G and Gaston, L.K. 1990. Mechanisms of organophosphate, pyrethroid, and DDT resistance in citrus thrips (Thysanoptera: Thripidae). *Journal of Economic Entomology* **83**: 1723–1732.

Sakuma, M. 1998. The Instructions for PriProbit ver. 1.6. Kyoto.

Sparks, T.C. 1998. New insecticides and acaricides: mode of action and potential role in IPM. *In*: Zalucki, M.P., Drew R.A.I. and White G.G. (eds.). Pest Management – Future Challenges: Proceedings of the Sixth Australasian Applied Entomological Research Conference, Univ. of Queensland, Brisbane, Australia, 29 September-2 October 1998. Vol. 2, pp: 123-130.

Sun, C. 1990. Insecticide resistance in diamonback moth. *In*: Talekar, N.S. (ed.). Management of diamondback moth and other crucifer pests: Proceedings of the Second International Workshop, Tainan, Taiwan, 10-14 December 1990. pp: 419-426.

FOLIAR INSECTICIDE FIELD TRIALS 2000-04

INTRODUCTION

Insecticides are currently the only means of controlling KCT in Australian commercial citrus orchards. Growers are limited to treating KCT with organophosphate and carbamate insecticides registered for use in citrus against other pests. Both of these insecticide groups have the same mode of action (acetylcholinesterase inhibition). If a pest is consistently exposed to insecticides with the same mode of action they can rapidly develop resistance to those insecticide groups. An important component of Integrated Pest Management is to delay the development of resistance by reducing the overall use of chemicals and alternating the use of insecticide groups.

At the initiation of this Project (CT00015), enquires were made with chemical companies regarding any new insecticides they were developing for the Australian market which had demonstrated efficacy against thrips species in other crops. Chemical companies were then asked to consider their interest in participating in our field trials and their commitment in taking the product through to registration, should it prove successful against KCT.

Syngenta, DowAgro Sciences and BASF provided one insecticide each in the first year. Importantly each of these new insecticides had unique modes of action. Despite chlorfenapyr performing well against KCT, BASF pulled out of subsequent trials, as they had no interest in pursuing chemical registration for the Australian citrus industry.

In addition to the assessment of new candidate chemistry, the use of a low rate regime of oil-sprays at ten day intervals throughout the flowering and early fruit set KCT risk period was field trialled in an unreplicated design, the synergistic effects of combining the Caltex oil product Brella™ with several of the KCT candidate insecticides were assessed, and a field trial was designed to test the efficacy of a sequence of Surround™ (a kaolin clay particle film product) sprays for KCT control.

2000-01 RIVERLAND TRIAL OF NEW CANDIDATE INSECTICIDES

METHODS

Site and Treatment details

The 2000/01 trial was conducted on a commercial navel block at Sunlands, in the Riverland of South Australia.

Table 2. Details of navel block used for the Sunlands insecticide trial, 2000-01.

Location	Sunlands
Soil Type	Sandy loam
Tree Age	35-40 years
Variety	Leng navels
Block Design	Single tree
Irrigation Design	Low level
Ground management	Bare earth

Treatments were replicated 4 times in a randomised block design. Each plot consisted of 9 trees (3 x 3), of which only the central tree was monitored for insect activity and crop damage. The single-tree plot design used in a previous project (CT97007) was replaced with a larger plot design in which unsprayed buffer trees were eliminated in an attempt to better represent the effect of applying insecticides on a commercial scale.

Sprays were applied using a commercial oscillating boom sprayer at 6000L/ha. Treatments were applied on 15th November when assessment of a 100 fruit sample indicated that >5% of the fruit was infested by KCT larvae. A follow-up spray was applied four weeks later (14th December).

To prevent any cross-contamination of treatments from residual spray within the sprayer, all replicates for one treatment were sprayed, then all replicates for each subsequent treatment. Before each new treatment was mixed, the sprayer was thoroughly flushed with water to remove residues of the previous chemical.

Table 3. Treatments applied in insecticide trial at Sunlands, Riverland 2000.

Dates of Application		**Treatment**	**Insecticide Product**	**Active Ingredient**	**Chemical Group**	**Rate of Product applied per 100L water**
-	-	A	Unsprayed (control)	-	-	-
15/11/00	14/12/00	B	Lorsban™	500g ai/L chlorpyrifos	Organophosphate	100ml
15/11/00	14/12/00	C	Actara™	250 g ai/kg thiamethoxam	Neo-nicotinoid	30gm + 0.5% summer oil
15/11/00	14/12/00	D	Secure™	360 g ai/L chlorfenapyr	Pyrrole	20ml
15/11/00	14/12/00	E	Success™	120 g ai/L spinosad	Spinosyn	40ml
15/11/00	14/12/00	F	Supracide™	400g ai/L methidathion	Organophosphate	125ml

Assessment of KCT activity and damage

To determine the timing of treatment application ($\geq$ 5% KCT larvae incidence), a 10× hand lens was used to monitor fruit in the field. One hundred fruit were sampled at random throughout the block, calyces peeled off and KCT incidence recorded. Treatments were applied when sampling revealed that KCT larvae infested 5% or more of the examined fruit. Other insects seen under the calyx were also noted.

To determine the effect of treatments on KCT, the central tree of each plot was monitored weekly for KCT adult and larvae incidence, as well as general insect activity under the calyx. Twenty fruit per plot were collected directly into 80% alcohol and taken back to the lab for processing. When fruit became too large to fit into sample jars, the top quarter of each fruit (including the calyx) was removed and placed directly into alcohol.

To determine the efficacy of treatments in controlling KCT damage, fruit from the central tree of each plot was assessed at harvest. 100 fruit per plot was picked from the top and sides of the central tree. Fruit were assessed for scurfing incidence and severity (Table 4).

Table 4. System for rating the severity of scurfing.

Scurfing Severity Rating	Definition
1	Nil halo to halo markings up to half circumference around calyx.
2	Halo markings half to full circumference around calyx.
3	Broad, expanded markings, band > 5mm.

Statistical Analysis

Statistix was used to analyse the data. The effect of treatments on the KCT population was analysed with ANOVA, with differences tested for significance using the t-table at the probability level of P= 0.05. Data were log (x+0.5) transformed for analysis of the effect of the treatments on KCT activity.

RESULTS

Effect of insecticides on KCT incidence

The first application of insecticide treatments (15/11/00) significantly reduced the number of KCT larvae from that observed in unsprayed control plots (Table 5). However, there was no significant difference in larval KCT control between the five insecticides.

The second application of treatments on 14 December 2000 had no significant effect on the KCT population, as densities were already low.

Treatment effect on KCT adults could not be assessed, as the density was too low in all plots for statistical analysis.

Table 5. Log transformed mean incidence of KCT larvae per 20 fruit assessed 8 days (23/11/00) after a single application of the treatments (15/11/00) at Sunlands, Riverland. Means followed by the same letter are not significantly different (LSD test).

Treat-ment	Control	Chlorpyrifos	Thiamethoxam	Chlorfenapyr	Spinosad	Methidathion	*LSD*	*F prob*
Log trans-formed mean	0.3874 a	-0.0625 b	-0.3010 b	-0.1817 b	-0.1817 b	-0.1817 b	*0.3689*	*0.017*
Untrans-formed mean	2.25	0.5	0	0.25	0.25	0.25		

Effect of insecticides on the incidence of halo fruit damage resulting from KCT feeding

All treatments significantly reduced the incidence of each KCT Scurfing Severity Rating below that of unsprayed control plots. Methidathion, chlorfenapyr, thiamethoxam all performed significantly better than chlorpyrifos in controlling the incidence of severe (Rating 3) and mild (Rating 1) KCT-inflicted halo damage. Spinosad treatments also significantly reduced the incidence of severe (Rating 3) KCT damage compared to chlorpyrifos treatments (Table 6).

These results are encouraging, with positive implications for the citrus grower. This experiment demonstrates that the use of these alternative insecticide treatments can result in significant financial gain. Fruit classed as Rating 2 or 3 are most likely to be sold as juice fruit for a minimal price. Significantly increasing the percentage of fruit with mild (Rating 1) or nil halo damage will also significantly increase the profit to be gained.

Table 6. Mean incidence of KCT damage per 100 fruit at Sunlands, Riverland (August 2001).

Scurfing Severity Rating**	**unsprayed control**	**Chlorpyrifos**	**thiamethoxam**	**chlorfenapyr**	**spinosad**	**methidathion**	***LSD***	***F prob***
3	11.5a*	5.0b	0.75c	0.75c	1.0c	1.25c	*3.0918*	*0.0*
2	10.25a*	4.0b	1.75b	0.75b	2.5b	0.5b	*3.7553*	*0.0*
1	78.25a*	91.0b	97.5c	98.5c	96.5bc	98.25c	*5.6326*	*0.0*

*means followed by the same letter are not significantly different (P=0.05)
**See Table 4 for Scurfing Severity Ratings.

DISCUSSION

Thiamethoxam (Actara™), chlorfenapyr (Secure™), spinosad (Success™), methidathion (Supracide™) and the industry-standard chlorpyrifos (Lorsban™) each resulted in a significant reduction in the incidence of KCT infestation on treated fruit eight days after the first application of these treatments. Although the level of larvae control provided by these five treatments ranged between approximately 80-100%, the differences between these treatments were not statistically significant.

At the August 2001 harvest assessment of halo damage the thiamethoxam, chlorfenapyr, spinosad and methidathion treatments were shown to have resulted in greater reduction in the incidence and severity of halo damage compared with chlorpyrifos.

These results are encouraging. They are very similar to the results of the Waite trials of January-February 2001 (reported below), and indicate that the new chemistries of chlorfenapyr, spinosad and thiamethoxam each have promise as candidate insecticides for KCT control in citrus orchards. Each of these new chemicals has unique modes of action and would therefore be valuable in an insecticide resistance management strategy in rotation with chlorpyrifos.

2001 WAITE CAMPUS TRIAL

INTRODUCTION

A study was initiated in January 2001 at the Waite campus to compare the efficacy of five insecticides for the control of KCT. This study was undertaken because of concern that low KCT abundance may have compromised the value of the 2000-01 Riverland (Sunlands) insecticide trial reported above.

METHODS

The insecticides (and spray rates) tested were chlorfenapyr (20 ml Secure™ 100L^{-1}), spinosad (40 ml Success™ 100L^{-1}), methidathion (125 Supracide™ 100L^{-1}), thiamethoxam (30 g Actara™ 100L^{-1}) and chlorpyrifos (100ml Lorsban™ 100L^{-1}).

The treatments, including a water control, were each applied with yellow fluorescent dye by hand sprayer to orange trees in the Waite orchard. At 2 hours and 3 to 11 days after spraying, leaves were picked and examined under UV light. A single 25 mm diameter leaf disc was cut from leaves with similar spray deposition (4 leaf replicates per treatment) and embedded in agar in 35 mm diameter petri dishes. 15-30 adult or larval KCT were added to each arena, and mortality was assessed after 24 hours.

In the first experiment (commenced on 18 January 2001) both adult and second instar larval KCT were tested. In the second experiment (commenced on 5 February 2001) only adult KCT were tested.

RESULTS

In the first experiment with adult KCT, chlorfenapyr (Secure™), spinosad (Success™), methidathion (Supracide™) and chlorpyrifos (Lorsban™) each caused 100% mortality on 0 DAT (days after treatment) (Fig. 1). In the thiamethoxam (Actara™) treatment arenas 7.1% of the adult KCT were still surviving at the 24 hour assessment. Thiamethoxam is known to be somewhat slow to act, so this lesser performance after only 24 hours is not unexpected. At 4 DAT chlorfenapyr (Secure™) and methidathion (Supracide™) produced 100% mortality, spinosad (Success™) and thiamethoxam (Actara™) resulted in very good control (94% and 94.5% respectively), and chlorpyrifos gave significantly lesser control (62.8%). At 11 DAT the insecticidal effect of all five products had largely dissipated, but the greater persistence of the chlorfenapyr (Secure™) and methidathion (Supracide™) treatments was still evident.

The susceptibility of the 2nd instar larval KCT closely resembled that of the adults, as evidenced by the similar mortality estimates for the 0 DAT and 3 DAT (larvae) vs 4 DAT (adults) data-sets in the first experiment (Fig. 2). Further, the relative performance of the five insecticides in the larval experiment was broadly similar to that observed in the adult experiment. At 0 DAT chlorfenapyr (Secure™), spinosad (Success™), methidathion (Supracide™) and chlorpyrifos (Lorsban™) each caused 100% larval mortality, while the thiamethoxam (Actara™) treatment produced 92.9% mortality. At 3 DAT all five insecticides provided a reasonably good kill of KCT larvae, with the lowest mortality unexpectedly provided by the chlorfenapyr (Secure™) treatment. At 7 DAT the chlorpyrifos (Lorsban™) treatment was significantly less effective than the other four products (Contingency table analysis, ©^{2}=30.78, P=0.0000), which parallels the lesser residual performance of this insecticide observed in the 4 DAT adult KCT data-set.

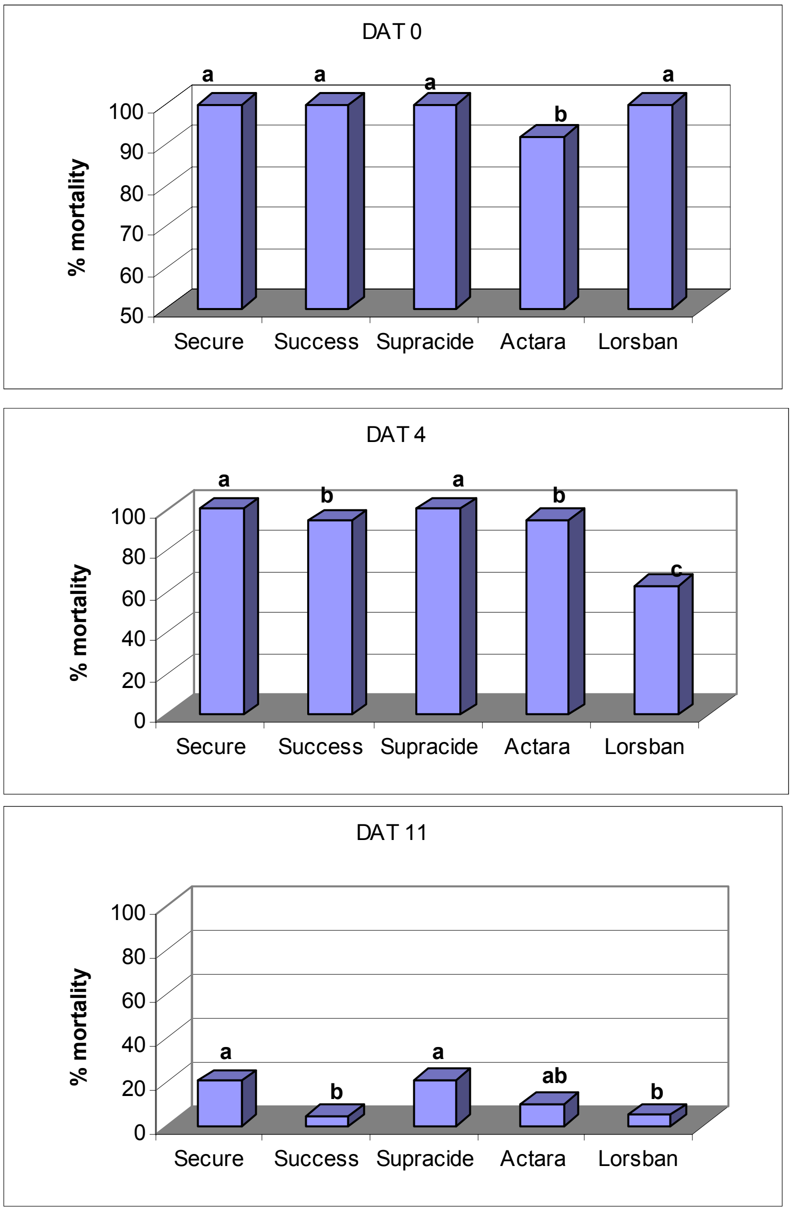


Fig. 1. Mortality of KCT adult after 24 hours exposure to weathered insecticide residues on Waite orchard citrus foliage sprayed on 18 January, 2001.

In the second experiment with KCT adults similar results were obtained (Fig. 3). On 0 DAT all five insecticides gave good to excellent control; chlorfenapyr (Secure™), methidathion (Supracide™) and chlorpyrifos (Lorsban™) gave 100% mortality, and spinosad (Success™) and thiamethoxam (Actara™) 90.9% and 95.9% respectively. At 3 DAT spinosad (Success™), thiamethoxam (Actara™) and chlorpyrifos (Lorsban™) were significantly less effective than either chlorfenapyr (Secure™) or methidathion (Supracide™) (Contingency table analysis, ©^{2}=8.41, *P*=0.0037). At 7 DAT the greater persistence of the insecticidal performance of both chlorfenapyr (Secure™) and methidathion (Supracide™) was again evident, and, in contrast to the results of the first experiment, chlorpyrifos (Lorsban™) demonstrated good persistence by still causing 90.6% mortality.

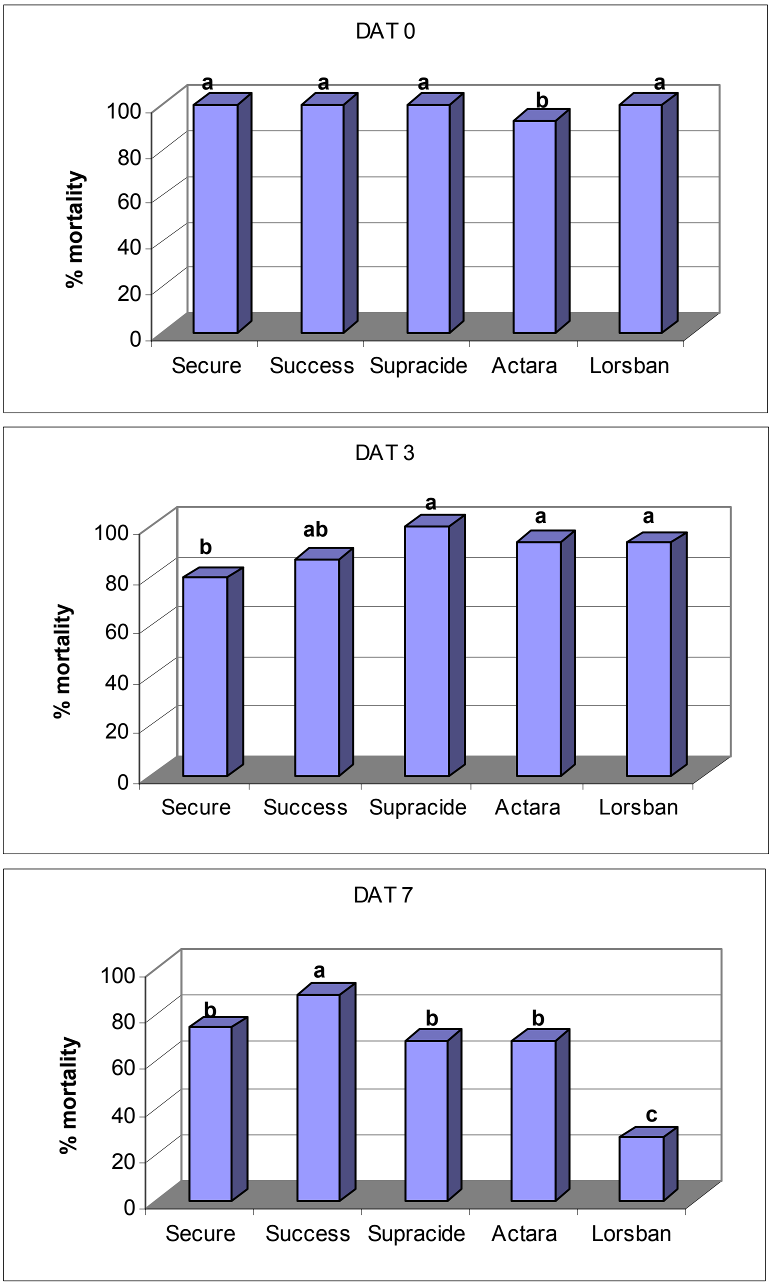


Fig 2. Mortality of KCT larvae after 24 hours exposure to weathered insecticide residues on Waite orchard citrus foliage sprayed on 18 January, 2001.

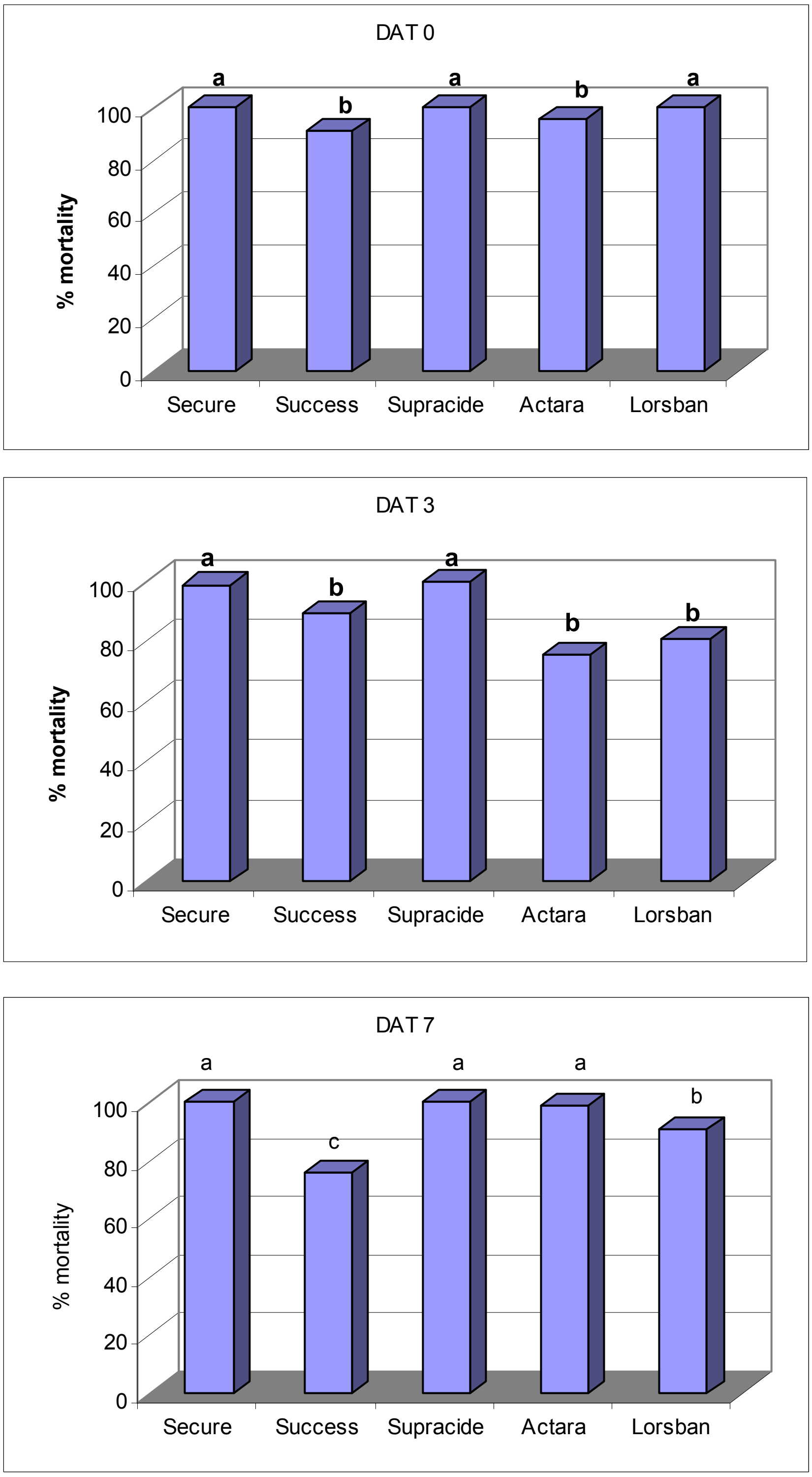


Fig 3. Mortality of KCT adults after 24 hours exposure to weathered insecticide residues on Waite orchard citrus foliage sprayed on 5 February, 2001.

DISCUSSION

Chlorfenapyr (Secure™) and methidathion (Supracide™) each performed as well as, or superior to chlorpyrifos, which is the industry standard for KCT control in the Riverland-

Sunraysia-Riverina region. They both caused 100% mortality in each experiment at 0 DAT, and provided the most persistent kill of the five products tested. However both of these compounds are broad-spectrum insecticides. This characteristic, along with their considerable field persistence, means that they are likely to cause significant disruption to the biological control provided by numerous beneficial organisms in the citrus ecosystem. For this reason they should only be considered for KCT registration if other suitable compounds are not available.

Spinosad (Success™) and thiamethoxam (Actara™) performed as well as, or better than, chlorpyrifos in seven and six of the nine data-sets respectively. In the case of thiamethoxam, which is known to be slow acting, these 24 hour assays may actually underestimate the field efficacy of this compound. Generally in these experiments both spinosad and thiamethoxam were as initially toxic to KCT as chlorpyrifos, and exhibited similar field persistence to that of chlorpyrifos. In contrast to chlorfenapyr and methidathion, both spinosad and thiamethoxam are relatively narrow-spectrum insecticides with a moderate level of field persistence. For these reasons it is expected that they would cause significantly less disruption to citrus ecosystems than chlorfenapyr, methidathion or chlorpyrifos. On this evidence it is considered that both spinosad and thiamethoxam should be further trialed for KCT control.

2000-01 RIVERLAND TRIAL OF A LOW-RATE, MULTIPLE OIL SPRAY STRATEGY FOR KCT CONTROL

INTRODUCTION

Liu *et al*. (2002) demonstrated in a laboratory study significant potential for using 0.5%-1.0% horticultural spray oil applications to maintain greenhouse thrips populations below economic threshold on citrus through behavioural effects of the oil deposits on feeding by larvae and adults and on oviposition by adult females. This small, unreplicated field trail was undertaken in 2000 to assess the effect of a sequence of 0.75% oil spray applications on the incidence of KCT damage in a Leng navel orange crop. The trial was arranged through James Altmann of Fruit Doctors (Loxton), and Fruit Doctors staff made the pre-harvest assessments.

METHODS

The effect on KCT abundance and feeding damage of a series of five 0.75% DC Tron Plus sprays applied at approximately 8,000 L/ha by oscillating boom at 10 day intervals from 11 October to 21 November 2000 to a crop of Leng navels at Solora, Loxton North was assessed and compared to the KCT abundance and feeding damage in several adjacent rows of unsprayed (control) Leng navels.

RESULTS AND DISCUSSION

The oil spray regime had no significant effect on KCT abundance. The pre-harvest assessment of the incidence of halo damage on sampled fruit is presented in Table 6 below. Each of the treatment and sample height pre-harvest assessments consisted of 20 quadrat (approx 0.5m x 0.5m) samples of 0-19 fruits. Contingency table analyses revealed that there were no statistically significant differences in damage incidence between either the oil-treated and control fruit samples (©^{2}=0.04, *P*=0.98), nor between the low and high height samples (©^{2}=2.72, *P*=0.26).

Table 6: The percentage of Leng navel fruit in each of three damage categories in untreated and DC Tron Plus treated samples, Solora, Loxton North, 2000-01.

Treatment	Sample height	% of fruit in each of 3 damage categories †			No. of fruit sampled
		Low	Moderate	High	
Control	Low	79.6	10.5	9.9	152
	High	83.2	10.7	6.1	197
Oil	Low	84.2	7.5	8.3	133
	High	80.2	13.5	6.3	126

† The three damage categories of low, moderate and high respectively refer to fruit with a halo circumference of 0-25%, 25-75% and >75%.

The sequence of five oil applications, applied at 10-day intervals from well before the period of crop risk from KCT feeding, and continuing until well after this risk period, was a simple, preliminary means of determining whether an oil-spray strategy might influence KCT behaviour and thereby reduce crop damage. Because of this discouraging outcome, and the limited project resources, this avenue of investigation was not pursued further.

2001-02 RIVERLAND TRIAL

INTRODUCTION

In the 2000-01 foliar insecticide trial, thiamethoxam and spinosad provided effective control of KCT damage, and resulted in lesser scurfing than the industry standard chlorpyrifos. These encouraging results prompted the re-selection of thiamethoxam and spinosad for further trailing with chlorpyrifos.

METHODS

Site and Treatment details

The site used for the 2000-01 Riverland trial was again used in 2001-02.

Treatments (Table 7) were replicated 4 times in a randomised block design. Each plot consisted of 9 trees (3 x 3), of which only the central tree was monitored for KCT activity and damage.

Sprays were applied using an oscillating boom sprayer at 5000L/ha. Treatments were applied when KCT larval density exceeded 5% incidence (13/11/01). A second application of treatments occurred on 5/12/02.

To prevent any cross-contamination of treatments from residual spray within the sprayer, all replicates for one treatment were sprayed, then all replicates for each subsequent treatment. Before each new treatment was mixed, the sprayer was thoroughly flushed with water to remove residues of the previous chemical.

Table 7. Treatments applied in insecticide trial at Sunlands, Riverland 2001-02.

Date of Application		**Treatment**	**Insecticide Product (and Chemical Group)**	**Active Ingredient**	**Product Rate per 100L water**
-	-	A	Unsprayed (control)	-	-
13/11/01	5/12/01	B	Lorsban™ (Organophosphate)	500 g ai/L chlorpyrifos	100ml
13/11/01	5/12/01	C	Success™ (Spinosyn)	120 g ai/L spinosad	40ml
13/11/01	5/12/01	D	Actara™ (Neo-nicotinoid)	250 g ai/kg thiamethoxam	30gm + 0.25% DC-Tron Plus

Assessment for KCT activity and damage

To determine the timing of treatment application (>5% KCT larvae incidence) one hundred fruit were sampled, at random throughout the block, into jars of 80% alcohol for processing in the laboratory using a microscope.

To determine the effect of treatments on KCT incidence, 125 fruit were sampled from the central tree of each plot 10 days post-treatment (30/11/01).

To determine the efficacy of treatments in controlling KCT damage, 100 fruit from the central tree of each plot were scored for KCT damage using the Scurfing Severity Rating (Table 4) at harvest (18/7/02). Fruit with a Scurfing Severity rating of '1' are considered acceptable for most markets (J. Altmann, pers. comm.) and for the purpose of this experiment, are added to the count of fruit with a Scurfing Severity rating of '0'.

Statistical Analysis

Data were analysed with ANOVA using Statistix for Windows. For the analysis of the effect of treatments on KCT larvae and adult activity, data were log (x+1) transformed.

RESULTS

Effect of insecticides on KCT activity

Thiamethoxam was the only treatment to cause a significant reduction in KCT larval incidence 10 days post application (Table 8). It is possible that the two other insecticides caused a significant reduction in KCT larval incidence in the days immediately following spraying, but because these insecticides have a limited residual impact this effect was not detectable 10 days after treatment application

There was no treatment effect on KCT adult incidence.

Table 8. Log transformed mean incidence of KCT per 125 fruit sample 10 days post-treatment (30/11/01) at Sunlands, Riverland, 2001. Means in the same row followed by the same letter are not significantly different using the LSD test (*P*=0.05).

Treatment	**A (control)**	**B (chlorpyrifos)**	**C (spinosad)**	**D (thiamethoxam)**	***LSD***	***Fprob***
Larvae log transformed mean	1.29b	1.07b	1.17b	0.27a	*0.44*	*0.00*
Larvae un-transformed mean	19.25	12.75	16.75	1.0	-	-
Adult log transformed mean	0.15a	0.23a	0.12a	0.08a	*0.29*	*0.69*
Adult un-transformed mean	0.50	0.75	0.50	0.25	-	-

Effect of insecticides on the incidence of halo fruit damage resulting from KCT feeding

Chlorpyrifos and thiamethoxam treatments significantly reduced the incidence of KCT damage below that observed in unsprayed control plots. Scurfing severity in the spinosad treatment plots was intermediate between that recorded in the control and the other two insecticide treatments, and did not differ significantly from any of these other treatments (Table 9).

Table 9. Mean percentage of fruit with scurfing of Severity Rating 2-3* at harvest, Sunlands, Riverland, 2002. Means followed by the same letter are not significantly different using the LSD test (*P*=0.05).

Treat-ment	**A (control)**	**B (chlorpyrifos)**	**C (spinosad)**	**D (thiamethoxam)**	***LSD***	***Fprob***
Mean damage incidence	14.0b	2.5a	8.0ab	2.75a	*8.59*	*0.04*

*Refer to Table 4 for Scurfing Severity Ratings.

DISCUSSION

The application of thiamethoxam resulted in a significant reduction in the incidence of KCT larvae infestation on treated fruit 10 days after treatment application. Results of the 2001 Waite Campus trial reported earlier in this Section suggests that the efficacy of spinosad and chlorpyrifos residues on the mortality of adult KCT drops from 82% and 93% respectively three days after application to less than 5% seven days after application. Whereas thiamethoxam residues still achieve 50% adult KCT mortality nine days after application. This information provides a possible explanation for spinosad and chlorpyrifos not achieving significant control of KCT larvae 10 days after treatment.

At harvest there was significantly less KCT damage recorded in plots treated with thiamethoxam and chlorpyrifos than in unsprayed control plots. Although the incidence of scurfing damage in the spinosad plots was almost half that recorded in the control plots, this difference was not statistically significant.

This 2001-02 trial provides further evidence of the potential of thiamethoxam as an effective alternative insecticide for the control of KCT.

2002-03 ACTARA™ RIVERLAND TRIAL

INTRODUCTION

The 2002-03 trial was undertaken in order to ensure an adequate data set for the foliar application of Syngenta's product Actara™ (thiamethoxam) and therefore aid Syngenta's application for registration of this product in citrus. At Syngenta's request two alternative application methods were also trialled, including water-soluble bags attached to the trunk and trunk sprays. Syngenta's in-house preliminary trials had indicated that while it takes the active constituent 2-3 days to reach the foliage, these alternative application methods would provide longer and better efficacy (Dr Craig Clarke, Syngenta, *pers. comm.*).

METHODS

Site and Treatment details

The 2002-03 Actara™ trial was split over two commercial navel blocks, which were situated within a kilometre of each other at Sunlands in the Riverland. Block A was comprised of 3 rows of +25year old navels and was flanked by Washington navels, Valencias and grapevines and watered by overhead sprinklers. Block B contained +25year old navels and was flanked on both sides by Valencias and watered by under-tree sprinklers.

Foliar and trunk applications were trialed at a range of rates (Table 10). Treatments were replicated 4 times in a randomised block design (2 reps each in navel blocks A and B). Each plot consisted of 9 trees (3 x 3) with the exception treatment J, which had a plot size of 3 trees (1 x 3) due to a limited number of trunk bands. Only the central tree of each plot was monitored for KCT activity and damage.

Foliar treatments were applied using a commercial oscillating boom sprayer at 5000L/ha. Trunk sprays were applied using a 750mL hand held atomiser, 20mL of treatment applied per trunk. Trunk bands comprised of 4 water-soluble bags (0.5g WG25 Actara each) fixed to the inner side a of 8cm wide PVC band, the bottom of the band taped around the trunk. Water, from rain or irrigation, penetrates between the trunk and the top of the band, dissolving the water-soluble bags. The active ingredient then penetrates through the bark into the tree, where it is transported in the xylem up to the leaves and fruit.

Table 10. Application and rate treatments for Actara (thiamethoxam) field evaluation trials for the control of KCT in citrus, Sunlands, Riverland, 2002-03.

Date of Application	Treatment	Chemical	Application method	g ai/100L	Product/100L
			-	-	-
27/11/02	B	Thiamethoxam	foliar	2.5	10
27/11/02	C	Thiamethoxam	foliar	5	20
27/11/02	D	Thiamethoxam	foliar	7.5	30
27/11/02	E	Thiamethoxam	foliar	10	40
27/11/02	F	Thiamethoxam + wetter	foliar	5	20
27/11/02	H	Methidathion	foliar	50	125
11/11/02	G	Thiamethoxam	trunk spray	0.5/tree	2g/tree
11/11/02	I	Thiamethoxam	trunk spray	1.0/tree	2g/tree
11/11/02	J	Thiamethoxam	trunk band	2	4 bags/tree

Assessment of KCT activity and damage

To determine the timing of treatment application (>5% KCT larvae incidence) one hundred fruit were sampled, at random throughout the block, into jars of 80% alcohol for processing in the laboratory using a microscope.

Trunk treatments were applied on 11/11/02. Unfortunately foliar application of treatments did not occur until 27/11/02 due to a combination of irrigation scheduling, windy conditions and farm-hand illness.

Assessments at pre-treatment and 7, 21 and 42 days after treatment were made by collecting 100 fruit from the central tree of each plot into 80% alcohol (due to a lack of fruit, only 25 fruit per plot were collected for 21 and 42 days after treatment assessments). These samples were then processed in the laboratory using a microscope.

To determine the efficacy of treatments in controlling KCT damage, 50 fruit from the central tree of each plot were scored for KCT damage using the Scurfing Severity Rating (Table 4) at harvest.

Statistical analysis

Statistix was used to analyse the data. The effect of treatments on the KCT population was analysed with ANOVA, with differences tested for significance using the t-table at the probability level of P=0.05. In some cases the data were log (x + 0.5) transformed for analysis of the effect of the treatments on KCT activity.

RESULTS

Effect of treatments on KCT activity

Trunk applied treatments

Due to the delay in the application of foliar treatments, foliar and trunk application of treatments must be analysed separately.

There was no significant treatment affect on the densities of adult or larvae 7, 22 or 36 DAT.

Foliar applied treatments

By the time the foliar treatments were applied (27/11/02) the KCT larval density had plummeted (Fig. 4). As a result none of the post-treatment differences between treatments were significant.

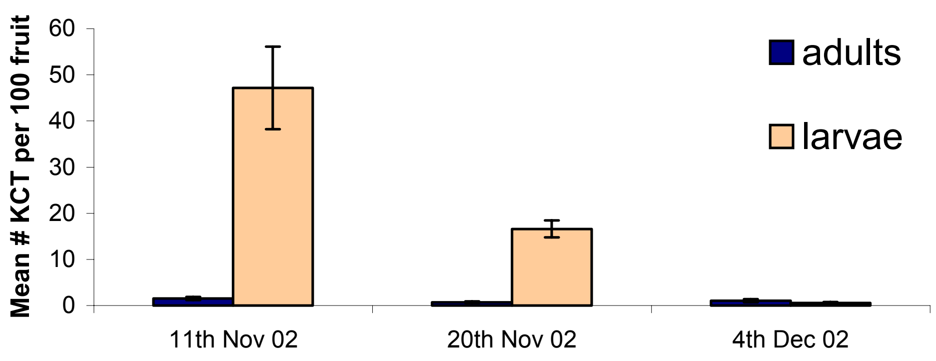


Fig. 4. Mean # KCT per 100 fruit at Sunlands site, Riverland, 2002.

Effect of treatments on the incidence of halo fruit damage resulting from KCT feeding

Damage levels were too low in all treatments (including control) to enable any statistical analysis (Table 11).

Table 11. Percentage of fruit in each of four categories of KCT scurfing damage severity, after treatment applications in November, harvest June 2003, Sunlands, Riverland.

Treatment	**Scurfing Severity Score***			
	0	**1**	**2**	**3**
A	97	2	1	0
B	98	2	0	0
C	93.5	4	2.5	0
D	93	4	1	2
E	97	1	2	0
F	94.5	2	1	2.5
G	93.5	2	2.5	2
H	98	1	1	0
I	93	1	1	5
J	98	2	0	0

*Refer to Table 4 for Scurfing Severity Ratings.

DISCUSSION

Little new information about the efficacy of different modes and rates of thiamethoxam was obtained from this trial, largely due to the regrettable delay in foliar treatment application that was beyond the control of the project team. However, earlier project data is anticipated by Syngenta to be sufficient for registration of the product Actara.

2002-03 SUCCESS™ RIVERLAND TRIAL

INTRODUCTION

Spinosad (Success™) as a foliar spray has provided poor to mediocre control of KCT in past field trials. Despite this, we maintained an interest in this insecticide because it is relatively "soft" on many natural enemies. As part of a laboratory study within the LBAM project (CT01023) it was demonstrated that the performance of spinosad could be significantly enhanced when used in combination with the oil Brella™. Brella™ is a medicinal paraffinic oil produced by Caltex.

METHODS

Site and Treatment details

This trial was conducted in a commercial navel orchard at Loxton North. Treatments (Table 12) were replicated 4 times in a randomised block design. Each plot consisted of 9 trees (3 x 3), of which only the central tree was monitored for KCT activity and damage.

The treatments were applied using a commercial turbine fan (Tornado) sprayer at 5000L/ha. The first application occurred on 10/11/02 at which time the KCT larval density had exceeded 5% incidence. A follow-up spray was applied two weeks later on 27/11/02.

Table 12. Treatments applied in insecticide trial, Loxton North, Riverland, 2002-03.

Treatment	Insecticide Product	Active ingredient	Product Rate per 100L water
A	Unsprayed control	-	-
B	Lorsban	500g ai/L chlorpyrifos	100mL
C	Success + Caltex Brella™	120 g ai/L spinosad	40mL plus 0.5% Brella™ oil

Assessment of KCT activity and damage

To determine the timing of treatment application (>5% KCT larvae incidence), one hundred fruit were sampled, at random throughout the block, into jars of 80% alcohol for processing in the laboratory using a microscope.

To determine the effect of treatments on KCT, 100 fruit per plot were collected 5 days after the first application and 50 fruit per plot 6 days after the second application of treatments. Fruit were collected directly into alcohol for processing in the laboratory.

To determine the efficacy of treatments in controlling KCT damage, 60 fruit from the central tree of each plot were scored for KCT damage using the Scurfing Severity Rating (Table 4) at harvest (2/6/03).

Statistical analysis

Statistix was used to analyse the data. The effect of treatments on the KCT population was analysed with ANOVA, with differences tested for significance using the t-table at the probability level of P=0.05. Data were log(x+0.5) transformed for analysis of the effect of the treatment on KCT activity. The effect of treatments on the percentage of fruit damaged by KCT was also analysed with ANOVA using an arcsin transformation, with differences tested for significance using the t-table at the probability level of P=0.05.

RESULTS

Effect of treatments on KCT activity

KCT adult and larval densities in treated plots were significantly reduced 5 days after the first application of insecticides (10/11/02). Spinosad + Brella™ oil caused a significant reduction in KCT adult and larval densities, while chlorpyrifos significantly reduced KCT larval densities only (Table 13).

KCT larval densities were again significantly reduced 6 days after the second application of Spinosad + Brella™ oil (Table 14).

Table 13. Log transformed mean incidence of adult and larval KCT per 100 fruit assessed 5 days (15/11/02) after a single application of the treatments (10/11/02) at Loxton North, Riverland. Means in the same row followed by the same letter are not significantly different (*P*=0.05).

Treatment		Unsprayed control	Chlorpyrifos	Spinosad + Brella™ oil	*LSD*	*Fprob*
Log transformed mean	Adults	1.30a	1.26a	0.98b	*0.21*	*0.02*
	Larvae	2.01a	1.08b	0.77b	*0.54*	*0.003*
Un-transformed mean	Adults	21.75	17.75	8.75		
	Larvae	115.75	16.25	5.00		

Table 14. Log transformed mean incidence of KCT larvae per 50 fruit assessed 6 days (3/12/02) after a second application of treatments (27/11/02) at Loxton North, Riverland. Means in the same row followed by the same letter are not significantly different (*P*=0.05).

Treatment	Unsprayed control	Chlorpyrifos	Spinosad + Brella™ oil	*LSD*	*Fprob*
Log transformed mean	0.75a	0.66a	0.15b	*0.46*	*0.0388*
Un-transformed mean	6	4	0.5		

Effect of treatments on the incidence of halo fruit damage resulting from KCT feeding

KCT damage incidence was significantly reduced in plots treated with chlorpyrifos and spinosad + Brella™ compared to plots that were left untreated (Table 15).

Table 15. Percentage of fruit damaged (Severity Rating 2-3*) by KCT, after 2 treatment applications in November, harvest June 2003, Loxton North, Riverland. Means in the same row followed by the same letter are not significantly different (*P*=0.05).

Treatment	**Unsprayed control**	**Chlorpyrifos**	**Spinosad + Brella™ oil**	***LSD***	***Fprob***
Arcsin transformed percentage	30.06**a**	14.08**b**	8.49**b**	*10.88*	*0.007*
Un-transformed percentage	25	6	3		

*Refer to Table 4 for Scurfing Severity Ratings.

DISCUSSION

The significant control of KCT halo damage recorded in the 2002-03 trial is the best result we have achieved with a spinosad treatment when compared with chlorpyrifos. Although our resources and the size of the block didn't allow the inclusion of a spinosad treatment alone, this superior result suggests that the performance of spinosad may have been significantly enhanced when combined with the Caltex oil Brella™.

Based on the results of this trial, a field trial to test a number of rate combinations of spinosad + Brella™ will be conducted in 2003-04.

2003-04 SPINOSAD+BRELLA™ TRIAL

INTRODUCTION

The results of the 2002-03 field trial suggested that field control of KCT with spinosad might be improved when applied with Brella™. Laboratory bioassays were conducted in September 2003 to compare the efficacy of weathered residues of a number of rate combinations of spinosad-Brella™ (applied to run-off on citrus foliage in the Waite orchard). The most cost-effective combinations were then trialled in a commercial navel orchard at Golden Heights, Riverland.

METHODS

Site and treatment details

Treatments were replicated 4 times in a randomised block design. Each plot consisted of 6 trees (3 x 2), of which only the inner side of the two middle trees was monitored for KCT activity and damage.

Sprays were applied using a commercial oscillating boom sprayer. Treatments (Table 16) were applied once KCT larval density had exceeded 5% incidence (25/11/02).

Table 16. Treatments applied in insecticide trial, Golden Heights, Riverland, 2003-04.

Treatment	Insecticide	Active ingredient	Rate	Volume (L/ha)
A	Unsprayed control	-	-	
B	Supracide™	400g ai/L methidathion	125mL/100L	6000
C	Success™ + Brella™	120 g ai/L spinosad + oil	20mL/100L plus 0.5% oil	6000
D	Success™ + Brella™	120 g ai/L spinosad + oil	20mL/100L plus 0.5% oil	3000
E	Success™	120 g ai/L spinosad	40mL/100L	6000
F	Success™ + Brella™	120 g ai/L spinosad + oil	40mL/100L plus 0.5% oil	6000
G	Success™ + Brella™	120 g ai/L spinosad + oil	40mL/100L plus 0.5% oil	3000

Assessment of KCT activity and damage

To determine the timing of treatment application (>5% KCT larvae incidence), one hundred fruit were sampled, at random throughout the block, into jars of 80% alcohol for processing in the laboratory using a microscope.

To determine the effect of treatments on KCT, 25 fruit per plot were collected 6 days after treatment. Fruit were collected directly into alcohol for processing in the laboratory.

To determine the efficacy of treatments in controlling KCT damage, 25 fruit from the central tree of each plot were scored for KCT damage using the Scurfing Severity Rating (Table 4) on 7th May 2004

Statistical analysis

Statistix was used to analyse the data. The effect of treatments on the KCT population was analysed with ANOVA. Data were square-root transformed, with differences tested for significance using the t-table at the probability level of *P*=0.05. Harvest data was not transformed.

RESULTS

Effect of treatments on KCT incidence

Due to the extremely low pest pressure at the trial site, a rigorous test of treatment efficacy was not possible. However, in the methidathion and three of the spinosad + Brella™ treatments (D, E & F) no KCT larvae were found on the sampled fruit (Table 17).

A total of only 4 adults were recorded from the 7 treatments (700 fruit).

Table 17. The mean density of KCT larvae per 100 fruit, six days after application, Golden Heights, Riverland 2003. Means followed by the same letter are not significantly different using the LSD test P=0.05).

Treatment	Insecticide	Rate	Volume (L/ha)	Mean density of KCT larvae
A	Unsprayed control	-	-	1.5a
B	Supracide	125mL/100L	6000	0a
C	Success + Brella™	20mL/100L plus 0.5% oil	6000	0.75a
D	Success + Brella™	20mL/100L plus 0.5% oil	3000	0a
E	Success	40mL/100L	6000	0a
F	Success + Brella™	40mL/100L plus 0.5% oil	6000	0a
G	Success + Brella™	40mL/100L plus 0.5% oil	3000	0.25a

Effect of treatments on the incidence of halo fruit damage resulting from KCT feeding

Damage levels were too low in all treatments (including control) to enable any statistical analysis (Table 18).

Table 18. Percentage of fruit in each of four categories of KCT scurfing damage severity*, after treatment applications in November, harvest June 2003, Golden Heights, Riverland.

Treatment	Scurfing Severity			
	0	**1**	**2**	**3**
A	95	4	1	0
B	99	0	1	0
C	99	1	0	0
D	96	4	0	0
E	98	1	1	0
F	98	2	0	0
G	99	1	0	0

*Refer to Table 4 for Scurfing Severity Ratings.

DISCUSSION

Extremely low KCT pressure at the time of this trial hampered any rigorous testing of Spinosad + Brella™ rate and volume treatments. However, along with methidathion, three of these treatments managed to keep KCT levels to zero.

Previous field trials have demonstrated that spinosad, particularly when combined with Brella™, is an effective insecticide for the control of KCT. Laboratory trials have shown that the efficacy of certain insecticides is increased up to 10-fold when used in combination with Brella™. It is unfortunate that we were unable to rigorously field-test the efficacy of reduced rates and volumes of the spinosad-Brella™ mix. However laboratory bioassays have been conducted to compare the toxicity on adult KCT of spinosad, abamectin and emamectin benzoate alone and in combination with Brella™; this lab study is reported below.

We anticipate that DowAgro Sciences, the producers of spinosad, have gained enough information from previous trial work to pursue the registration of spinosad for use in Australian citrus for the control of KCT at the rate recommended for use in other crops (40ml/100L).

2003 LAB STUDY OF THE SYNERGISTIC EFFECT OF USING BRELLA™ WITH THE INSECTICIDES SUCCESS™, PROCLAIM™ AND VERTIMEC™

INTRODUCTION

There is increasing evidence that the efficacy of Success™ (spinosad) against various pests is improved when this insecticide is applied with a horticultural spray oil (Paul Downard, Dow Agrosciences, *pers. comm.*.). The 2002-03 Riverland trial with Success™ reported earlier in this Section provided further evidence, indicating that the performance of spinosad may have been significantly enhanced when combined with the Caltex oil Brella™.

Proclaim™ (emamectin benzoate) was field trialled for KCT control in 1999 as part of Project CT97007, where it provided a level of control that was comparable to that of chlorpyrifos. Vertimec™ (abamectin) was included in the same trial, but the control of KCT that was achieved was poor. However, abamectin is being applied with horticultural spray oil in New Zealand for KCT control (Dr David Steven, *pers. comm.*).

This laboratory bioassay was undertaken to determine whether a synergistic benefit is obtained controlling KCT adults by applying these three insecticides in combination with several rates of Brella™.

METHODS

The bioassay methodology employed was the same full-dose response, Potter tower technique described in the Section titled "Resistance to chlorpyrifos, methidathion and methomyl in KCT populations" in this Final Report. The KCT strain tested was the 'OP-susceptible' Waite strain. The insecticides used were Success™ (120 g spinosad L^{-1}), Vertimec™ (18 g abamectin L^{-1}), and Proclaim™ (44 g emamectin benzoate L^{-1}).

RESULTS AND DISCUSSION

The results are presented in Table 19. They confirm that the addition of Brella™ to a spray of these insecticides can improve their insecticidal activity against KCT. Examination of the LC99 values reveals that the inclusion of Brella™ at the 1.0% rate has allowed 99% KCT mortality to be achieved with approximately 30%, 5% and 0.5% of the amount of spinosad, abamectin and emamectin benzoate required to achieve this mortality when these insecticides are used alone.

In Table 20 the ratios of the field use rate to the LC99 for these insecticides, alone and in combination with Brella™, are presented. To be certain of achieving good field control many entomologists and chemical companies consider that this field rate:LC99 ratio should be in the 30 to 40:1 range or greater. Referral to Table 20 reveals that this ratio is 'suboptimal' for spinosad alone, but the inclusion of Brella™ at both the 0.5% and 1.0% rates lifts this ratio into the range where good field control may be expected. By contrast, this ratio for abamectin and emamectin benzoate alone is two orders of magnitude below the desirable 30 to 40:1 range. The 7.5:1 ratio for abamectin+1.0% Brella™ suggests that, even with the enhanced efficacy that results from the inclusion of this rate of Brella™, abamectin is likely to be ineffective for field control of KCT. However the use of emamectin benzoate with 1.0% Brella™ resulted in a field rate:LC99 ratio of 25.9:1, which may mean that this emamectin benzoate-Brella™ treatment would provide effective field control of KCT.

Table 19. Dose-response data of Success™, Vertimec™ and Proclaim™ used alone and in combination with 0.5% and 1.0% Brella against adult 'susceptible' KCT.

Treatment (g or ml a.i. L^{-1})	n[1]	Slope (±s.e. [2])	LC50 (g ai L^{-1}) (95% F.L. [3])	LC99 (g ai L^{-1}) (95% F.L.)
0.048 g spinosad	273	3.49 (0.39)	0.00059 (0.00051 – 0.00069)	0.0027 (0.002 – 0.0043)
0.048 g spinosad + 5 ml Brella™	277	2.79 (0.66)	0.00015 (0.00010 – 0.00020)	0.0010 (0.00052 – 0.0083)
0.048 g spinosad + 10 ml Brella™	327	3.09 (0.39)	0.00014 (0.00012 – 0.00016)	0.00081 (0.00057 – 0.0014)
0.0045 g abamectin	350	2.99 (0.60)	0.0019 (0.0013 – 0.0027)	0.011 (0.0062 – 0.048)
0.0045 g abamectin + 5 ml Brella™	305	2.95 (0.48)	0.00016 (0.00013 – 0.00020)	0.0010 (0.00066 – 0.0022)
0.0045 g abamectin + 10 ml Brella™	386	2.89 (0.43)	0.000093 (0.000075 – 0.00011)	0.00060 (0.00040 – 0.0012)
0.0132 g ema. benzoate	430	2.23 (0.23)	0.0090 (0.0075 – 0.011)	0.099 (0.065 – 0.18)
0.0132 g ema. benzoate + 10 ml Brella™	303	2.45 (0.34)	0.000057 (0.000045 – 0.00007)	0.00051 (0.00032 – 0.0011)

[1] Number tested [2] Standard Error [3] 95% Fiducial Limits

Table 20. The insecticide field use rate:LC99 ratio for Success™, Vertimec™ and Proclaim™, used alone and in combination with 0.5% and 1.0% Brella against adult 'susceptible' KCT.

Brella treatment	Insecticide field use rate:LC99		
	Success™	Vertimec™	Proclaim™
Insecticide alone	17.8	0.4	0.13
Insecticide + 0.5% Brella™	48	4.5	-
Insecticide + 1.0% Brella™	59.3	7.5	25.9

In conclusion, although the inclusion of Brella™ has had a more dramatic impact on the performance of abamectin and emamectin benzoate than it has had on the performance of spinosad, these two macrocylic lactone compounds would appear to still remain ineffectual for KCT field control even if used with Brella™. By contrast, the performance of spinosad at the field rate of 0.048 g ai L^{-1} (40 ml $100L^{-1}$ of Success™) is likely to be significantly enhanced by the inclusion of 0.5% to 1.0% Brella™.

Two research areas need further investigation:

1. to confirm the field benefit of using Brella™ with Success™ and determine the field efficacy of several different Brella™-Success™ rate and spray volume ha combinations, and
2. to determine whether the detrimental impact of Success™ on important citrus parasitoid wasps is increased by the inclusion of Brella™.

SUMMARY OF FOLIAR INSECTICIDE TRIALS 2000-04

Chlorpyrifos and methidathion are organophosphate insecticides widely used by citrus growers in Sunraysia and the Riverland for KCT control. There is documented evidence that some KCT populations are developing resistance to organophosphates (see Section "Resistance to Chlorpyrifos, Methidathion and Methomyl in KCT Populations" in this report), and it is therefore essential that Riverland and Sunraysia growers soon have one or more alternative insecticide groups to use in an insecticide resistance management rotation strategy to help delay the further evolution of resistance to insecticides.

A low-rate (0.75%), multiple oil spray strategy tested in a small-scale, unreplicated trial in 2000-01 failed to prevent KCT scurfing damage.

The results of work conducted within this Project have led to Syngenta fast tracking the registration of their product Actara™ (thiamethoxam) for use against KCT in Australian citrus. It is anticipated that Actara™ will be available to growers by 2005-06.

It should be noted that Actara™ has good field persistence. Whilst this attribute contributes to its effectiveness at controlling KCT, it has the negative effect of prolonging the considerable toxicity that the foliar spray formulation has against beneficial wasp parasitoids. Given the importance of beneficial wasps to citrus IPM, foliar sprays of Actara™ should be used cautiously, and only in circumstances of high KCT pest pressure. In recognition of this issue, Syngenta are testing trunk and soil-applied formulations of thiamethoxam to avoid or limit this off-target IPM risk.

It is recommended that Dow AgroSciences pursue registration of Success™ (spinosad) for control of KCT in Australian citrus. Success™, especially when combined with the Caltex oil Brella™, can provide effective control of low to medium density KCT infestations. Combined with its "soft" effect on beneficial insects, it could fill a valuable role within an integrated management system for KCT.

Kaolin has been used to successfully control Scirtothrips citri in California and Arizona by applying from petal fall up to five applications, at approximately 20-day intervals (Kerns and Wright 2000). A Kaolin (Surround™) foliar spray trail was designed and initiated at Loxton in October 2003. However the collaborator, who was responsible for applying the specified three applications of Surround™ at 10-day intervals from petal fall, failed to spray the second and third applications, and hence the trail was abandoned. It is worth noting that Californian researchers have recently reported red-scale flare-ups in Kaolin-treated citrus, and are warning against the use of Kaolin treatments for pest control (Grafton-Cardwell and Reagon 2003).

It is essential for the long-term maintenance of the efficacy of these new KCT insecticides that an insecticide resistance management (IRM) strategy be devised. A "window" rotation IRM strategy, such as employed by the Australian cotton and Brassica vegetable industries to combat the development of resistance in cotton bollworm and diamondback moth respectively, is recommended (Roush 1989, Immaraju *et al.* 1990). Adoption of an effective "window" strategy can be expected to delay the onset of resistance and thereby increase the effective life of the insecticides in the strategy by as much as 2-2.5 times (Roush 1998).

Would a "window" strategy need to be universally adopted across the Riverland-Sunraysia to be effective? KCT populations in Riverland-Sunraysia citrus appear to be relatively sedentary (see Section "The Dispersal and Primary Population Sources of KCT in the Riverland-Sunraysia" in this report), with little or nil gene flow likely to occur between separate settlements. Therefore, the geographic unit for an effective IRM "window" strategy could be reduced to the scale of separate settlements. However a standardized strategy than can be readily promoted across the whole industry does have practical benefits.

Would a "window" strategy be effective if many citrus growers within a settlement were acting independently and using products in different windows from each other, resulting in any one insecticide being used across all windows within the settlement? The answer is an unequivocal no. Sufficient KCT population gene flow is likely to be occurring within settlements to render such a laissez faire strategy ineffective.

What would be the most appropriate window time-scale for rotating the use of the different KCT insecticides? Given that the main control period is short (November-December) and probably involves only two KCT generations, the appropriate rotation would be from year to year. For example, let us assume that five insecticides are available to the industry, three of which confer cross-resistance between each other (chlorpyrifos, methidathion and methomyl), and two others (thiamethoxam and spinosad) which show no cross resistance between each other or to the group of three. An example of a three-year "window" strategy for this choice of KCT insecticides would be:
Year 1 – Thiamethoxam
Year 2 – Chlorpyrifos, Methidathion or Methomyl
Year 3 – Spinosad
Year 4 – Thiamethoxam
Etc.

There are several likely grower/chemical company concerns about a yearly "window" strategy. Firstly, if these insecticides differ significantly in price, KCT efficacy, or impact on beneficials, there will be a tendency amongst many growers to depart from the strategy in particular years in favour a cheaper, or more effective, or less IPM-disruptive option. Secondly, the chemical companies are unlikely to support a strategy in which no sales of particular products are made for the control of the citrus industry's main pest in the Riverland-Sunraysia for two whole years.

Can these concerns be addressed? The issue of price will likely diminish over time as the price of the new products decline relative to the older product prices, but in the short-term will be difficult to address. Efficacy concerns can be allayed by properly informing growers. The IPM-disruption concerns are difficult to address, but concerned growers must be advised that over-reliance on one product (this would most likely be spinosad) is ill advised for IRM reasons (particularly given that a number of international pests have developed resistance to spinosad in only 3-4 years of selection). Finally, the chemical company concerns could certainly be addressed. For example, for a **three**-year "window" strategy as described above, the Riverland-Sunraysia could be divided into **three** geographically related groups of settlements, each of which have similar citrus acreage. The strategy could then be applied to each of these IRM "window" areas in a yearly offset pattern, which would mean that in any one year all of the products were being sold for KCT control across the entire Riverland-Sunraysia, but within any one of these three IRM "window" areas only the one product (or related group of old products) would be sold for KCT control.

LITERATURE CITED

Grafton-Cardwell, B. and Reagon, C. 2003. Surround use in citrus increases California Red Scale. *UC Plant Protection Quarterly* Vol. 13 (3). pg.5-8.

Kerns, D.L. and Wright, G.C. 2000. Protective and yield enhancement qualities of kaolin on lemons. In Wright, G. and M. Kilby (eds.). 2000 Citrus and Deciduous Fruit and Nut Research Report.

Liu, Z.M., Beattie, G.A.C., and R. Spooner-Hart 2002. Feeding and oviposition responses of greenhouse thrips to horticultural mineral oil deposits on Valencia orange fruit and mango leaves. In Beattie, G.A.C., Watson, D.M., Stevens, M.L., Rae, D.J. and R. N. Spooner-Hart (eds.). Spray Oils Beyond 2000: Sustainable Pest And Disease Management. Proceedings of a conference held 25-29 October, 1999, Sydney, NSW, Australia, pp: 147-151.

Immaraju, J. A., Morse, J. G., and Hobza, R.F.. 1990. Field evaluation of insecticide rotation and mixtures as strategies for citrus thrips (Thysanoptera: Thripidae) resistance management in California. *Journal of Economic Entomology* **83**: 306-14.

Roush, R.T.. 1989. Designing resistance management programs: How can you choose? *Pesticide Science* **26**: 423-41.

Roush, R.T.. 1998. Resistance management strategy for diamondback moth in southern Australia. Report to AVCARE Australian Insecticide Resistance Action Committee, 7 pp.

THE EFFECT ON KCT FECUNDITY OF EXPOSURE TO SUBLETHAL DOSES OF CHLORPYRIFOS

INTRODUCTION

Hormoligosis is the term used to describe the phenomenon whereby exposure of an insect to sublethal doses of a pesticide causes an increase in its fecundity. This phenomenon has been reported with the Californian citrus thrips, Scirtothrips citri (Morse and Zareh 1991). Do chlorpyrifos residues cause hormoligosis in KCT? If so, it may help explain the emergence of this thrips species as a major pest in Riverland-Sunraysia citrus in early 1990's, which took place four to five years after a dramatic increase in chlorpyrifos usage had occurred across this citrus region in response to changing market conditions and the arrival of several new pests.

This laboratory study was undertaken to determine whether the fecundity of female KCT was influenced by exposure to sublethal doses of chlorpyrifos.

METHODS

Two to three day old female KCT were taken from a laboratory culture which was maintained as described in Baker *et al*. 2000. Approximately 30 females were transferred onto 30 mm diameter leaf discs embedded in agar in 35 mm diameter plastic petri dishes, anaesthetized with carbon dioxide, and then sprayed in a Potter Tower with one of five treatments. The treatments were the LC05, LC10, LC25 and LC50 of chlorpyrifos (ie. the concentration that kills 5, 10, 25 and 50% of the KCT females) and a water control. Each petri dish was then covered with cling-film plastic that was peppered with micro-pin perforations to allow air exchange and prevent condensation build-up. After 24 hours the survivors were transferred to containers specially designed for the laying and harvesting of KCT eggs. 25 KCT were placed in each of these containers, and six replicates were set up for each of the five treatments.

The egg-laying containers were constructed by cutting the base out of a plastic 50 ml sample jar, and then stretching a piece of parafilm over the opening. The thrips were then introduced using the screw-cap lid, and the container inverted so that the screw-cap lid became the base of the container. A small quantity of cumbungi pollen was previously placed on the inside surface of the lid as a food source for the KCT. The base of a 35 mm plastic petri dish was then filled with RO water, and the parafilm end of the container placed over this dish, and then both the dish and container were inverted. This allows the thrips to lay their eggs through the parafilm into the water reservoir formed above. Every 2-3 days the eggs were harvested by pouring the water reservoir through a Buchner funnel (additional water was used to rinse the parafilm and petri dish to ensure all the eggs were collected). They were then counted by examining the Buchner filter paper under a stereomicroscope. Egg production was assessed over a total of 16 days.

The data was log-transformed to correct for non-additivity and analysed by ANOVA using Statistix 8.

RESULTS AND DISCUSSION

Although the mean number of eggs produced per female ranged almost four-fold between treatments (2.26 to 8.42), these differences were not statistically significant (Table 1).

Table 1. The mean number of eggs produced per KCT female over 16 days following exposure to four sublethal concentrations of chlorpyrifos.

Treatment	Mean number of eggs per female KCT	
	Log-transformed (Fprob=0.053)	Untransformed
Control (water)	0.6995 a	5.03
LC05 chlorpyrifos	0.4017 a	2.26
LC10 chlorpyrifos	0.6947 a	6.02
LC25 chlorpyrifos	0.8388 a	8.42
LC50 chlorpyrifos	0.4577 a	2.86

It is worth noting that the numbers of eggs produced per female in this experiment were relatively low, and that there was considerable variability between the replicates within each treatment. It is conceivable that the low egg productivity may have helped to mask any treatment effect. Nevertheless, the mean productivity of the control females (5.03 eggs per female) was marginally higher than the mean productivity of females exposed to the chlorpyrifos treatments (4.92 eggs per female), which provides no support for a hormoligosis effect.

On this evidence it would seem unlikely that the emergence of KCT as a Riverland-Sunraysia pest is due to insecticide-induced hormoligosis.

LITERATURE CITED

Morse, J.G., and N. Zareh. 1991. Pesticide-induced hormoligosis of citrus thrips (Thysanoptera: Thripidae) fecundity. *J. Economic Entomology* **84**: 1169-1174.

THE EFFECT OF FOLIAR RESIDUES OF CHLORPYRIFOS, THIAMETHOXAM AND SPINOSAD SPRAYS ON ADULTS OF *APHYTIS MELINUS*

INTRODUCTION

Following the promising results obtained in the 2000-01 and 2001-02 field trials controlling KCT with foliar formulations of spinosad (Success™) and thiamethoxam (Actara™), an experiment was conducted in April 2002 at the Waite Campus to determine the contact toxicity to *Aphytis melinus* (the key parasitoid of red scale) of field-weathered residues of these insecticides compared to those of the industry standard, chlorpyrifos.

METHODS

40 ml Success™ (120 g ai/L spinosad), 30 g Actara™ (250 g ai/kg thiamethoxam) and 100 ml Lorsban™ (500 g ai/L chlorpyrifos) per 100 L water respectively and a water control were each applied to runoff to sections of foliage on lemon trees in the Waite orchard. 1.5% yellow fluorescent dye was added to each of the spray solutions so that the treated leaves could be viewed under black light and leaf-discs with a similar spray deposition selected for the bioassay.

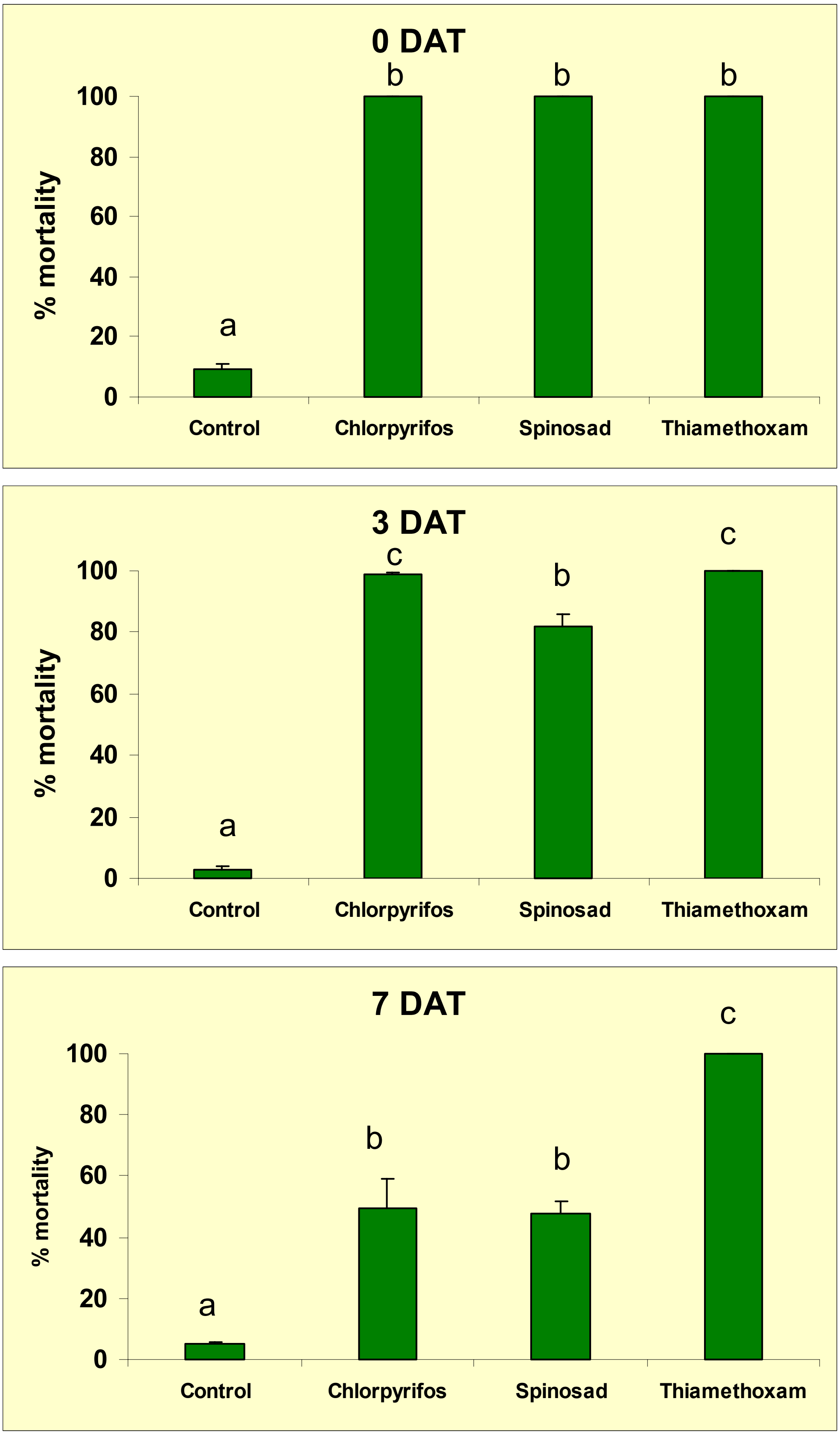
At 0, 3, 7 and 13 DAT 29 mm diameter leaf discs with similar spray coverage were cut from selected leaves (10 discs per treatment) and embedded in setting agar in 30 mm diameter petri dishes. Approximately 20 *A. melinus* adults were introduced to each petri-dish arena. The dishes were each covered with cling-film plastic that was peppered with micro-pin perforations to allow air exchange and prevent condensation build-up. The mortality of the *A. melinus* was assessed after 24 hours.

The percentage mortality data were arcsin square-root transformed and analysed by ANOVA using Statistix 8.

RESULTS AND DISCUSSION

The results are presented in Fig. 1. The 24-hour exposure to residues of each the three insecticides on leaf discs collected four hours after the spray application (0 DAT) resulted in 100 percent mortality of the tested adult *A. melinus*. At 3 DAT the percentage of *A. melinus* killed by exposure to the Success™-treated foliage had declined to 81.8%, but 100% and 98.8% of *A. melinus* were still killed by exposure to the Actara™ and Lorsban™-treated foliage respectively. At 7 DAT the mortality of *A. melinus* exposed to the Success™ and Lorsban™-treated foliage had dropped to 47.8 and 49.7% respectively, but exposure to the Actara™ residues still killed 99.8% of the tested wasps. At 13 DAT the *A. melinus* mortality from exposure to the Actara™ residues had declined to 64.7%, and the mortality from exposure to the Success™ and Lorsban™ residues had dropped to 16.1 and 30.3%.

These results suggest that, in the first 24-48 hours following spray application with any one of these three insecticides, contact with the treated citrus will kill near to 100% of *A. melinus* adults. The rate of decline in residual toxicity is relatively similar for Success™ and Lorsban™, with mortality declining to about 50% in 7 days. By contrast, Actara™ residues appear likely to kill nearly 100% of exposed *A. melinus* for at least one week post-treatment. Only at the fourth Actara™ assay, conducted at 13 DAT, did substantial survivorship occur.


0 DAT
% mortality
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Chlorpyrifos
Spinosad
Thiamethoxam
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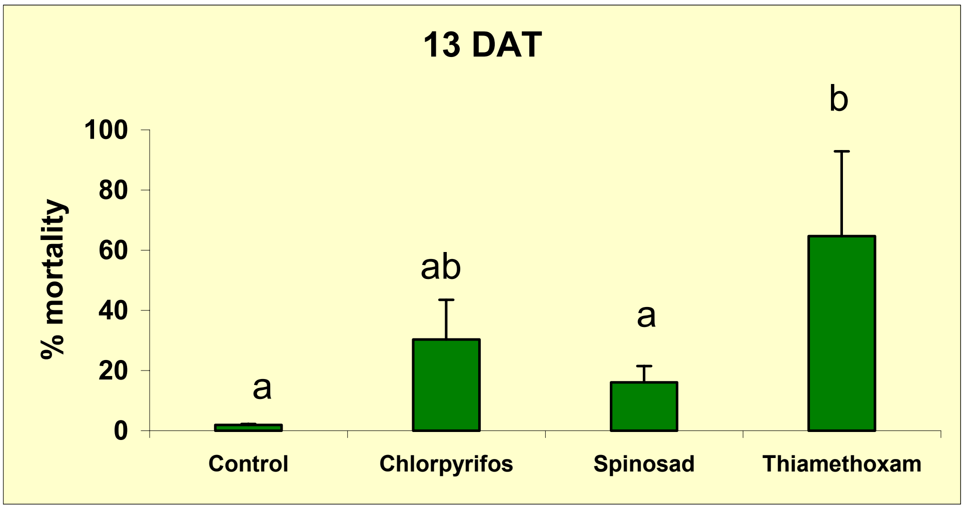


Fig. 1. The mean percentage mortality (bars indicate the standard error of adult *Aphytis melinus* after being exposed for 24 hrs to field-weathered foliar residues (0, 3, 7 and 13 DAT) of chlorpyrifos, spinosad and thiamethoxam. For each of the four assessment dates, treatments accompanied by the same letter are not significantly different, Tukey HSD All-Pairwise Comparisons Test test (P>0.05).

This study is limited in scope, having only tested the contact toxicity of these insecticides on the adult stage of one species of aphelinid parasitoid. However, *A. melinus* and a number of other aphelinid species are key parasitoids of Australian citrus scale and mealybug pests, and it is likely that these results would be broadly indicative of the effect of these three compounds on these aphelinids and other related citrus wasp parasitoids.

THE IMPACT OF CHLORPYRIFOS ON PREDATORY MITE POPULATIONS IN RIVERLAND CITRUS ORCHARDS

INTRODUCTION

The 1990's increase in KCT activity coincided with an increase in insecticide usage in the citrus industry. If KCT populations are naturally controlled by beneficials, these events may be causally related through off-target impacts of these insecticide treatments on beneficial populations. The main broad-spectrum insecticide used in inland Australian citrus over the past decade has been the organophosphate (OP) chlorpyrifos. The natural enemy group that appear to exert the greatest influence on KCT abundance in the Riverland is predatory mites. In this project three experiments were conducted to determine the impact of chlorpyrifos on arboreal and soil-dwelling predatory mite populations of Riverland citrus.

METHODS

Experiment 1

This trial was initiated on 6 November 2002 in a navel orange orchard at Loxton Research Centre. The treatments consisted of 100 ml Lorsban (500 g ai/L chlorpyrifos) per 100L water and a water control each applied at a volume of 5000L/ha to nine-tree plots (five replicates) using a commercial oscillating boom sprayer. Each plot consisted of a single row of 9 trees, with each treatment row separated by an unsprayed buffer row.

The arboreal predatory mite population was assessed using double-sided sticky tape bands around citrus limbs. Five sticky bands were placed around limbs as close to foliage as practical (average diameter of limbs 1cm) on each of four trees per replicate row (the 2nd, 4th, 6th and 8th tree in the 9 tree row). Pre-treatment bands were placed on the treated and untreated trees for 48 hours prior to treatment application. To address the re-entry prohibition, post-treatment bands were placed on trees immediately prior to treatment application and collected after 48hrs.

The soil-dwelling predatory mite population was assessed by collecting soil scrapes (25cm x 25cm x 1cm + leaf litter) 48 hours prior to treatment application. Soil was then placed in Berlese funnels for 72 hours. 24 hours post-treatment soil scrapes were again collected and placed in funnels for 72 hours. Two samples were collected per replicate.

Experiment 2

Because of concerns that the arboreal and soil-dwelling predatory mite faunas were relatively impoverished at the Loxton Research Centre site, and that as a consequence the results of Experiment 1 may underestimate the demographic impact of chlorpyrifos spraying on these mite populations, the following experiment was initiated in September 2004.

This study was restricted to the soil-dwelling predatory mite population, and was conducted using soil collected from the organic navel orange orchard (Brown's) which was part of the KCT orchard management and biological control study (see Section "The Biological Control of KCT in Riverland Citrus Orchards" in this report). Twelve plastic Decor™ containers (~30 x 20 x 8 cm, and fitted with a sealed lid) were each filled to a depth of 5 cm with soil and litter collected by taking trowel scrapes from the upper 1-2 cm of the soil profile along the drip line of this orchard. Preliminary work had demonstrated that the invertebrate community of detrivores, fungivores and predators in these soil samples could be maintained for at least several months in these containers with occasional fine water spraying to maintain the moisture level.

These 12 containers were transported to the Waite campus, and randomly divided into two groups of six. One group was treated on two occasions (1/10/04 and 2/11/04) with a 0.1% solution of Lorsban™ (500 g ai/L chlorpyrifos) applied at the equivalent rate of 2,000L ha^{-1} using a track sprayer. The other group were sprayed on the same occasions with water at the equivalent rate of 2,000L ha^{-1}. The 2,000L ha^{-1} was chosen because this is the minimum volume of run-off that occurs when the average citrus canopy is sprayed at 4-5,000L ha^{-1} with current types of spray equipment used by the Australian citrus industry. Given that many Riverland orchards are now sprayed at least twice during the late spring to early summer with synthetic pesticides, of which chlorpyrifos is the most common treatment, this two spray chlorpyrifos treatment regime was considered representative of current industry insecticide practice.

Three 150cc soil samples were taken from each of the 12 containers on three occasions (pre-treatment date of 28/9/04, and post-treatment dates of 5/10/04 and 4/11/04) and were each placed in a Berlese funnel for 72 hours for invertebrate extraction.

RESULTS

Experiment 1

Arboreal mites: Species of predatory mites recorded on the sticky bands in this first trial included species of the families Erythreidae, Phytoseiidae, Stigmaeidae and Tydeidae (Table 1). Chlorpyrifos did not appear to have a detrimental impact on the abundance of arboreal predatory mites. The numbers of stigmaeds actually increased post-treatment. This is more likely to be an effect on the degree of stigmaeid activity, perhaps an excitation effect caused by the chlorpyrifos treatment, rather than an effect on the density of these mites. Based on this result, the trial was repeated in April 2003 with modification to assessment procedures.

Soil-dwelling mites: The very low densities of mites species (of the families Tydeidae, Anystidae, Bdellidae and Ascidae) collected in the Berlese samples, pre- and post- treatment, were insufficient to detect any impact of chlorpyrifos on the predatory mites.

Table 7. Mean # of predatory mites recorded on sticky bands placed on citrus limbs pre and post-treatment, November 2002.

Sampling date	Treatment	Erythreidae	Phytoseiidae	Tydeidae	Stigmaeidae
Pre-treatment 5/11/02	Unsprayed control	13.4	2.0	13.2	2.4
	Chlorpyrifos	15.2	0.8	20.6	3.6
Post-treatment 8/11/02	Unsprayed control	12.0	1.4	13.6	3.0
	Chlorpyrifos	12.6	1.2	28.2	20.2

Experiment 2

Prior to the first treatment application, the mean numbers of predators in the containers of soil allocated to the two treatment groups were similar (ANOVA, P=0.17) (Fig. 1). Following the first treatment application, a modest suppression of predatory mite densities in the treated containers occurred relative to the untreated containers. However following the second chlorpyrifos treatment, which took place one month after the first application, a substantial decline in predator densities was observed, and the difference in predator density between the treated and untreated containers was highly significant (ANOVA, P<0.01). Meanwhile, the predator population densities in the control arenas had remained unchanged.

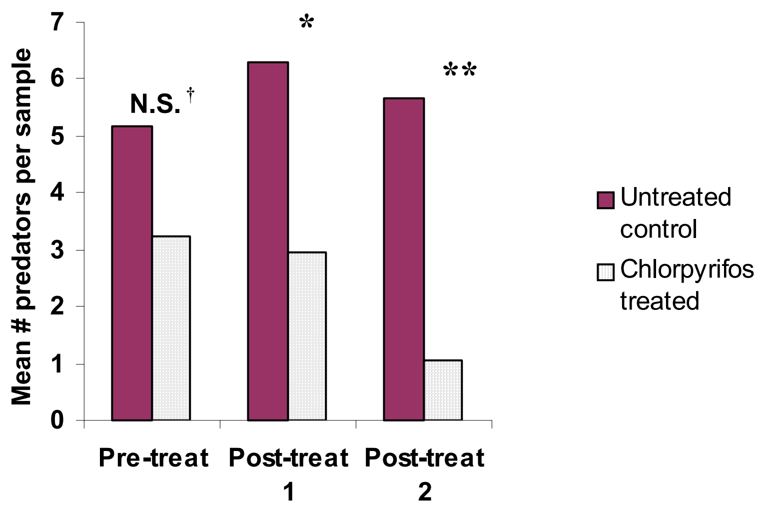


Fig 1. The numbers of predatory mites per Berlese funnel sample in untreated (water) control and chlorpyrifos treated soil at pre-treatment and two post-treatment assessments. ([†]Indicates the significance of the difference between the control and treated; N.S. - P>0.05, * - 0.01<P<0.05, ** - P<0.01)

Both direct and indirect factors may have contributed to the decline in predatory mite abundance that followed the application of the two chlorpyrifos spray treatments. A direct cause may be the toxicity of the chlorpyrifos treatment to the predatory mites, which may have killed active stages of the mites and had sublethal effects on fecundity, etc.. An indirect impact may have been starvation that resulted from the substantial reduction in the abundance of many of the detrivores and fungivores (Fig. 2), which are an important food source for these predatory mites.

As discussed in the "Biological control of KCT in Riverland citrus orchards" section of this report, the predatory mite species *Athiasella relata* (Ologomasidae), *Pachylaelaps* sp. (Pachylaelapidae), and *Protogamasellus mica* and *P. massula* (Ascidae)were notable by their abundance at Brown's organic orchard (the source of the soil samples used in this experiment) and paucity at other Riverland study sites. The authors speculate that these species may be particularly important in regulating KCT at this site. It was therefore of concern to note that no *A. relata*, *P. mica* nor *P. massula* were retrieved from the chlorpyrifos treatment arenas following the second spray application, and that the density of *Pachylaelaps* sp. had declined by 51 percent.

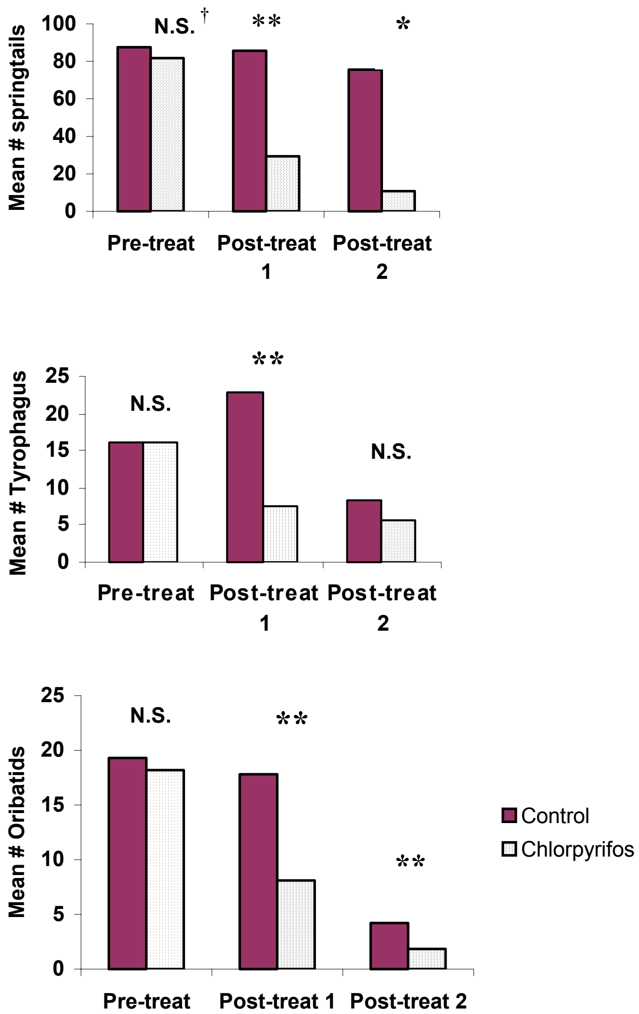


Fig. 2. The numbers of springtails (collembolans), *Tyrophagus* and Oribatid mites per Berlese funnel sample in untreated (water) control and chlorpyrifos treated soil at pre-treatment and two post-treatment assessments.
(†Indicates the significance of the difference between the control and treated; N.S. - $P>0.05$, * - $0.01<P<0.05$, ** - $P<0.01$)

DISCUSSION

Although no substantial impact from foliar spraying of chlorpyrifos was observed in the two Loxton Research Centre trials, it must be noted that the predatory mite population densities at this site were very low and may have masked any treatment effect.

By contrast, the laboratory assay using soil samples collected from Brown's Organic navel orchard, which has an abundant and diverse population of soil-dwelling predatory mites, has revealed a significant detrimental impact from two chlorpyrifos treatments applied one month apart. It is therefore of considerable concern to note that many Riverland citrus orchards are now treated in the spring with two to five chlorpyrifos or methidathion sprays, and that these spray treatments are often applied in closer succession than the one-month interval used in this experiment. Many of these sprays are being applied to target KCT. Ironically, this spring regime of OP sprays may well be releasing KCT populations from the biocontrol regulation that these predatory mite populations are likely providing (See the Section "Biological control of KCT in Riverland citrus orchards" in this report), thereby locking the industry into a classical insecticide treadmill.

The Australian citrus industry currently uses a range of spray technologies, all of which require high spray volumes to achieve effective pest control. In addition to the substantially inflated labour and chemical costs incurred by the industry, this high-volume spray strategy generates high levels of pesticide run-off and hence high pesticide load in the soil (and potentially in ground-water run-off). The findings of this study strongly suggest that chlorpyrifos contamination of orchard soils may be helping to release KCT from effective biocontrol. It would seem appropriate for the Australian citrus industry to critically appraise the potential cost, pest control (and nutrient and growth regulator) efficacy and IPM benefits of multi-fan spray technology (as has occurred, with dramatic benefits realized, in the Australian viticultural industry). In addition to the IPM benefits for KCT management, the improved spray coverage and efficacy that such multi-fan spray technology is likely to provide, would allow 'softer' insecticides, which are less disruptive to natural enemies, to be substituted in many instances for current 'hard' chemistry (eg. organophosphates).

Further KCT predator studies are required:

(1) to determine the resilience of the soil-dwelling predatory mite populations following OP spraying (i.e. how long do they take to recover),
(2) to determine whether the detrimental impact is primarily due to an indirect effect on food sources,
(3) if the answer to (2) is yes, to then determine whether this detrimental impact can be countered by boosting the normal density of the food source detrivore/fungivore populations (eg. through building the organic content of the soils),
(4) to determine the impact of other insecticides (in particular methomyl, thiamethoxam and aldicarb (Temik™)) and key herbicides on these soil-dwelling predatory populations, and
(5) to determine the effect of chlorpyrifos (and ideally other citrus foliar insecticides) on the arboreal predatory mite complex.

EFFICACY OF SOIL-APPLIED CONTACT INSECTICIDES FOR THE CONTROL OF KCT

INTRODUCTION

The primary spring source of immigrant KCT in orange orchards in the Riverland/Sunraysia region appears to be lemon orchards. The strategic application of soil insecticidal treatments in lemon orchards in the late-winter to mid spring may significantly limit the build-up of KCT in flowering orange orchards. Two laboratory experiments (2002) and then two field trials (2003) were designed to assess the effect of two soil-applied contact insecticide treatments on the survival of late 2nd instar larvae and pupae of KCT. The laboratory approach was adopted in 2002 because of the low field abundance of KCT.

METHODS

Experiments 1 (April 2002) and 2 (July 2002)

Sandy soil collected from a citrus block at Loxton Research Centre was placed to a depth of 20 mm in 15 plastic containers of 100 mm diameter and 30 mm height, and cumbungi pollen was lightly sprinkling on top. Thirty late 2nd instar KCT larvae were then added to each container, and the containers covered with Glad-wrap™. Three days were allowed for the larvae to pupate, and then the containers (5 per treatment) were sprayed under a Potter Tower with one of the following treatments: (i) 1.25 µl Talstar™ per 2 ml aliquot of water delivering 0.0025 µl Talstar™ per cm^2 soil surface (equivalent to 100 ml per ha of Talstar™), (ii) 3.125 µl Regent™ per 2 ml aliquot of water delivering 0.001 µl Regent™ per cm^2 soil surface (equivalent to 250 ml per ha of Regent™), and (iii) 2 ml aliquot of water. The containers were then held at 22^{o}C and the numbers of emergent adult KCT counted.

The results were analysed with ANOVA using Statistix 7 software, with differences tested for significance using the Bonferoni test (P=0.05).

High levels of control mortality occurred in Experiment 1. Subsequent testing identified soil moisture as a key determinant of KCT pupal survival, and established that a RH of around 65% was optimal for their survival. The methods employed for Experiment 2 were the same as for Experiment 1, but the soil was sufficiently moistened to provide a RH in the test arenas of approximately 65%.

Experiment 3 (May 2003)

This trial was conducted in a navel orchard on the Loxton Research Centre. Treatments included unsprayed control, fipronil (Regent®) and bifenthin (Talstar®) (Table 1). An Enviromist® sprayer was used to apply treatments to 2m x 2m plots along the drip line of citrus trees. Treatments were replicated 10 times and applied on 7th May 2003.

Table 8. Treatments applied in the Soil Insecticide trial, Loxton Research Centre, Riverland, 7 May 2003.

Treatments	Product	Product Rate	Water Volume
Unsprayed control	-	-	-
Fipronil (200 g a.i. L^{-1})	Regent®	250ml/ha	30L/ha
Bifenthrin (100 g a.i. L^{-1})	Talstar®	100ml/ha	30L/ha

A 15cm x 15cm area near the centre of each plot was seeded with 20-25 late 2nd instar KCT larvae 48hrs prior to treatment application. Large emergence traps (45cm diameter) were placed over the seeded area, removed in order to apply the treatments, and then replaced about one hour after applying the treatments. Fourteen days after treatment the emergence traps were removed and the numbers of adult KCT that had been caught inside each trap was counted.

The results were analysed with ANOVA using Statistix 7 software, with differences tested for significance using LSD (P=0.05).

Experiment 4 (November 2003)
This trial was conducted in a navel orchard at Bookpurnong, Riverland. Treatments included an unsprayed control, fipronil (Regent®) and bifenthin (Talstar®). The insecticides were applied at the same per ha rates as in Experiment 3. A backpack sprayer was used to apply treatments to 2m x 2m plots along the drip line of citrus trees. Treatments were replicated 10 times and applied on 19th November 2003. The spray volume was increased from 30L/ha (May trial) to 200L/ha to provide a more accurate representation of commercial operations (J. Altmann, *pers. comm*.).

The impact of the treatments on KCT survival in the soil was assessed in two ways, firstly by seeding lab-reared KCT larvae into each plot, and secondly by placing a succession of adult emergence traps in each plot pre- and post-treatment to assess the impact on the natural field population.

Plots were seeded with 20-25 late 2nd instar KCT larvae 24hrs prior to treatment application. Emergence traps (45cm diameter) were placed over seeded larvae, removed in order to apply treatments, and then replaced. Given the concern that not enough time was allowed for pupal development before collecting the May emergence traps, this time emergence traps were then left in the field for 27 days before assessments were made.

Emergence traps were also used to determine the residual activity and efficacy of soil insecticides on the existing field population of KCT. One trap per plot was set 7 days prior to treatment application and collected the day before application to provide an indication of population activity. These traps were re-set and serviced at one, eight and sixteen days post treatment.

The results were analysed as for Experiment 3.

RESULTS AND DISCUSSION

Experiment 1
The results of this trial are presented in Table 2. Both the Talstar™ and Regent™ treatments significantly reduced the number of KCT adults that emerged. However, the control mortality of approximately 70% was unacceptably high. Further, this experiment only assessed the effect of these treatments on the adult KCT emerging from the soil. The treatments' effects on mature larvae entering the soil to pupate were not determined.

Table 2. The mean number (and standard error) of KCT adults that emerged from insecticide-treated and untreated soil arenas in Expt. 1, April 2002.

Treatment	Mean±s.e.
Control	9.40±1.33 a[†]
Regent™	4.60±1.75 b
Talstar™	1.20±0.37 b

[†]Means followed by different letters are significantly different at *P*… 0.05 by the Bonferoni test.

Experiment 2

The results of this second laboratory trial were encouraging (Table 3). The control mortality was substantially lower than that of the first experiment, and the numbers of KCT adults that emerged in both the treatments was greatly reduced compared to the control. Based on these results it was concluded that a field test of these treatments was warranted.

Table 3. The mean number (and standard error) of KCT adults that emerged from insecticide-treated and untreated soil arenas in Expt. 2, July 2002.

Treatment	Mean±s.e.
Control	19.10±0.39 a[†]
Regent™	1.20±0.39 b
Talstar™	1.00±0.42 b

[†]Means followed by different letters are significantly different at *P*… 0.05 by the Bonferoni test.

Experiment 3

The treatments had no significant effect on the emergence of KCT seeded into the trial plots two days prior to application (Table 4).

Table 4. Mean incidence of seeded KCT adults emerging from plots 14 days after treatment. Means followed by the same letter are not significantly different (*P*=0.05).

	Unsprayed control	Bifenthrin	Fipronil	*LSD*	*Fprob*
Mean number of adult KCT	0.6a	0.7a	0.2a	*0.85*	*0.439*

No significant treatment effect on the mortality of seeded KCT pupae was demonstrated in this trial. The very low level of adult KCT emergence recorded in the control plots indicates that there may have been problems with our technique of seeding lab-reared pupae into the field environment. This may be due to handling causing damage to the larvae and/or having not allowing enough time for development (the traps were collected after 14 days).
With these factors in mind, the trial was repeated in November 2003.

Experiment 4

The treatments had no significant impact on the natural population of KCT at the trial site (Table 5).

Table 5. Mean number of KCT adults caught on emergence traps pre-treatment (18/11/03) and one (27/11/03), eight (5/12/03) and 16 (15/12/03) days after treatment. Means followed by the same letter are not significantly different (*P*=0.05).

Date	Unsprayed Control	Bifenthrin	Fipronil	*LSD*	*Fprob*
Pre-trt 18/11/03	36a	33a	26a	*14.4*	*0.361*
Post-trt 27/11/03	8a	7a	6a	*3.8*	*0.548*
Post-trt 5/12/03	15a	12a	10a	*4.8*	*0.114*
Post-trt 15/12/03	14a	11a	8a	*7.5*	*0.254*

By contrast, the bifenthrin treatment significantly reduced the emergence of seeded KCT pupae (Table 6). However, the percentage emergence of adult KCT in the controls, although higher than in the May 2003 trial, is again relatively low at approximately 25%. Unfortunately this high control mortality confounds the interpretation of the results. These concerns aside, if the 64% reduction in KCT emergence recorded in the bifenthrin treated plots does reflect the likely impact of such a treatment, it is unlikely to be considered a commercially viable control tactic by most growers.

Table 6. Mean incidence of seeded KCT pupae emerging from plots 27 days after treatment. Means followed by the same letter are not significantly different (*P*=0.05).

Date of Seeding	Unsprayed Control	Bifenthrin	Fipronil	*LSD*	*Fprob*
18/11/03	5.9a	2.1b	6.0a	*2.7*	*0.0115*

DISCUSSION

The strategic application of insecticides to the soil of lemon orchards may be a cost effective and less disruptive (district-wide) control option for KCT. Our research suggests that KCT densities build up in lemon blocks prior to the main flowering period in navel oranges. If KCT densities in lemon orchards can be controlled before they move into neighbouring navel blocks, it is likely there will be less of a reliance on repeated foliar insecticidal. However, we anticipate that any soil treatment used is likely to have detrimental effects on the existing soil fauna. Hence the general use of such a control option would be discouraged, and instead selective and strategic application in particularly troublesome areas would be advised.

The November 2003 trial demonstrated a significant reduction in emergence of KCT seeded into plots treated with bifenthrin. The 64% reduction achieved is unlikely to be commercially acceptable, as industry generally requires pest treatments to achieve 90-95% or greater control. However, the rates that both these insecticides were tested at were extremely low. It is envisaged that if this control tactic were adopted by the industry, only the drip-line zone, where KCT pupae are most concentrated, would be treated. This treated zone would be about 25-33% of the orchard area, which at the rates tested in these trials, would equate to only 2.5-3.3 g.a.i. bifenthrin or 12.5-16.5 g.a.i. fipronil being applied per hectare. These are very low use rates indeed, and further investigation using higher rates of these two products is recommended.

THE DISPERSAL AND PRIMARY POPULATION SOURCES OF KCT IN THE RIVERLAND-SUNRAYSIA REGION

INTRODUCTION

The question of the source of KCT infestations in navel orange orchards in mid-late spring is central to the management of this pest. We now know that KCT populations are present in navel orange orchards all year round, albeit generally at low population densities from mid-summer to early spring. It remains less clear whether these in situ populations are the primary source of the mid-late spring population outbreaks, or whether these in situ populations are supplemented by the influx of KCT from other sources, such as lemon orchards or as a result of a regional immigration from further a field. It should be noted that earlier studies (Baker *et al.* 2000) have established that there are no significant non-citrus hosts of KCT in the Riverland and Sunraysia districts.

Testing KCT populations for their susceptibility to insecticides has provided some circumstantial evidence that localized KCT movement occurs between blocks. KCT collected from an insecticide-free orchard that is surrounded by orchards which are heavily sprayed with chlorpyrifos and methidathion, have been shown to have reduced susceptibility to these organophosphate insecticides, which implies that these KCT have moved from the nearby orchards. This suggests that movement from nearby lemon orchards, which often support high KCT densities because of their more frequent flowering flushes, is feasible.

No data or information was available to determine whether regional migration of KCT occurs.

This study was undertaken using suction traps to measure the relative flight activity of KCT in and outside irrigation settlements in spring.

MATERIALS AND METHODS

The suction traps were constructed using household exhaust fans mounted on top of a support frame, which was then placed over a star dropper. A fine “thrips proof” voile net attached to a collection container was placed below this exhaust fan. Due to the predominantly sandy soils present in the Riverland region, most of the suction trap frames were secured using rope lines pegged around the trap. The operation of the traps required access to mains power.

The collection container consisted of a 500 ml plastic screw-top container. A hole (60 mm diameter) was cut into the lid, and the lid was attached to the fine voile net. Each collection container contained 400 ml of a 25 % NaCl solution with 5 ml of detergent to preserve the trapped insects.

In 2001 a suction trap was placed at each of two sites, one within and one outside the Loxton irrigation district. The former was situated at North Loxton next to a shed in a vineyard approximately 300 m from the nearest citrus. The latter was situated at a residence near Wunkar in the mallee, approximately 30 km from the nearest citrus orchard. These traps were operated from 28th September until 30th November 2001 and were checked every one to two weeks.

In 2002 the trial was expanded to include six traps. Two traps were placed within or adjacent to citrus orchards; one in a Valencia orange orchard at Ingerson’s, Bookpurnong, and the other adjacent to a navel orange orchard at the Loxton Research Centre, Loxton. Two traps were placed within the Loxton irrigation district but distant from citrus orchards; one at North

Loxton in a vineyard approximately 300m from the nearest citrus orchard and the other at the Loxton Research Centre approximately 200m from the nearest citrus orchard. Finally, two traps were placed outside the Loxton irrigation district, one located in the mallee at a residence near Wunkar, approximately 17 km from the nearest citrus orchard, and the other at Calperum station, approximately 5 km from the nearest citrus orchard. These traps were checked every one to two weeks from 16th August until 27th December 2002.

RESULTS & DISCUSSION

In both seasons of these trials no KCT were captured at traps located outside the irrigated settlements. However in both years large numbers of other thrips species were captured in these traps placed in the mallee locations, and KCT were captured at four of the five locations within the irrigated settlements. This result would be expected if there was no significant migration of KCT into Riverland citrus during the spring (Table 1).

Table 1. Expected capture (E) of KCT in traps at three different locations under three different movement/migration scenarios, and the observed capture (O) in the spring 2001 and 2002 suction trapping trials.

Movement/ migration scenario	**Trap location**					
	Within irrigated district				**Outside irrigated district**	
	Within or adjacent to citrus		**≥300m from citrus**		**(> 15 km from citrus)**	
	E	O	E	O	E	O
Large-scale regional movement	Yes	Yes	Yes	Yes	Yes	No
Broad-scale within-district movement, no regional movement	Yes	Yes	Yes	Yes	No	No
Very localized within-district movement, no broad-scale district or regional movement	Yes	Yes	No	Yes	No	No

In the spring of 2001 a total of 15 KCT were captured in the trap located within the irrigated settlement at Loxton North approximately 300 m from the nearest orchard.

In the spring of 2002 the traps placed in or adjacent to citrus orchards did capture KCT, as did the trap located at Loxton North 300 m from the nearest orchard (Fig. 1). However, the trap located at the LRC approximately 200 m from the nearest citrus orchard did not yield any KCT; there were problems with this trap's collection net and container early in the trial, and power supply problems later in the trial. Clearly these results indicate that there is movement of KCT within and near citrus orchards, confirming that some block-to-block movement does occur.

All traps in both the spring of 2001 and 2002 captured a large number of thrips of various species, confirming all traps ability to capture thrips.

In summary, these suction trap trials provide no evidence that a large-scale regional movement of KCT occurs in the Riverland during spring. It is likely that the same would apply for the Sunraysia district. Based on this evidence, and the observation that there are few non-citrus hosts of KCT in the Riverland-Sunraysia region, it seems most probable that the KCT that infest citrus in this region is largely a "self-contained" population cycling within citrus. Whilst movement of KCT between citrus orchards is undoubtedly occurring at the local level, the extent of this population movement and the resultant population gene flows remains unclear.

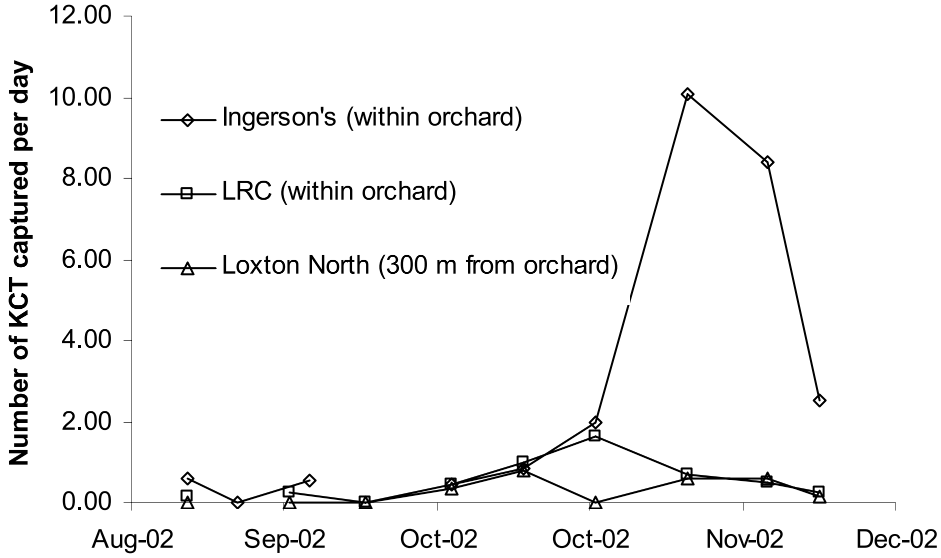


Fig. 1. Number of KCT captured per day per suction trap at three Loxton district locations, spring 2002. (NB. No KCT were captured in traps located at Wunkar or Calperum Station. And because of problems with the trap no KCT were captured at the Loxton Research Centre site.)

ROLE OF COLOUR IN THE ATTRACTION OF ADULT KCT TO FLOWERS

INTRODUCTION

Extensive field observations and collections indicated that Kelly's citrus thrips (KCT) is most commonly collected from flowers that are white or pale yellow in colour. However, KCT are found in abundance in some white flowers (e.g., lemons) but not others (e.g., potato flowers). Insects are known to see colours in the ultraviolet region of the spectrum, and many flowers have ultraviolet reflectance patterns that guide pollinators (Kevan and Baker 1983). So it is conceivable that differences in attraction to flowers with apparently the same colours have different levels of reflectance in the invisible regions of the spectrum that influence that degree of attraction to adult KCT. Moreover, sampling had shown that the most attractive colours among a range of paper cards were either white or light blue. We compared the spectral reflectance of some known host flowers and some flowers that are known not to be inhabited by KCT to see if there are any consistent differences that could account for differing levels of floral infestation.

MATERIALS AND METHODS

The reflectance curves of seven surfaces were recorded, including freshly collected flower petals and leaves (Table 1). Spectral reflectance was measured using a Varian Cary 500 Scan Ultraviolet-Visible-Near Infrared Spectrophotometer that was fitted with a Labsphere DRA-CA-500 reflectance sphere. The light source changeover was programmed to occur at 350 nm. Data were processed with Cary WinUV software (version 02.00 (25)), using zero/baseline correction, a scanning rate of 0.5 nm, 5 nm interpolation. Samples were held on a black card with cellophane tape. A blank with cellophane tape only was scanned as an experimental control. In some instances, two separate curves were recorded for different samples of the same type to check if the curves were representative, or varied from sample to sample.

Table 1. Plant and paper surfaces that were evaluated to measure their spectral reflectance.

Surface	Host plant of KCT	Description
Lemon flower petal	Yes	3 petals
Ornamental mandarin petals	Yes	4 petals
Star jasmine corolla	Yes	Entire corolla
Iceberg rose	No	One petal
Potato vine corolla	No	Entire corolla with anthers removed
Lemon leaf	No	Single fully-expanded leaf.
Blue card	No	Previously shown to be attractive to KCT in the field
White card	No	"

RESULTS

In each of the white flowers, spectral reflectance rose at around 400 nm and peaked at less than 450 nm, above which reflectance was virtually static at the maximum level (Figure 1). Reflectance curves had the same characteristic shape regardless of whether or not the flower was a host of KCT. The lemon leaf had a small reflectance peak at 550 nm and then reflectance rose sharply above 700 nm. Replicated recordings of different petal surfaces were virtually identical in all cases, indicating that the curves were representative of the samples tested.

The two cards displayed substantially different reflectance patterns (Figure 1). The white card reflected light at all wavelengths tested between 300 and 800 nm, with the highest reflectance occurring between 350 and 800 nm. In contrast, the blue card displayed peak reflectance around 450 nm and 800 nm, with lower reflectance between these peaks.

The tape used to mount the specimens did not contribute significantly to the estimated reflectance curves (Figure 1).

DISCUSSION

There are no significant differences in the reflectance (colour) of host and non-host plants, so the reflectance patterns cannot explain differences in attractiveness of various flowers to adult KCT. It is likely that odour is a more significant cue that is used in finding host plants.

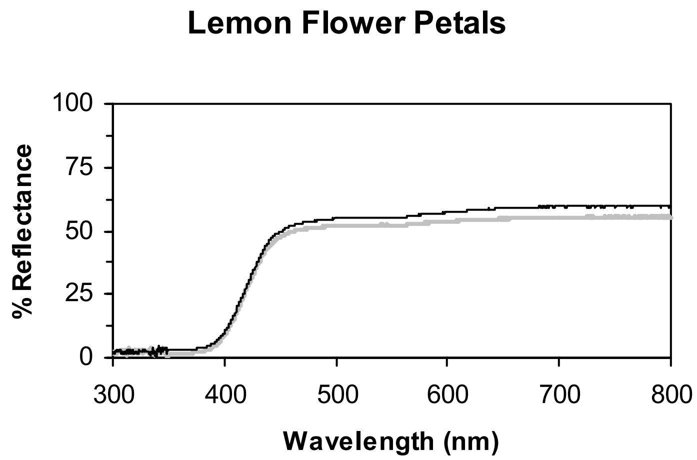
LITERATURE CITED

Kevan, P. G., and H. G. Baker. 1983. Insects as flower visitors and pollinators. *Annual Review of Entomology* **28**: 407-453.

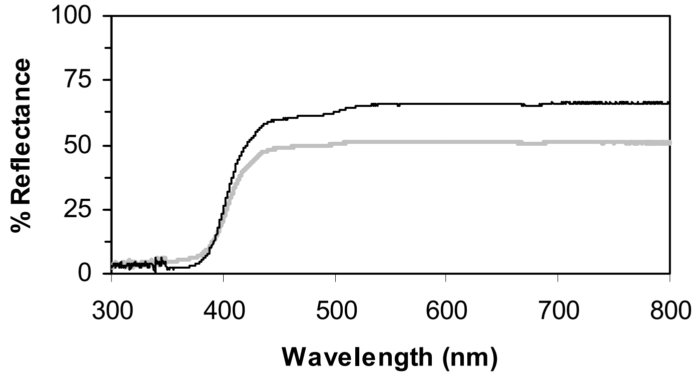
ACKNOWLEDGEMENT

We are grateful to *Sola Optical* for use of their spectrophotometer, and the expert assistance of Christine Voits who helped to set up and calibrate it.

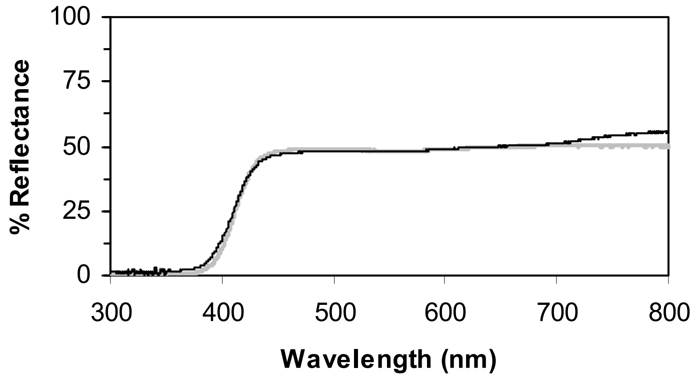
Figure 1. Spectral reflectance curves for selected flowers, leaves and paper cards. Replicated recordings are shown in grey.



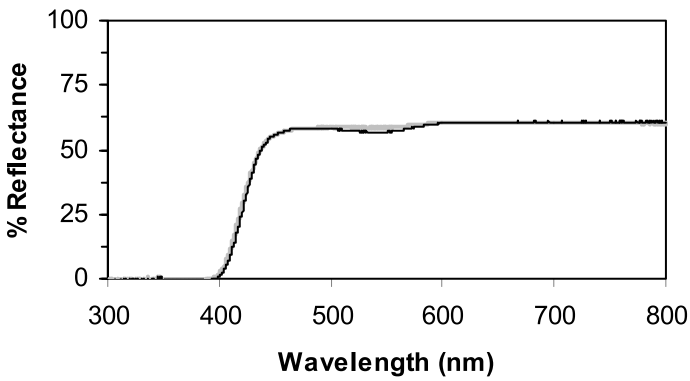
Star Jasmine Corola



Potato Vine Corola



Iceberg Rose Flower Petal



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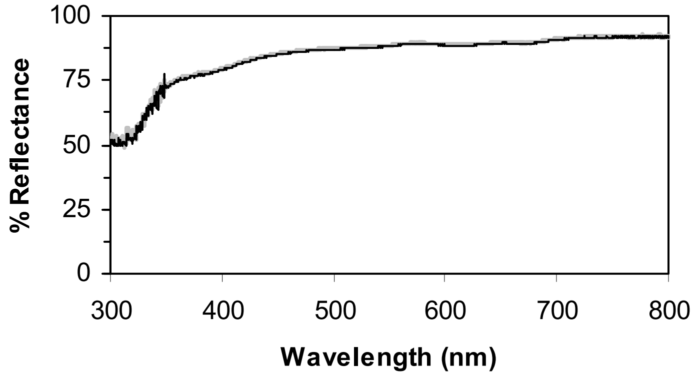

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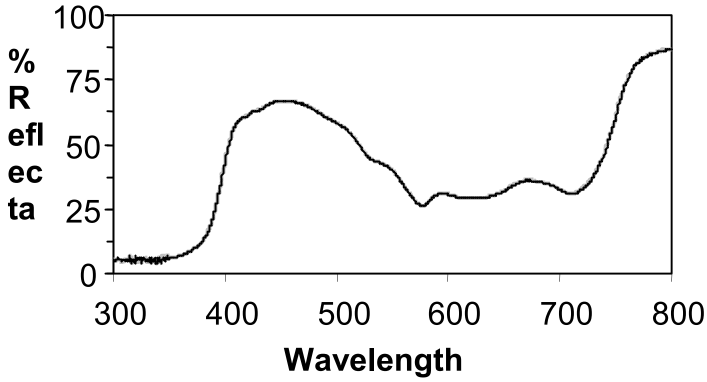
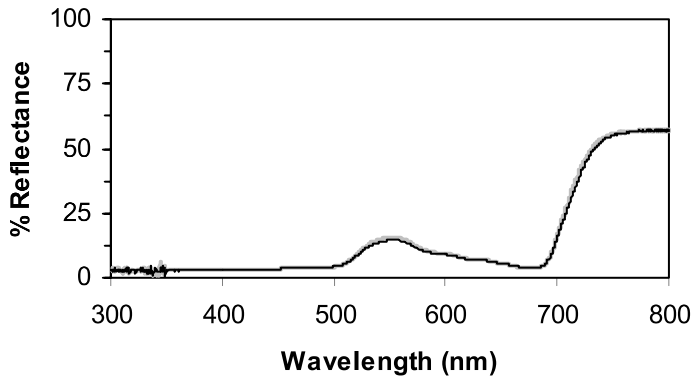
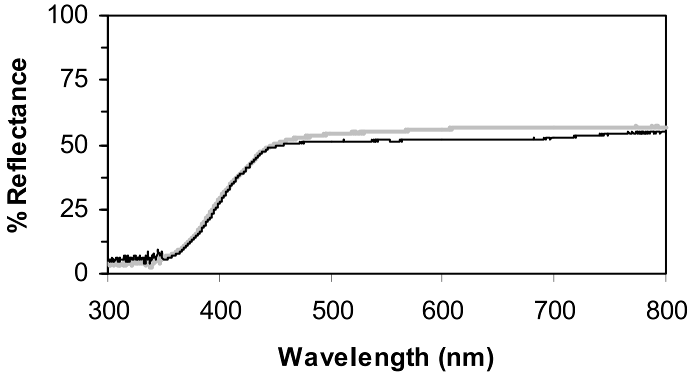


Lemon Leaf, Upper Surface



Ornamental Mandarin Flower Petals



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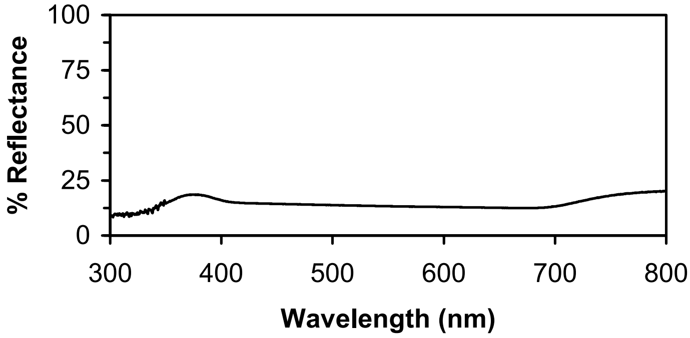


THE BIOLOGICAL CONTROL OF KCT IN RIVERLAND CITRUS ORCHARDS

INTRODUCTION

Kelly's citrus thrips (KCT) (*Pezothrips kellyanus* (Bagnall) (Thysanoptera: Thripidae) emerged in the 1990's as a serious citrus pest in southern Australia. Information about KCT biology and ecology has been scarce, and in Australia the management of KCT has been limited to the use of several insecticides. In previous studies very few natural enemies of KCT were identified in the canopy of citrus trees in Riverland-Sunraysia orchards (Baker *et al.* 2000). In recognition of the KCT's soil-pupating behaviour, the search for biocontrol agents was redirected to the soil and soil-litter, in particular to the potential role of predatory mites. As the Project team gained acarological research experience, attention was then re-focused on the canopy to identify arboreal predatory mites.

The primary objectives of this study were:
1. To identify soil-dwelling and arboreal natural enemies of KCT in Riverland citrus orchards.
2. To evaluate the potential of these natural enemies to regulate KCT populations in a commercial citrus IPM system.

METHODS

One organic (Loxton North) and two conventional-production navel orange orchards (Sunlands and Ramco) in the Riverland were chosen. The organic and one of the conventional (Conv. 1) orchards had a known history of low KCT abundance, and the other conventional orchard (Conv. 2) a history of high KCT abundance.

KCT abundance and pupal mortality were estimated using water traps (which sampled KCT larvae dropping from the canopy) and Tanglefoot®-coated ground traps (which sampled KCT adults emerging from ground) placed under the canopy as described in Baker *et al*. 2000, and the latter calculated by the formula:

$$\% \text{ pupal mortality} = \frac{\text{cumulative \# KCT larvae m}^2 - \text{cumulative \# KCT adults m}^2}{\text{Cumulative \# KCT larvae m}^2} \times 100$$

Soil-dwelling predators were sampled with pitfall traps (300 ml plastic tapered cups, 75 mm diameter at top, dug into the soil with the top flush with ground level) and Berlese funnel extraction of soil-litter samples. The pitfall sampling consisted of 10 pitfall traps left in the field for 24 hours. The pitfall catch was then rinsed into a specimen container using 80% ethanol. The soil-litter sample consisted of 20 trowel scrapes taken from 1-2 cm depth and each placed in a plastic bag. The pitfall traps were placed along, and the soil-litter samples were taken from, the drip-line. A 250 cc subsample of each of these samples was then placed in a Berlese funnel (sealed 22 cm diameter funnel fitted with a 15 watt light globe) and left for a 72-hour extraction period. The extracted invertebrates were collected in a specimen container containing 80% ethanol.

The soil-dwelling predatory mite data were analysed for site-to-site differences in abundance using ANOVA, and for differences in diversity and richness using the Shannon-Weiner diversity index (Southwood and Henderson 2000) and the species richness index d (where d=(S-1)/log N, and S = the number of species and N = the number of individuals) .

Arboreal natural enemies were sampled by placing 20 double-sided sticky bands (10 mm width) on twigs in the outer canopy near fruits, and on fruits. Sampling occurred quarterly.

Ground cover plant diversity and composition was quantified using the Levy Point quadrat method (Levy 1927) (Fig. 1). The ten probe Levy Point quadrat assessment was made at 20 randomly selected sites along the interrow over 3-4 rows in the study section of each orchard on four occasions (December 2001, April 2002, July 2002, October 2002). These quadrat data were used to calculate (i) the faunistic composition of the ground cover, (ii) the percentage of the ground covered with plants, (iii) % broadleaf cover, (iv) % grass cover, (v) the leaf area index, and (vi) the Shannon-Weiner diversity index. Soil organic carbon content was measured by taking 10 random trowel scrapes of 1-2 cm depth along the drip-line over 3-4 rows in the study section of each orchard, and then analysing a composite subsample using the modified Walkley-Black method (Rayment and Higginson 1992).

Fig. 1. Levy Point quadrat device used to quantify the diversity and composition of ground cover in the study citrus orchards.

RESULTS

The relationship between KCT canopy abundance and soil mortality

Organic and Conv. 1 had a relatively low abundance of KCT in the canopy in both study years, as evidenced by the estimated cumulative number of 100 - 300 KCT larvae m^2 $year^2$ captured in the water traps placed under the drip line. By contrast, Conv. 2 had 1500 – 1800 KCT larvae m^2 $year^2$ captured in water traps over the same time period (Fig. 2).

The abundance of KCT in the canopy is inversely related to the mortality of the soil-dwelling life-stages of KCT at the three study orchards.

In both years the mortality of KCT pupae in the soil was significantly higher at Organic and Conv. 1 compared to Conv. 2. The mean % mortality for the pooled data of Organic and Conv. 1 over the two years of the study is 97.3%, compared to 74.9% for Conv. 2. That is, for every 100 mature larvae that fell from the canopy to pupate at Organic and Conv. 1, on

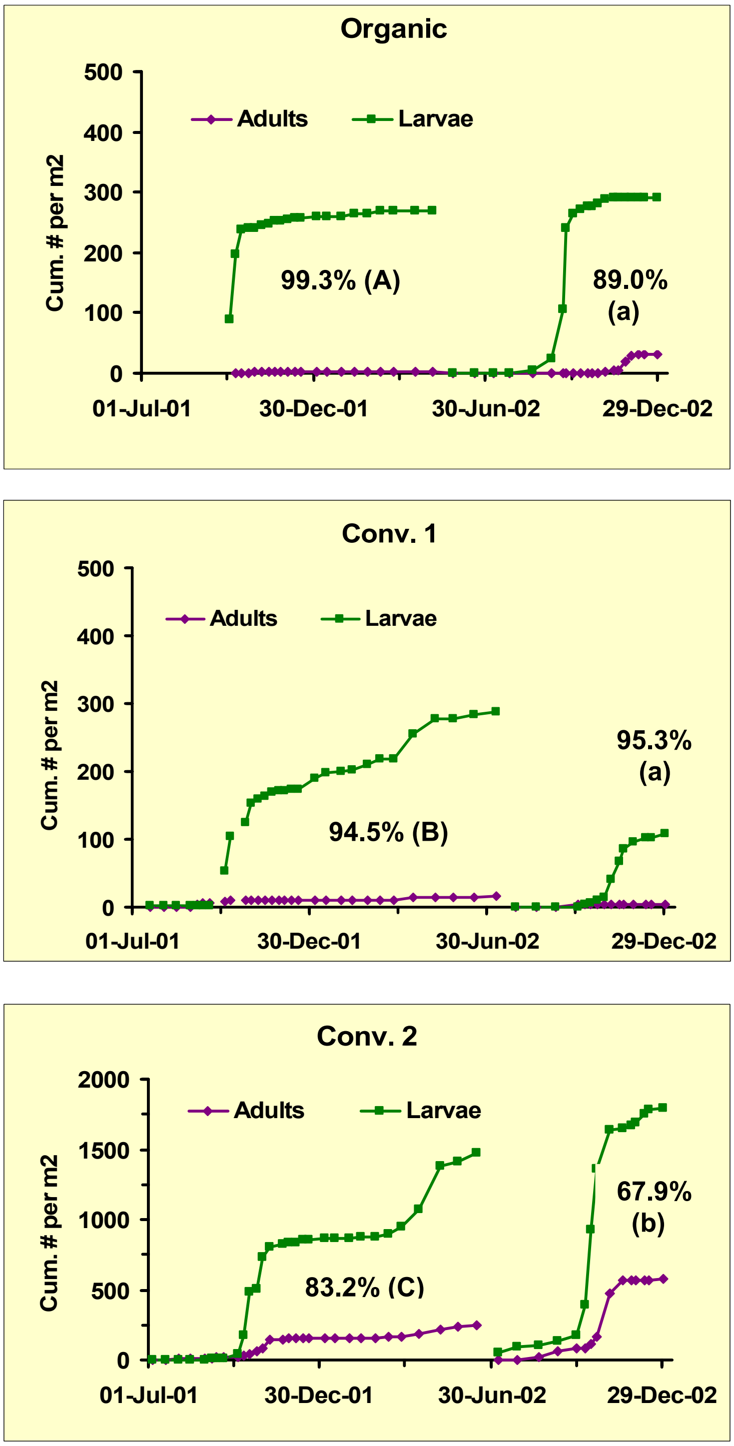


Fig. 2. The cumulative number of KCT 2nd instar larvae captured in water traps and KCT adults captured in sticky emergence traps placed under the canopy of the 3 study orchards, 2001-03. The mean % mortalities of KCT in the soil in each year at these 3 sites are presented. For each year, means followed by different letters are significantly different, ANOVA (P<0.05).

average only 2.7 adult KCT later emerged from the soil. By contrast, for every 100 mature larvae that fell from the canopy at Conv. 2, on average 25.1 adult KCT later emerged. This represents a rate of adult KCT soil-emergence (or KCT pupal mortality) and reinfestation of Conv. 2 that is more than nine times greater than that which was occurring at Organic and Conv. 1 over the same time period. This difference in soil survival between Conv. 2 and the other two study sites is of sufficient magnitude to largely explain the differences in KCT abundance between these two.

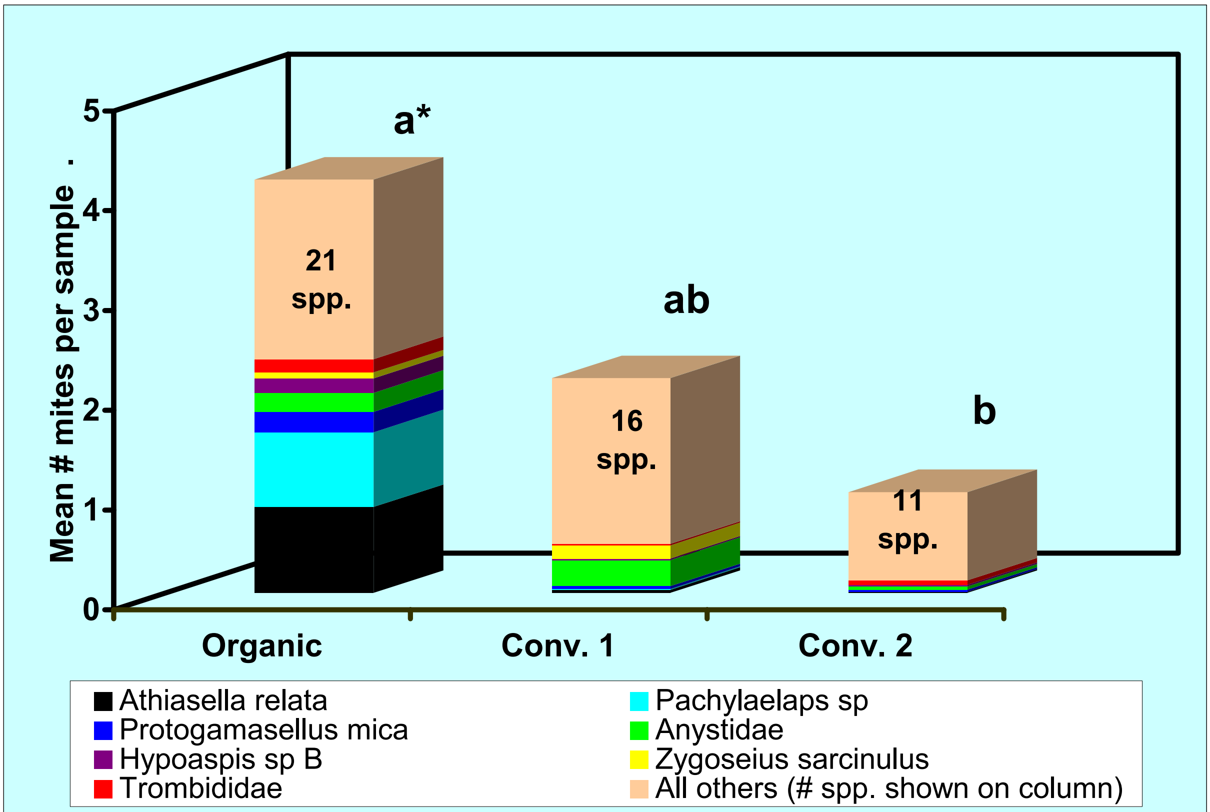
In the first year of the study the mortality of KCT pupae in the soil was significantly higher at Organic compared to Conv. 1, but in the second year this difference between these two sites was not statistically significant (Fig. 1).

The soil-dwelling predators, and their relationship to KCT abundance, ground cover composition and soil organic carbon content

The abundance and diversity of soil-dwelling predatory mites was greatest at the Organic orchard and least at Conv. 2 (Fig. 3 and Table 1).

The mean number of predatory mites per sample collected at the Organic, Conv. 1 and Conv. 2 sites throughout the two-year study were 4.14, 2.15 and 1.01 respectively. This is a significantly higher abundance of predatory mites at the Organic orchard compared to the Conv. 2 orchard, whilst the intermediate abundance recorded for Conv. 1 is not significantly different from either of the other orchards.

Several predatory mite species were notably more abundant at the organic orchard. These species include *Athiasella relata* (Ologomasidae), *Pachylaelaps* sp. (Pachylaelapidae), and *Protogamasellus mica* and *P. massula* (Ascidae).



*Different letters indicate that the mean # of all predatory mites sampled at these orchards differ significantly, LSD test (P<0.05).

Fig. 3. The mean number of predatory mites per sample at the 3 study orchards, 2001-03.

Table 1. The mean number of specimens of nineteen predatory mite species per Berlese funnel sample collected at the 3 Riverland study orchards, 2001-03.

Species (family)	Site			LSD*
	Organic	Conv. 1	Conv. 2	
Anystidae spp.	0.188a[†]	0.261a	0.032a	*0.348*
Arctoseius sp. (Ascidae)	0.133a	0.0a	0.01a	*0.035*
Athiasiella relata (Ologomasidae)	0.86a	0.02a	0.01a	*1.160*
Bdella sp. (Bdellidae)	0.36a	0.561a	0.357a	*0.564*
Cyta sp. (Bdellidae)	0.133a	0.036a	0.102a	*0.162*
Cunaxidae sp.	0.005a	0.035a	0.015a	*0.058*
Dendrolaelaps sp. (Digamasellidae)	0.052a	0.025a	0.005a	*0.057*
Erythracarus sp. (Anystidae)	0.018a	0.378b	0.089ab	*0.352*
Hypoaspis sp. A (Laelapidae)	0.03a	0.0a	0.0a	*0.055*
Hypoaspis sp. B (Laelapidae)	0.144a	0.015a	0.016a	*0.191*
Lasioseius sp. (Ascidae)	0.05a	0.047a	0.037a	*0.052*
Pachylaelaps sp. (Pachylaelapidae)	0.758a	0.011b	0.0b	*0.438*
Pergamus sp. (Parasitidae)	0.011a	0.0a	0.022a	*0.092*
Protogamasellus mica (Ascidae)	0.260a	0.026b	0.022b	*0.160*
Protogamasellus massula (Ascidae)	0.116a	0.021a	0.037a	*0.164*
Stigmaeidae spp.	0.016a	0.178a	0.005a	*0.279*
Trombididae spp.	0.134a	0.015a	0.045a	*0.129*
Zygoseius sarcinulus (Pachylaelapidae)	0.06a	0.161a	0.0a	*0.161*
Other spp.	0.893a	0.168b	0.155b	*0.510*
Total spp.	**4.14a**	**2.15ab**	**1.01b**	***2.063***

[†] Numbers followed by different letters are significantly different at $P \leq 0.05$ by the Least Significant Difference test.
* The critical LSD value at P=0.05.

Trends in the data suggest that a greater number of soil-dwelling predatory mite species were collected from the Organic orchard, and that a lesser number were collected from Conv. 2 (Table 2). Similarly, the greatest and the least values of species diversity (Shannon-Weiner diversity index) and of species richness were recorded for the Organic and Conv. 2 orchards respectively (Table 2). However, the differences between these three orchards in (i) the number of predatory species and (ii) the values of the Shannon-Weiner diversity indices were not significantly different, and no tests were conducted to test the significance of the differences between these orchards in their species richness indices.

These comparative orchard studies suggest there is a negative correlation between KCT pupal survival and the abundance of soil-dwelling predatory mite populations. In addition to these field correlations between KCT pupal survival and soil-dwelling predatory mite abundance, laboratory feeding trials with several of these soil-dwelling predatory mite species have demonstrated that they successfully develop and reproduce on a diet of KCT propupae (see Appendix 1).

Table 2. The number of soil-dwelling predatory mite species and the Shannon-Weiner diversity indices and species richness indices for these mite populations at the three Riverland study sites, 2001-2003.

Mite population statistic	Site		
	Organic	**Conv. 1**	**Conv. 2**
Number of predatory species	28a*	23a	19a
Shannon-Weiner diversity index	1.57a†	1.13a	0.94a
Species richness index (d)	9.23	8.59	6.67

*Means followed by the same letter are nsd, X^2 Goodness of Fit test (0.50<*P*<0.25).

†Means followed by the same letter are nsd, ANOVA (*P*=0.05).

Differences between the 3 study orchards in the abundance and species composition of soil-dwelling predatory insects and spiders captured in pitfall traps placed along the drip-line were not significant (ANOVA, *P*>0.05) (Table 3). Although the differences in abundance of these macro-invertebrate predators were not significant, it is interesting to note that 30.1% more macro-invertebrate predator specimens were collected at the Organic orchard compared to the Conv. 2 orchard.

Table 3. The mean number of four taxa of macro-invertebrate predators captured per pitfall trap placed along the drip-line of trees in the three study orchards, Riverland, 2001-03.

Macro-invertebrate predator taxon	Site			*P* value
	Organic	**Conv. 1**	**Conv. 2**	
Ants (Formicidae)	2.42a†	2.24a	1.64a	*0.7774*
Rove beetles (Staphylinidae)	0.04a	0.02a	0.08a	*0.5203*
Ground beetles (Carabidae)	0.11a	0.02a	0.08a	*0.0955*
Spiders (Araneida)	0.50a	0.54a	0.57a	*0.6237*
Total	**3.07a**	**2.82a**	**2.36a**	***0.7535***

†Means in each row followed by the same letter are nsd, ANOVA.

The results of the Levy Point quadrat study of ground-cover plant composition and diversity at the three study orchards are summarized in Table 4. A total of 29 plant species were identified amongst the ground cover of these orchards. Nine of these were grasses, and the remainder were a range of pasture legumes (*Trifolium* and *Medicago*) and broad-leaf weeds. The Organic and Conv. 2 orchards each had a significantly greater percentage of the ground covered with plants, percentage broadleaf cover and Shannon-Weiner diversity index values than Conv. 1 (Fig. 4). However these differences do not correlate with the observed differences between these orchards in KCT abundance, KCT mortality and soil predatory mites.

Please note that the Levy Point sampling only occurred in the interrow area. Hence the percentage of ground covered with plants is a measure of the density of the ground cover in this interrow area, not the percentage of total orchard ground area covered with ground vegetation. This latter statistic would have a much higher percentage value for Organic, compared to Conv. 1 and Conv. 2, than the values for '% ground covered with plants' presented in Table 4. This is because the conventional orchards were herbicided under the drip-line and under-tree area, whereas the Organic orchard had ground-cover growing under the canopy of the trees (Fig. 4).

Fig. 4. The ground cover at the Organic (top), Conv. 1 (mid) and Conv. 2 (bottom) orchards. Note the ground cover extending under the trees at Organic, as opposed to the herbicided, ground-cover free drip-line and under tree areas of Conv. 1 and Conv. 2. Also note the grass-broadleaf mix at Organic, the dominant grass component at Conv. 1 and the absence of grasses at Conv. 2.

Table 4. The percentage of the ground covered with plants, percentage broadleaf cover, percentage grass cover and the Shannon-Weiner diversity index for the ground cover of the three study sites, Riverland, 2001-03.

Ground cover statistic	**Site**			***P***
	Organic	**Conv. 1**	**Conv. 2**	***value***
% ground covered with plants	64.6a[†]	32.9b	66.6a	*0.0024*
% broadleaf cover	38.7a	4.5b	52.1a	*0.0001*
% grass cover	25.6a	28.3a	11.9a	*0.0629*
Shannon-Weiner diversity index	1.51a	0.58b	1.26a	*0.0003*

[†]Means in each row followed by the same letter are nsd, ANOVA.

Although the other ground cover statistic, the percentage grass cover, did not significantly differ between the three orchards (*P*=0.06), Organic and Conv. 1 had more than twice the amount of grass cover compared to Conv. 2 (Fig. 4). At one other orchard (Altmann, New Residence) observed in this study a high grass component in the ground cover was also associated with low KCT incidence and high soil-dwelling predatory mite activity. Do the grasses provide a benefit to the soil predators and thereby aid the biological control of KCT? This remains unclear. However the arboreal predatory mite *Amblyseius victoriensis* is favoured in Queensland citrus where growers retain and allow flowering of Rhodes grass, because the wind-blown grass pollen provides a supplementary food source for the *A. victoriensis* (Smith and Papacek 1991).

Colloff *et a.l.* (2003) note that properties "free" of KCT "tended to have inter-row ground cover consisting of fairly dense and diverse swards of perennial grasses and herbs, whereas those with a thrips problem had a bare soil, annual weeds or a monoculture (eg. lucerne)". They also state "species-rich, perennial ground cover probably provides better habitat for predatory mites, as well as a reliable source of pollen, which many soil-dwelling predatory mites use as a supplementary food source". In our study the three main study sites, plus a range of other sites which were visited, had a succession of different ground cover plant species replacing each other as the seasons progressed. Generally a range of broadleaf and grass species were present at all times, albeit the percentage ground cover tended to be somewhat less in most orchards in the summer. That is, each of our main study sites possessed a fairly dense and diverse sward of ground cover species. Albeit not statistically significant, the lesser incidence of grasses at the Conv. 2 site compared to the Organic and Conv 1 sites was noticeable, and given the observations of Collof *et al.* (2003) and the Queensland evidence with the Rhodes grass – *A. victoriensis* system, the role of flowering grasses in the maintenance of these Riverland citrus predatory mite populations should perhaps be further investigated.

Soil organic carbon content positively correlates with predator abundance and KCT mortality. (5.3, 2.5 and 1.5 g C/ kg soil at Organic, Conv. 1 and Conv. 2 respectively.) Whether this is an important determinant of predator abundance, and if so, what is the nature of the relationship between organic carbon levels and mite abundance, remain unknown. However studies in Asian irrigated rice (Settle *et al.* 1996) have demonstrated that by increasing organic matter in test plots, populations of detrivores and plankton-feeders were boosted, and in turn the abundance of generalist predators were boosted. The mesostigmatid predatory mites of Riverland-Sunraysia citrus soils are similarly generalist feeders, and would be expected to benefit from most conditions that boost the abundance of detrivores, fungae and other food sources in these soils. The influence that soil organic carbon content has on the abundance and diversity of KCT natural enemies in the soils of Riverland citrus should be further studied to help develop an orchard management system that optimizes the contribution of soil-dwelling natural enemies to the control of KCT.

The Arboreal Predators

In Project CT97007 Baker *et al*. (2000) determined that a range of generalist predatory insects (mirids, chrysopids, coccinellids, *Haplothrips* sp.) and the eulophid parasitoid *Ceranisus menes* were present in most Riverland orchards at low densities. These predators and parasites were also present in the three orchards of this study at similar low densities.

Arboreal predatory mites, which belonged to four families (Erythraediae, Phytoseiidae, Anystidae and Stigmaeidae), were more abundant in the canopy of the Organic and Conv. 1 orchards compared to the Conv. 2 orchard (Table 5). The extent to which these populations of arboreal predatory mites are contributing to the control of KCT remains unclear.

Table 5. Mean number of arboreal predatory mites per 20 sticky band sample placed in the canopy of the three Riverland study orchards, 2001-03.

Orchard	Mean # of arboreal predatory mites
Organic	33.0a[†]
Conv. 1	19.1a
Conv. 2	11.8b

[†]Means followed by same letter are nsd, Tukey HSD test (P>0.05).

In summary, there are complexes of soil-dwelling and arboreal mite species that are either demonstrated or likely predators of KCT in Australian citrus orchards. Negative correlations between the abundance of KCT and the predators, and between the survival of soil-dwelling KCT and the abundance of the soil predatory mites suggest an important causal link between predatory mites and low thrips numbers. Further research is needed to fully answer several key questions, namely:

1. Can the boosting of organic carbon (OC) levels in Riverland-Sunraysia citrus soils cost-effectively promote KCT predator activity and biocontrol, and thereby cost-effectively reduce KCT packout losses? If so, what is the minimum OC level required?
2. Can increasing the grass-pollen production of Riverland-Sunraysia citrus ground covers promote KCT predator activity and biocontrol? Which grasses are most beneficial?
3. What impacts are commonly used insecticides and herbicides having on KCT predator activity and biocontrol in Riverland-Sunraysia citrus? (See Section titled “Impact of chlorpyrifos on predatory mite populations in Riverland citrus orchards” in this report.)

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LITERATURE CITED

Levy, E.B. 1927. The grasslands of New Zealand. *New Zealand Journal of Agriculture* **34**: 147-148.

Colloff, M.J., Fokstuen, G. and T. Boland. 2003. Towards the triple bottom line in sustainable horticulture: biodiversity, ecosystem services and an environmental management system for citrus orchards in the Riverland of South Australia. CSIRO Entomology, Canberra.

Rayment, G.E. and F. R. Higginson. 1992. Australian laboratory handbook of soil and water chemical methods. Inkata Press.

Settle, W., Ariawan, H., Tri Astuti, E., Cahyana, W., Hakim, A.L., Hindayana, D., Lestare, A.S. and Pajarningsih. 1996. Managing tropical rice pests through conservation of generalist natural enemies and alternative prey. *Ecology* **77**: 1975-1988.

Smith, D. and D.F. Papacek. 1991. Studies of the predatory mite Amblyseius victoriensis (Acarina: Phytoseiidae) in citrus orchards in south-east Queensland: control of Tegolophus australis and Phylocoptruta oleivora (Acarina: Eriophyidae), effects of pesticides, alternative host plants and augmentative release. *Exp. Appl. Acarol*. **12**: 195-217.

Southwood, T.R.E. and P.A. Henderson. 2000. Ecological Methods. Third edition. Blackwell Science.

GROUND-COVER MANAGEMENT TRIAL

INTRODUCTION

Research conducted by Colloff *et al.*2001) indicates that citrus orchards with diverse under-story vegetation and ground cover, that provides moisture retention and important habitat requirements, are more likely to support an abundant and diverse soil fauna of predatory invertebrates. Further to this, Californian research (Hoddle *et al*. 2002) has demonstrated a 50% reduction in adult avocado thrips emergence from under avocado trees mulched to a depth of 6-8 inches with organic yard waste. The Californian research suggests that avocado thrips pupation rates may be significantly reduced through the application of organic mulches and an increase in the soil predatory fauna.

A field trial was initiated in 2001 in a commercial orchard in the Riverland, to assess the influence of several ground-cover treatments on the abundance of predatory mites and KCT larval and pupal survival. As it can take some time for changes in ecological systems to become measurable, it was anticipated that this trial would run for a number of years.

METHODS

The trial was conducted in a commercial Valencia orchard at Bookpurnong in the Riverland. Trees were +20 years old, un-skirted and watered using under-tree sprinklers. The existing ground cover consisted of medium ground coverage with perennial grasses and weeds growing in an approximately 2m wide swath. The under-tree canopy was kept clear of weeds through herbicide usage and covered with 1-2cm of leaf litter.

Treatments were applied during the last week of July 2001. Treatment 1 (lucerne hay mulch) was re-applied in September 2002.

The treatments were:
1. Lucerne hay mulch (5cm depth) under tree canopy,
2. Perennial grasses (60% Drylander Perennial ryegrass, 20% Wimmera Annual Ryegrass, 20% Kambria Cocksfoot orchard grass) planted into mid-row and
3. Control (untreated).

The treatments were replicated 4 times in plots of 4 rows x 4 trees. Two larval pan and two adult emergence traps were set in each plot and monitored at fortnightly intervals during Spring 2001, when the experiment was initiated, to confirm that KCT abundance was similar across the trial plots prior to the ground-cover treatments taking effect.

In July 2002, soil samples (25cm x 25cm x 2cm) were collected from the tree drip-line of plots to assess treatment effects on soil-dwelling predatory mite populations.

RESULTS

Because of poor establishment by the newly-sown perennial grasses, there was actually less vegetation mid-row than in the control treatments. Despite a large amount of leaf litter under most trees, the under-tree area of the mulching treatment was still appreciably different from that of the other two treatments.

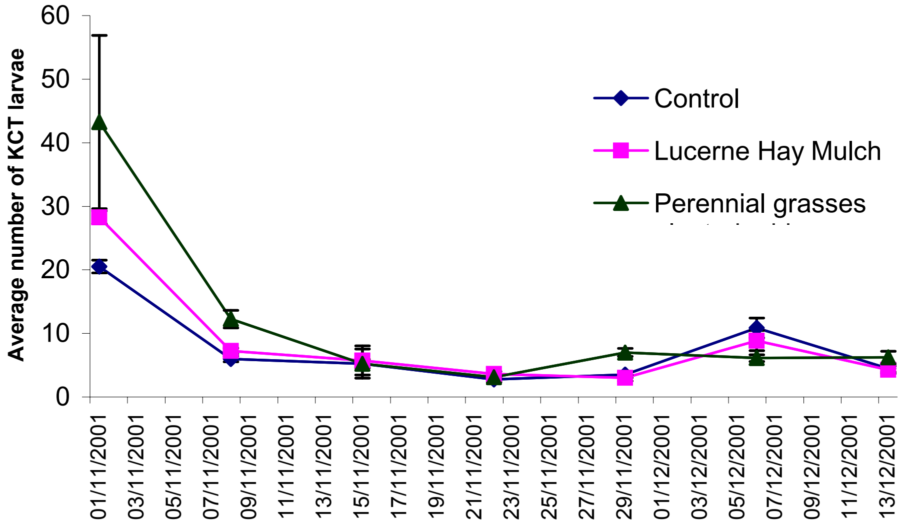

Fig. 1. Average number of KCT larvae collected from pan traps placed on the drip-line of trees at the Ground Management trial, Bookpurnong, Spring 2001.

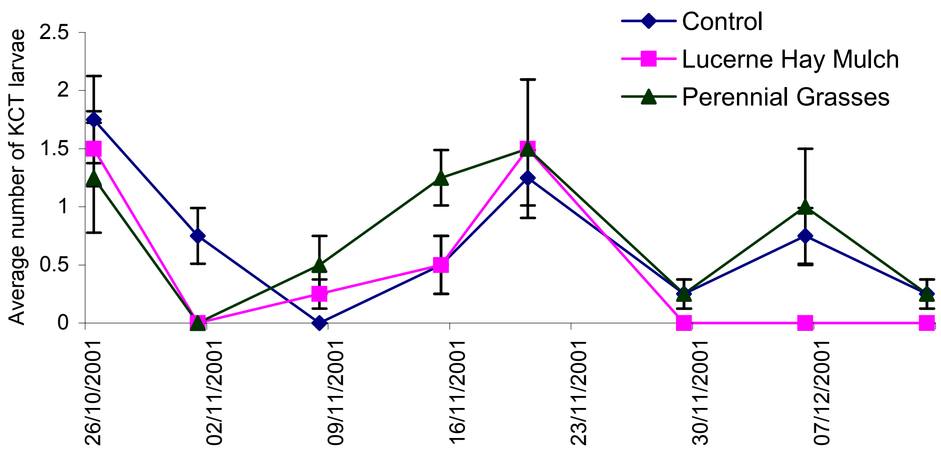

Fig. 2. Average number of KCT larvae collected from samples of 20 fruitlets at the Ground Management trial, Bookpurnong, Spring 2001.

Generally the KCT larval densities recorded in the pan trap and fruitlet samples in November 2001 were similar across the three treatments (Figs. 1 and 2). However on several sample occasions differences in KCT larval densities were recorded. Given that the ground management treatments had only been in place for several months, these differences in larval density more likely reflect natural field population variability and the need for larger sample sizes, rather than being causally linked to the newly-imposed ground treatments.

The results of the first assessment of soil predatory mite populations in the three treatment areas are presented in Fig. 3. This assessment occurred one year after the trial's commencement. Whether this is sufficient time for any soil faunistic responses to the ground-cover management changes to have become detectable remains unclear. However, the single July 2002 data-set provides no evidence of any positive effect on soil predatory mite populations from either of the new ground-cover treatments.

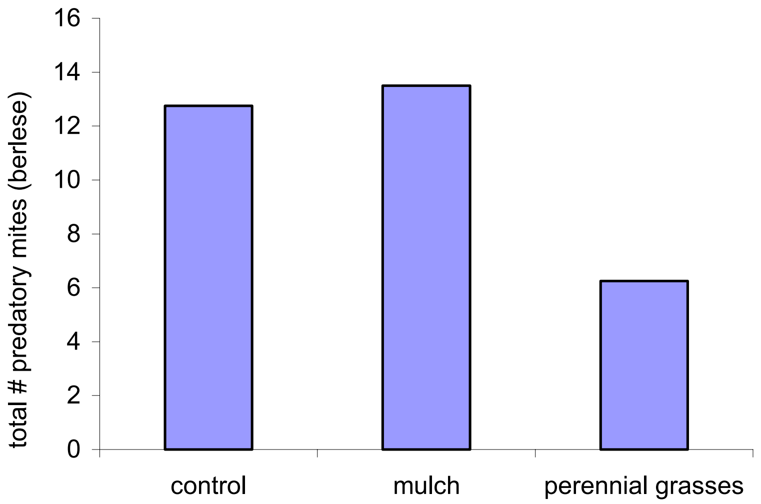

Fig. 3. The total number of predatory mites extracted from Berlese soil samples collected from the drip line of trees on July 26, 2002.

DISCUSSION

Up until February 2003, when the grower co-operator unfortunately bulldozed the site without informing us, no significant treatment effects had been detected in regards to KCT larval/pupal survival or the abundance of predatory mites. Given that the response by beneficial organisms to such changes in orchard management may be slow, it remains unclear whether the non-response was due to the limited duration of the study or to the unsuitability of the treatments.

Hoddle *et al*. (2002) have used composted garden refuse, laid ~20cm deep under avocado trees, to substantially reduce the survival of the soil-pupating avocado thrips in commercial Californian orchards. These Californian researchers are still trying to determine whether this effect was primarily due to a direct effect on the thrips (eg. lethal effect of compost heat on the thrips), or indirectly through benefiting natural enemies (eg. predatory mites) or pathogens of the thrips. In light of this Californian work with a soil-pupating thrips that has similar biology to KCT, should further funding arise, an investigation of the impact that this form of mulching has on the survival of KCT is warranted.

LITERATURE CITED

Colloff, M.J., Fokstuen, G. and T. Boland. 2003. Towards the triple bottom line in sustainable horticulture: biodiversity, ecosystem services and an environmental management system for citrus orchards in the Riverland of South Australia. CSIRO Entomology, Canberra.

Hoddle, M.S., Morse, J. G., Oevering, P., Phillips, P. A. and Faber, B. A. (2002). Further progress on Avocado thrips biology and management. www.avocado.org/growers/symposium_2002/a1.pdf

THE IMPACT OF IRRIGATION REGIME (SOIL MOISTURE) ON KCT PUPAL SURVIVAL

INTRODUCTION

A useful spin-off from the *Metahizium* study has been evidence that KCT pupae are killed by dry soil conditions. Hence it may be possible to modify irrigation scheduling at critical times to control KCT pupal infestations. The impact of irrigation regime (soil moisture) on KCT pupal and predatory mite survival was investigated in conjunction with Loxton Fruit Doctors and Solora Orchards.

METHODS

Site and Treatment details

The unreplicated trial was conducted in a navel orchard comprised of 6 adjacent sections (A1-A6). Each section contained 9 rows of 53 trees. Sections A1, A2 and A3 were treated with the regular irrigation schedule, while sections A4, A5 and A6 had their irrigation schedule restricted.

The covercrop in sections A4, A5 and A6 was sprayed with herbicide in August and September and the trees received two applications of Success™ in November. Sections A1, A2 and A3 were treated with a single Supracide application in November, and no herbicide applications. Unfortunately, we were unable to influence the decision to confound the design of this trial with the application of these different insecticide and herbicide treatments.

For the purpose of this trial, assessments were made from row 5 of sections A2 (the 'regular irrigation' treatment) and A5 (the 'restricted irrigation' treatment).

Table 1. The irrigation, herbicide and insecticide treatments applied to the regular and restricted irrigation sections, Solora Orchards, Bookpurnong, Riverland, Aug-Dec 2003.

	Regular irrigation	**Restricted irrigation**
Section	A2	A5
Herbicide application	-	19/8/03 (Roundup Max @ 1.7L/ha) 5/9/03 (Sprayseed @ 2.5L/ha)
Insecticide application	17/11/03 (Supracide 125ml/100L @ 6000L/ha)	27/11/03 and 11/12/03 (Success 20ml/100L @ 4500L/ha)
Irrigation applied	23/9/03 8hrs, 30.4mm 13/10/03 6hrs, 22.8mm 23/10/03 6hrs, 22.8mm 24/10/03 4hrs, 15.2mm 1/11/03 6hrs, 22.8mm 5/11/03 4hrs, 15.2mm 11/11/03 10hrs, 38mm 20/11/03 10hrs, 38mm 1/12/03 12hrs, 45.6mm	19/9/03 8hrs, 30.4mm 19/10/03 10hrs, 38mm 10/11/03 6hrs, 22.8mm 12/11/03 4hrs, 15.2mm 29/11/03 12hrs, 45.6mm
Total irrigation applied	**66 hours, 250.8mm**	

Assessment of Irrigation Regime on survival of KCT pupae

Twenty large adult emergence traps (45cm diameter) were set along the drip line of alternate trees in row 5 of each of sections A2 and A5, and serviced weekly from 24/10/03 until 8/12/03.

In an attempt to address the confounding effects of the different herbicide and insecticide treatments applied to the two irrigation treatment areas, ten 2m x 2m plots were marked out under the drip line of every second tree along row 4 of section A2. These plots were given the same irrigation schedule as that applied to section A4-A6 by kinking sprinklers as required. On the 24th November, 25 KCT pupae were seeded into each of these plots, as well as in ten sites along row 5 of A2 and A5. Seeded pupae were covered with a fine mesh tent (15cm diameter). A yellow sticky trap was suspended within the tent to catch emerging adults. The mesh traps were designed to allow irrigation and rain to penetrate the seeded soil. These traps were collected after 15 days.

Assessment of Irrigation Regime on survival of predatory mites

On the 27th October, twenty soil samples (25cm x 25cm x 2cm) were collected from the drip line of trees along row 5 of sections A2 (regular) and A5 (restricted). The Berlese funnel method was used to extract predatory mites from the soil samples. Mites were collected directly in vials of 80% alcohol for short-term storage, and then slide-mounted for species identification.

Statistical analysis

Statistix was used to analyse the data. The effect of treatments number of adult KCT caught on emergence traps was analysed with ANOVA, with differences tested for significance using the t-table at the probability level of P=0.05

RESULTS

Influence of Irrigation Regime on Survival of KCT Pupae

Large emergence trap assessments

For all but the last two monitoring dates, there was no significant difference in the number of adult KCT caught on emergence traps for each treatment (Table 1). On the last two monitoring dates (2/12/03 and 8/12/03) significantly more adult KCT were caught on emergence traps set within the restricted irrigation treatment. For the duration of the trapping period, the total number of adult KCT caught on emergence traps set within the restricted irrigation treatment was also significantly greater than the number caught on traps set within the regular irrigation treatment.

This runs counter to our working hypothesis that restricting irrigation, and thereby reducing soil moisture, would have a detrimental impact on KCT pupal survival within the soil and reduce KCT densities. However, due to the confounding treatment of different herbicide and insecticide applications, it is impossible to determine the cause of the significant differences in adult KCT emergence.

Table 9. Mean number (± s.e.) of KCT adults captured per 20 emergence traps in the 'regular' and 'restricted' irrigation sections, Solora Orchards, Riverland 2003. Means followed by the same letter for a particular date are not significantly different (*P*=0.05).

Date Collected	Mean number of adult KCT (±s.e.)	
	Restricted Irrigation	Regular Irrigation
24/10/03	0.35±0.15a	0.20±0.12a
3/11/03	0.95±0.38a	1.20±0.63a
10/11/03	3.40±1.82a	2.15±0.45a
17/11/03	1.70±0.47a	0.95±0.31a
24/11/03	5.05±2.10a	1.55±0.44a
2/12/03	1.30±0.43a	0.05±0.05b
8/12/03	0.20±0.09a	0.00b
Total catch	**1.86±0.43a**	**0.87±0.15b**

Seeding Assessment

The seeding trial was unsuccessful in determining the influence of soil moisture on the pupal development of KCT. Extremely low numbers of adult KCT were captured by the mesh traps in each treatment (Table 2). Irrespective of which treatment it appears that the seeding technique has resulted in high KCT mortality, and as a result it remains unclear whether the treatments would have affected KCT survival.

Table 10. The mean number of adult KCT captured per 'seeded' mesh trap in the Regular, Restricted and Small plot Restricted sections, Solora Orchards, 2003. Means followed by the same letter are not significantly different using the LSD test (*P* = 0.05).

Date		Mean Adult Emergence		
Set	Collected	Regular Irrigation	Restricted Irrigation	Small Plots (Restricted Irrigation)
24/11/03	9/12/03	0.0a	0.0a	0.6a

Influence of Irrigation Regime on Survival of Predatory Mites

The mite sampling data is presented in Table 3.

Two species were numerically dominant, namely *Pergamasus* sp. and *Zygoseius sarcinulus*, and both were more prevalent in the samples taken from the restricted irrigation section samples. The total number of predatory mites in the samples taken from the restricted irrigation section is significantly greater than the number in the regular irrigation sample (©^{2} Goodness of Fit test, ©^{2}= 11.8, v=1, *P*=0.001). However, because of the confounding treatments, it remains unclear whether these differences are due to the irrigation treatments.

Table 3. The numbers of predatory mites extracted by Berlese funnel from soil samples collected from the regular and restricted irrigation sections, Solora Orchards, 27 October 2003.

Predatory mite taxa	Irrigation section	
	Regular	Restricted
Anystidae	2	2
Arctoseius cetratus (Ascidae)	0	1
Cyta sp. (Bdellidae)	3	2
Pergamasus sp. (Parasitidae)	24	56
Phytoseiidae	2	1
Protogamasellus massula (Ascidae)	0	8
Zygoseius sarcinulus (Pachylaelapidae)	2	16
Total	**33**	**86**

DISCUSSION

This trial was conducted under adverse conditions. Firstly, there was low KCT pressure at the trial site, which makes it difficult to achieve statistically significant differences between treatments. Secondly, it was a wet spring, which counteracts the purpose behind the 'restricted' irrigation treatment. And thirdly, the confounding treatments (herbicide and insecticides) applied to section A2 make it impossible to determine which of these factors might have had a significant influence on KCT survival.

However, despite this trial disappointment the concept that soil moisture may influence KCT pupal survival is still worthy of further investigation. Should further funding arise, it is recommended that this alternative control strategy be investigated in a replicated trial with non-confounding treatments.

THE ASSESSMENT OF A *METARHIZIUM* ISOLATE AS A MYCOINSECTICIDE FOR THE CONTROL OF KCT

INTRODUCTION

Another area of investigation that was brought about by the discovery that KCT pupates solely in the soil was the potential of soil borne pathogens to control KCT. In collaboration with Richard Milner of CSIRO Entomology in Canberra, various soil pathogens were tested to determine the potential role of fungal diseases in the management of KCT in citrus orchards.

There was a need to assess the level of potentially useful fungi in the soil of citrus orchards; to laboratory bioassay isolates of *Beauveria* and *Metarhizium* as possible mycoinsecticides to be applied to the soil under citrus trees; and if suitable isolates were found to test these in field trials.

MATERIALS AND METHODS

Survey of potentially useful fungi in the soil of citrus orchards

Soil scrapings were taken from seven citrus orchard sites consisting of Fechner's and McLean's near Waikerie, Pickering's and Radomi's near Loxton, Lindsay Point, and Yandilla Park near Renmark. These samples were sent to CSIRO Entomology and assessed for the presence of *Beauveria* and *Metarhizium* isolates.

Testing of isolates in the laboratory

A Potter tower was used to mix the fungal conidia suspension evenly into a vermiculite/peat substrate containing the KCT pupae. This substrate was then placed into a small conical flask and the number of adults emerging each day assessed using small yellow sticky traps placed at the neck of the flask. By comparing the number of KCT emerging in the controls and the treatments the efficacy of an isolate could be assessed.

There were difficulties in supplying adequate numbers of pupae to CSIRO Entomology to undertake these tests. The logistics of supplying adequate numbers of pupae from the laboratory culture at the right time, and of transporting the fragile KCT pre-pupae and pupae resulted in highly variable emergences, and with many of the pupae emerging as adults prior to their arrival in Canberra. Finally it was decided to treat the test substrates with the fungal suspensions in Canberra, and then to ship these to Adelaide where the tests were undertaken.

CSIRO Entomology provided five substrate treatments for testing:
1. Substrate treated with water (control).
2. Substrate treated with water and x 0.1 % Tween 85 (surfactant control).
3. Substrate treated with 0.25% *Metarhizium* FI-1248 isolate and 0.1 % Tween 85.
4. Substrate treated with 0.5% *Metarhizium* FI-1248 isolate and x 0.1 % Tween 85.
5. Substrate treated with 1.0% *Metarhizium* FI-1248 isolate and 0.1 % Tween 85.

Each treatment (4 replicates) was then placed into a small conical flask. Twenty KCT pupae were placed into each conical flask. The number of adults that emerged each day was assessed using small yellow sticky traps placed at the neck of the flask.

Testing of isolates in the field

A small orchard of navel oranges at the Loxton Research Centre was used for this trial. At this orchard 30 2 x 2 m plots were randomly allocated in 15 replicate blocks (one treatment and one control per block) along the drip line in a representative section of an orchard row. Each plot was positioned central to a corresponding tree.

At each of the control plots (15), a 2 x 2 m black plastic sheet was pegged down (Fig. 1). A 0.01 g ml^{-1} suspension of *Metarhizium anisopliae* var. *anisopliae* isolate FI-1248[1] was sprayed over all the 30 sites at a rate of 5 ml per m^2 . After removing all of the plastic sheets a 4-5 cm^2 area near centre of each plot was covered with a 1 cm deep layer of leaf-litter and soil. A container of 25 KCT pupae were then seeded into this leaf-litter. A sticky ground trap was then placed over each of these seeded areas to recover emerging adults. These traps were subsequently removed one week later and assessed.

Fig. 1. *Metarhizium* trial site illustrating the placement of 2 m x 2 m black plastic sheets for the control plots.

RESULTS & DISCUSSION

Survey of potentially useful fungi in the soil of citrus orchards

From the soil scrapings taken from the seven sites, no Metarhizium isolates were recovered. However there were four isolates of *Beauveria bassiana* all recovered from Yandilla Park. There was also an isolate of Paecilomyces sp., another fungal pathogen of insects, recovered from Yandilla Park. However, the number of spores of the various isolates recovered from

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[1] The *Metarhizium* isolate was prepared at the CSIRO laboratories and while attempts were made to contact Richard Milner to determine the concentrations of various components of the isolates, no contact was established and the isolate was applied as supplied.

the soil at Yandilla Park was very low, suggesting that the fungus was unlikely to be influencing the KCT population.

Testing of isolates in the laboratory

The logistical problems experienced transporting sufficient numbers of viable pupae to Canberra limited the value of the initial experiments. The mean numbers of KCT adults that were captured, even in the controls, were low, and treatment differences were not significant (Table 1). However, with the high *Metarhizium* dose (0.1 g ml^{-1}) adult KCT were observed to emerge and die exhibiting sporulation symptoms typical of *Metarhizium*. The observation that some KCT emerged and died later suggests that the actual number of pupae killed may be an underestimate of the treatment's efficacy. These survivors may well die before laying eggs or may lay fewer eggs than healthy adults.

Table 1. The mean number of adult KCT that had emerged five days after treating a vermiculite/peat substrate containing KCT pupae with several *Metarhizium anisopliae* var. *anisopliae* isolate FI-1248 and control treatments, and this number expressed as percentage survival.

Treatment	Mean number of adult KCT at 5DAT	% survival
Untreated	4.13 a	20.7
Tween only (surfactant)	6.25 a	31.2
FI-1248 (0.001 g ml^{-1}) plus Tween	8.25 a	41.3
FI-1248 (0.01 g ml^{-1}) plus Tween	3.50 a	17.5

Subsequent testing of isolates in the laboratory in Adelaide was a little more successful. Elimination of the interstate transportation resulted in a slightly higher and less variable emergence of KCT adults (Fig. 2). Importantly, there did appear to be a suppression of KCT emergence at the greatest *Metarhizium* concentration of 0.01 g ml^{-1} (mean of 2.8 adult KCT) compared to the water and surfactant Tween controls (means of 9.5 and 7.6 adult KCT respectively).

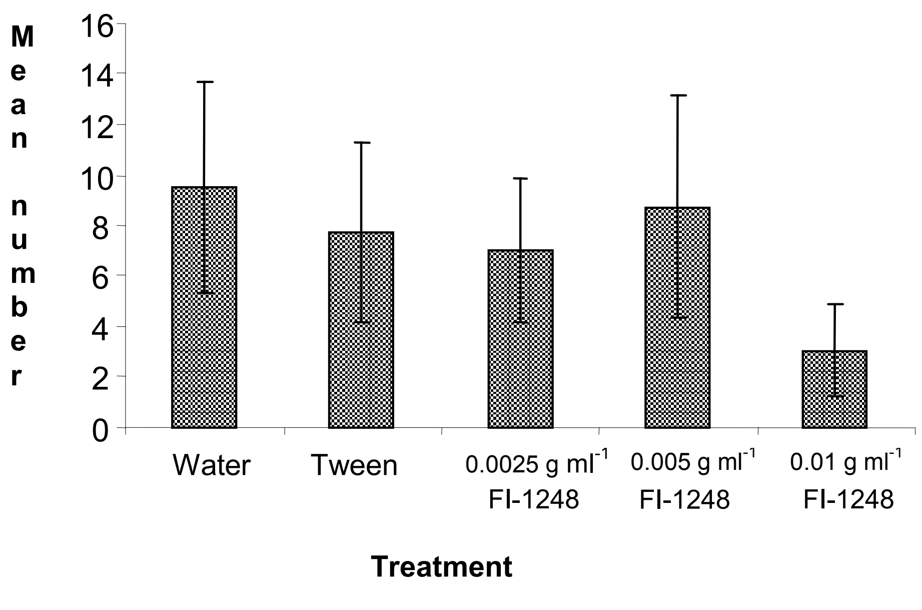


Fig. 2. Mean number of KCT adults emerging from a substrate of peat/vermiculite treated with various concentrations of *Metarhizium* isolate FI-1248.

Testing of isolates in the field

Unfortunately there was no observed treatment effect in the field test of the *Metarhizium* isolate FI-1248 (Fig. 2). Given that the dosage rate of *Metarhizium* applied in this field trail was extremely high (500 g spores ha^{-1}, when for most field uses it is applied at ~100 g spores ha^{-1}, Dr Richard Milner, *pers. comm.*.), and coupled with the reasonably poor laboratory trial results, there are no further plans to investigate the use of *Metarhizium* as an alternative control for KCT.

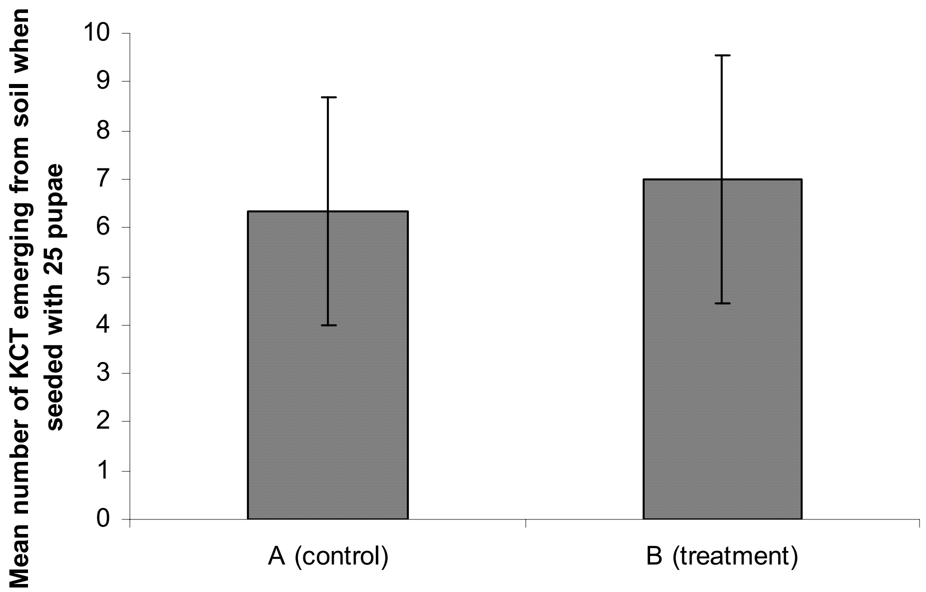


Fig. 2. Mean number of KCT emerging from the soil when seeded with 25 pupae (Error bars are 95% CI).

EXTENSION ACTIVITIES

EVENTS AND MEETINGS

2000-01

- Eleven KCT management presentations to Cittgroups in the Riverland, Riverina and Sunraysia districts, Aug-Oct 2000. (Two of these included presentations by visiting scientist Professor Carl Childers on citrus pest and disease management in Florida orchards.)
- Professor Carl Childers interviewed about citrus IPM by ABC Regional Radio and WIN TV.
- Research results and activities were presented at the ACG National Field Days, Renmark, May 2001. A written report on the KCT project was included in the 2001 ACG's Annual Conference and National Field Days book.
- Mildura District Horticultural Field days – KCT identification and monitoring posters in Riverlink tent. Colour A4 laminated copies of posters available to growers free due to MVCMB sponsorship.
- Five IPM career presentations to students at National Science Week.
- SA Citrus Board briefing on KCT research findings.

2001-02

- Poster presentation and live thrips identification exhibit with A4 colour laminated "KCT ID & Management" posters available to citrus growers free of charge at Riverland Field Days, September 2001.
- KCT management presentation at MVCMB IPM Workshop, Mildura, October 2001.
- Riverland Cittgroup presentation on KCT HAL project, December 2001.
- KCT presentation at Mildura Field Days, May 2002.
- WIN TV interview about KCT research activities.

2002-03

- Poster presentation at ACG Conference, Leeton NSW.
- Display at Riverland Field Days, Sept 2002.
- Presentations at four Riverland Cittgroups and the Waikerie Ag Bureau, Oct 2002.
- Presentation at IPM course, Loxton, Oct 2002.

2003-04

- Display at Riverland Field Days, Sept 2003.
- Presentations at two Riverland Cittgroups, Oct 2002.

PAPERS and ARTICLES

2000-01

- Three colour posters produced (Poster 1 - Adult KCT identification; Poster 2 - Larval KCT identification; Poster 3 - The key to controlling Kelly's citrus thrips is finding them early.)
    - A4 colour, laminated copies of the posters available to growers for $3 each.
- *Murray Pioneer* and *The Grower* article, Aug and Sept 2000: 'The key to controlling Kelly's citrus thrips is finding them early'.
- *The Loxton News* article, Dec 2000: 'Searching for pest solution'.

- *CGSA News* article, Nov 2000: ‘The key to controlling Kelly’s citrus thrips is finding them early’.
- *OpenGate (Stock Journal* insert) article, June 2001: “Scientists target Kelly’s citrus thrips”.

2001-02
- *Sunraysia Citrep and Australian Citrus News,* Sept 2001: ‘The key to controlling Kelly’s citrus thrips is finding them early’. This article also sent to The Loxton News, Murray Pioneer and Sunraysia Daily.
- *SA Grower Magazine* article, Nov 2001: KCT management and the HAL research project.
- *ACN* article, Dec 2001: ‘Kelly’s citrus thrips – a challenging new citrus pest’.
- *Citrus Insight* article, Jan 2002: ‘Changing environment may favour thrips’.
- *The Loxton News* article, March 2002: ‘Sheridan's research rewarded’.
- "Talking Thrips in Citrus" Newsletter produced in May 2002, and distributed at the Mildura Field Days and in the CGSA newsletter.
- *Countryside Quarterly* article, June 2002: ‘Sheridan's research helping eradicate Kelly's citrus thrips’.
- *Open Gate* (*Stock Journal* insert) article, June 2002: ‘Thrips researchers look for flaw in Kelly's armour’.

2002-03
- *Murray Pioneer* article, Sept 2002: ‘KCT early warning’.
- Issue 2 of “Talking Thrips in Citrus” Newsletter distributed in Oct 2002.
- *CGSA News* article, Nov 2002: ‘The key to controlling Kelly’s citrus thrips is finding them early’.
- *Citrus Insight* article, Jan 2003: ‘KCT, an export issue’.
- Issue 3 of “Talking Thrips in Citrus” Newsletter distributed in July 2003.

2003-04
- *Citrus Insight* article, Jan 2004: ‘Improving the management of Kelly’s citrus thrips’.
- ‘KCT Biology and Management’ Factsheet draft prepared, May 2004.
- Poster paper (‘The Biological Control of Kelly’s Citrus Thrips in Australian Citrus Orchards’) prepared for International Congress of Entomology, June 2004.
- Glossy colour ‘fact-sheet’ on KCT biology and management drafted, June 2004.

RECOMMENDATIONS

Industry recommendations

1. The citrus industry should encourage Syngenta and Dow Agrosciences to **swiftly advance the registration applications** for the use of thiamethoxam and spinosad respectively for control of KCT in citrus.

2. An **IRM rotation strategy**, similar to the cotton and Brassica vegetable industry strategies and based on an understanding of the cross-resistance relationships between the KCT insecticides, must be devised and implemented as soon as new chemistry is registered for KCT control. This is essential to ensure long-term, effective insecticidal control of KCT. Expert assistance should be sought to assist the interested parties (the citrus growers and their representative bodies, the insecticide manufacturers and the insecticide distributors) with concept development (refer to pages 53-54 of this report) and the planning negotiations.

3. Given that the singular reliance on insecticidal control of KCT will be unsustainable even if an IRM strategy is adopted, the citrus industry should support efforts to **enhance the contribution of biocontrol agents to KCT control**.

4. The citrus industry should critically **appraise the potential benefits of multi-fan (lower-spray volume) spray technology** (as has occurred, with dramatic spray efficacy, labour and chemical cost benefits, in the Australian viticultural industry). In addition to the benefits to the biocontrol of KCT by predatory mites, the improved spray coverage and efficacy that such multi-fan spray technology is likely to provide would benefit citrus IPM generally by allowing the industry to replace many 'hard' insecticides with IPM-compatible 'soft' compounds.

Research recommendations

Further research is required in four key areas

1. **Insecticide resistance studies**
   Important questions requiring answer are (i) what are the cross-resistance patterns of the insecticides used for KCT control? (ii) what is the stability of the resistance to chlorpyrifos and methidathion in KCT populations? and (iii) how much KCT population gene flow is occurring between neighbouring citrus orchards? (Answers to (i) and (ii) are required for the design of a sound IRM rotation strategy, and if the answer to (iii) indicates that sufficient amounts of population exchange are occurring, then a 'refuge' strategy, whereby insecticides that are used for KCT control are not used in blocks that are low risk from KCT damage (eg. mandarins and juicing Valencias), would slow the development of resistance in nearby high-risk blocks (eg. navel oranges, lemons, grapefruit).)

2. **Insecticide field trials**
   Different rates and spray volumes of spinosad+Brella™ should be trialled to determine the most cost-effective treatment for KCT control, and higher rates of the soil-applied contact insecticides bifenthrin and fipronil should be tested.

3. **Enhancement of KCT biological control**
   . Further research is needed to answer several key questions, namely:

- Will the raising of organic carbon (OC) levels in Riverland-Sunraysia citrus soils cost-effectively enhance KCT predator activity and biocontrol, and thereby reduce KCT packout losses? If so, what is the minimum OC level required?
- Can increasing the grass-pollen production of Riverland-Sunraysia citrus ground covers promote KCT predator activity and biocontrol? If so, which grasses are most beneficial?
- What impacts do insecticides (chlorpyrifos, methidathion, methomyl, aldicarb, spinosad and thiamethoxam) and commonly-used herbicides have on KCT predator activity and biocontrol in Riverland-Sunraysia citrus?

4. Irrigation modification

Further research is recommended to determine whether irrigation restriction in spring can be managed to reduce KCT pupal survival (and subsequent crop attack) without adversely affecting tree health.

APPENDIX A: AN EVALUATION OF THE PREDACIOUS BEHAVIOUR OF MITES AS PREDATORS OF KCT

BACKGROUND

This portion of the project evaluated the predatory behaviour of a selection of mites with the intention of determining their value as biological control agents of Kelly's citrus thrips (KCT)(*Pezothrips kellyanus*). Previous research was focussed on locating potential biological control agents for KCT, but no natural enemies were found in the citrus canopy (Baker *et al*., 2000). KCT pupate exclusively in the soil (Baker *et al*., 2000). This exposes pupal KCT to the soil biota for a period between two and eleven days. Thus predators that reside in the soil can attack and kill them. Davidson (2001) reported on research by Colloff, who linked a high diversity of soil-dwelling predatory mites to the reduced pest status of KCT in some orchards. Thus there is some evidence that predators may suppress KCT in the soil.

OBJECTIVES

It was the primary aim of this research to compare the activity of various soil dwelling mites as predators of KCT. The research focused on questions pertaining to the biology and activity of predatory mites:

- Is there a significant difference in the functional responses to prey density among mite species?
- Is soil type or composition an important factor influencing the efficiency of predatory mites?

MATERIALS AND METHODS

Mites from seven laboratory cultures were evaluated in the course of this research project (Table 1). Trials had demonstrated that each of the predators could complete its development with pupal KCT as the sole food source. For convenience, a unique letter was used to identify each mite culture was used. Cultures of mites A, B, D, E, and F were initially extracted from the soil at Grant Brown's organic citrus orchard near Loxton, South Australia and reared on a diet of nematodes. Mite K was first found in Queensland by Irene Vanninen and has been used in biological control experiments on other thrips species (Vanninen, 2002). Mite W was found in a culture of KCT at the Waite Campus, Urrbrae, South Australia. It was accidentally introduced into the culture, but its origin is unknown.

Methods for culturing predatory mites and their nematode prey were based on instructions provided by Irene Vanninen (Agrifood Research Finland [MTT], Jokioinen, Finland.). Cultures of predatory mites were maintained on a diet of that consisted of a mixture of two species of nematodes, *Panagrellus siluridae* (Panagrolaimidae) and *Rhabditis spp.* (Rhabditidae) (Barbour 2003).

Rearing and experimental arenas were constructed using plastic containers (Polarpak GS125, Genfac Plastics Pty. Ltd., Melbourne, Australia) with a diameter of 60 mm, height of 45 mm and a volume of 125 ml. Tap water was added to a mixture of plaster of Paris (9 parts) and carbon powder (1 part) to create a thick, well-mixed liquid. This mixture was then poured into the plastic containers to a depth of approximately 5 mm. The plaster/carbon mixture was allowed to set until hard and cool. Lids for the containers had an 8 mm diameter hole covered with fine mesh voile to allow for gas and moisture exchange. To prepare the arenas for use, tap water was added until an excess was present on top of the substrate. The substrate was allowed to absorb the water for 15 minutes before the excess water was removed by blotting the substrate with paper towel.

Table 1. Identities of predatory mites. Due to the death of all mite B samples in culture, no identification was possible. All other mite specimens were initially identified by Peter Crisp (The University of Adelaide, Australia) and subsequently checked by David Walter (The University of Queensland, Australia) (Table 3.2).

Mite	Family	Identity	Notes
A	Laelapidae	*Dendrolaelaps spp.*	Mite A and D have been identified as the same species
B	[unknown]	[unknown]	
D	Laelapidae	*Dendrolaelaps spp.*	
E	Rhodacaridae	*Protogamasellopsis spp.*	*P. corticalis* or a close relative
F	Parasitidae	[unknown]	
K	Laelapidae	*Geolaelaps aculifer*	
W	Ascidae	*Lasioseius spp.*	*L. dentatus* is most likely - a number of key diagnostic features were compared with a known *L. dentatus*

The functional responses of predatory mites from seven laboratory cultures were examined in experiments. Prey densities ranging from 1 to 50 KCT were tested in the experiments. Late second instar larvae or early pro-pupal KCT were collected from rearing containers. The KCT and vermiculite were then transferred into a prepared experimental arena. Clean vermiculite was added to obtain a quantity of approximately 4 cm^3, thereby providing a thin uniform layer of vermiculite. One predatory mite that had been starved for 24 hours was added to the arena. Predation arenas were stored in cardboard boxes during their incubation period so the mites were held in near darkness at 25°C. Three sprays of water from an atomiser increased the humidity inside the box. After 48 hr the number of surviving KCT was counted.

Nonlinear regression (JMP version 4.0.2, SAS Institute Inc.) was used to fit the data to Holling's Disk Equation (Holling 1959). The regression estimated both the handling time (T_h) and instantaneous rate of discovery (a). The initial estimate of both 'a' and 'T_h' was set as 0.1.

Outlying data points were visually identified on the initial functional response plots and excluded from the data set. These have been identified by open circle data points on the functional response graphs. To obtain better regression fits, the nonlinear regression was repeated using the reduced data set, which excluded the outlying data points.

Individual curves were compared to determine whether the functional responses of the species could be distinguished statistically. The data from two mite species were analysed first separately and then as pooled data. To determine whether an entire curve was statistically different from another curve, the F statistic was calculated with the equation
:

$$F = \frac{(SSE_{combined} - SSE_{separate}) / (DF_{combined} - DF_{separate})}{SSE_{separate} / DF_{separate}}$$

$SSE_{separate}$ is the separate error sum of squares and $DF_{separate}$ is the error degrees of freedom. The values for $SSE_{separate}$ and $DF_{separate}$ were calculated by adding the two SSE and DF values from the individually fitted curves. The probability value to test the null-hypothesis was calculated in Microsoft Excel with the formula:

$$= FDIST('Fvalue', 2, 'DF_{separate}')$$

A total of 21 curve comparisons were performed. To adjust for the inherent error in comparing a large number of curves, the sequential Bonferoni procedure was applied (Rice 1989). First the curve comparisons were ranked from the least significant, ranked 1, to the most significant, ranked 21. Each comparison was tested for significance at a level of 0.05/rank. Starting at the most significantly different curves, once the first curve comparisons that did not pass the significance test were found, all curves ranked lower were automatically rejected as insignificantly different.

RESULTS

All predatory mites exhibited a type 2 functional response, which is typical of many invertebrate predators (Figure 1). The instantaneous rate of discovery was greatest for mite K (*Geolaelaps aculifer*), which indicates this species has the highest searching rate at low prey densities (Table 2). Mite F (a parasitid mite) had the shortest handling time, which indicates that it can consume the greatest numbers of prey when prey density is extremely high. Statistical analysis indicated that the functional response of mite K differed from all other species (Table 3).

Table 2. Summary of functional response parameters and statistics

		Parameter Estimates		Regression Statistics	
Mite	n	a[1]	T_h[2]	SSE[3]	DFE[4]
A	105	0.3710	0.2138	125.7100	103
B	90	0.1359	0.1198	166.0729	88
D	55	0.2139	0.1207	70.4526	53
E	98	0.2101	0.1972	255.7456	96
F	105	0.1285	0.1157	262.3881	103
K	109	0.4764	0.1592	247.4357	107
W	128	0.2470	0.1089	348.8056	126

[1] Instantaneous rate of discovery (day^{-1})
[2] Handling time (days)
[3] Error sums of squares for nonlinear regression
[4] Error degrees of freedom for nonlinear regression

Table 3. Summary of statistical comparisons among functional response curves.

Mite Species	W	K	F	E	D	B
A	*	*	*	*	NS	*
B	*	*	NS	NS	*	
D	NS	*	*	NS		
E	*	*	NS			
F	*	*				
K	*					

* = $P < 0.05$; NS = No Significant Statistical Difference.

DISCUSSION

Each of the predatory mites consumed pupal KCT. When KCT densities were high, the mites could consume 3-5 pupal thrips per day, or possibly more. Consumption rates were lower at lower thrips densities. These results confirm that predatory mites can kill pupae of KCT. Several mites probably feed on this pest in citrus orchards, since four of the species tested were collected from a Riverland citrus orchard. These mites were reared on nematodes, so none of them is a specific predator of thrips pupae. Hence their significance as thrips predators cannot be established in the simple arenas that were used here. Preliminary experiments (Barbour 2003) indicated that some of the predators (Species A, E, F, K, W) did feed on pupal thrips when other potential prey species were present in soil.

Figure 1. Functional response curves for six species of predatory mites in experimental arenas. Outlying data points (open circles) were excluded from statistical analysis.

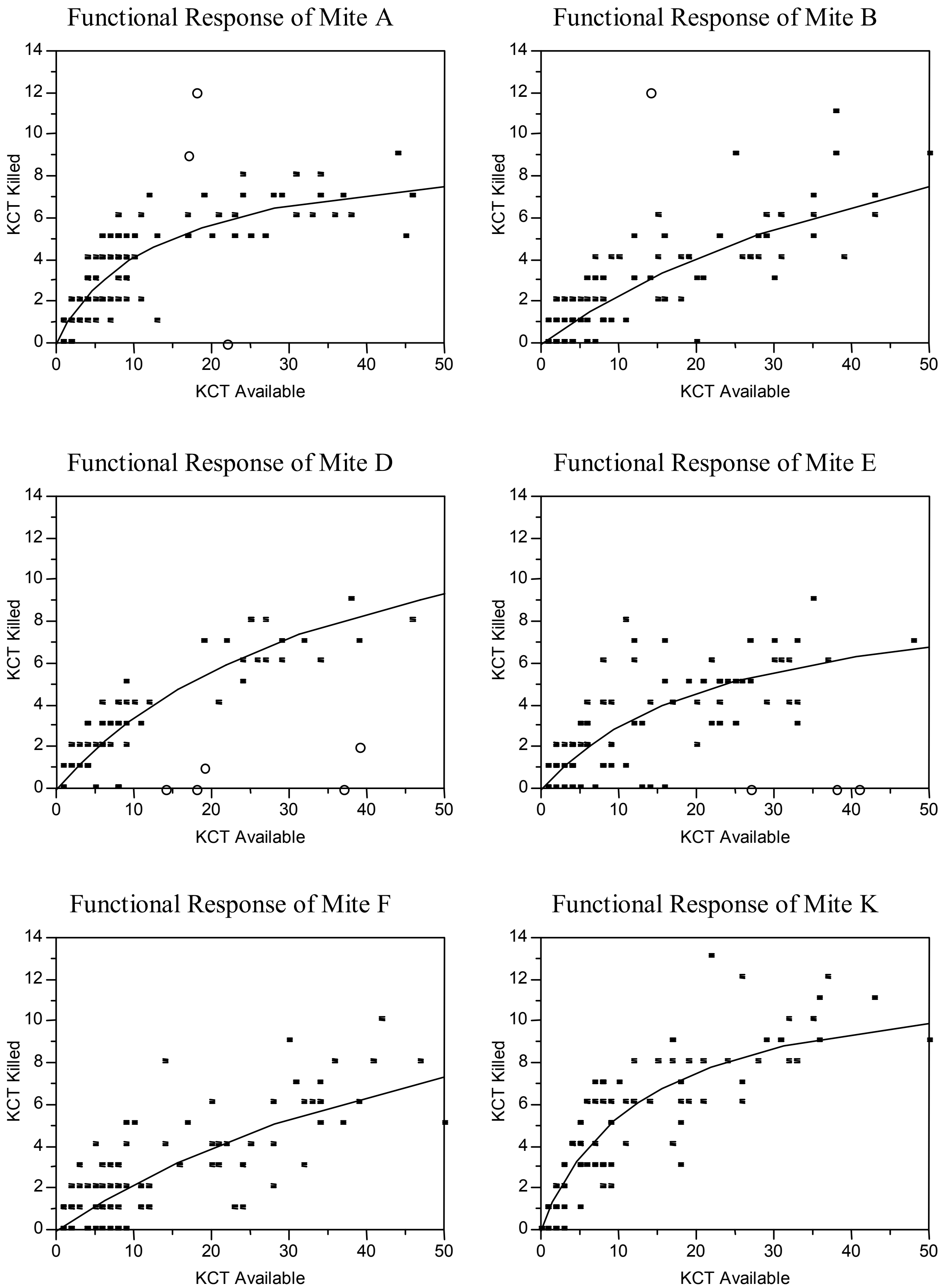
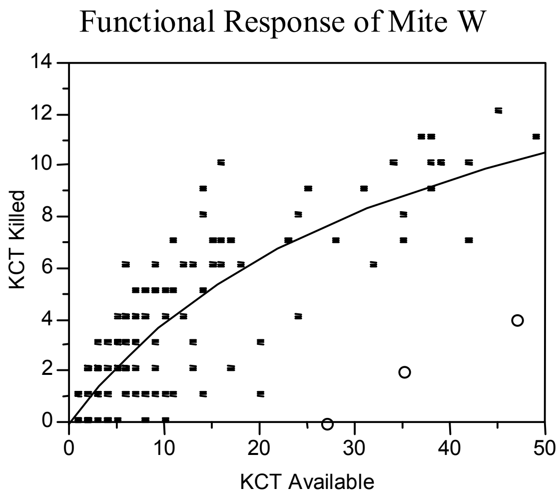


Figure 1 (continued). Functional response curves for six species of predatory mites in experimental arenas. Outlying data points (open circles) were excluded from statistical analysis.



ACKNOWLEDGMENT

This section of the report was extracted and partially rewritten from:

Barbour, D. 2003. An evaluation of the predaceous behaviour of mites as predators of Kelly's citrus thrips (*Pezothrips kellyanus*). Honours thesis, the University of Adelaide. 64 p.

LITERATURE CITED

Baker, G., Jackman, D. J., Keller, M., MacGregor, A., & Purvis, S. (2000). Development of an Integrated Pest Management system for thrips in Citrus (Adelaide, SARDI).

Davidson, S. (2001). Dirty Deeds, Ecos *106*, 12-14.

Holling, C. S. (1959). Some Characteristics of Simple Types of Predation and Parasitism, The Canadian Entomologist *91*, 385-398.

Vanninen, I. (2002). Unpublished poster presentation at the First International Symposium on Biological Control of Arthropods, Honolulu, Hawaii, USA, 13-18 January 2002.

APPENDIX B: SELECTED EXAMPLES OF EXTENSION PRODUCTS


Who does the damage?


Who does the damage?

Natural Resources and Environment

AGRICULTURE

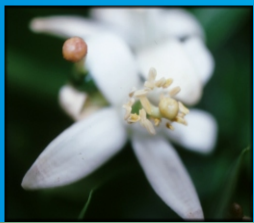
RESOURCES

CONSERVATION

LAND MANAGEMENT

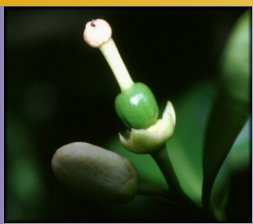
The key to controlling Kelly's citrus thrips is finding them early

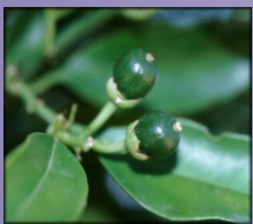
Darryl Jackman[3], Mike Keller[3], Greg Baker[2], Alison MacGregor[1] Sheridan Purvis[1]
[1]Agriculture Victoria, SARDI, [3]University of Adelaide

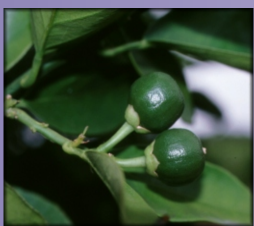


You may see many adult KCT in flowers

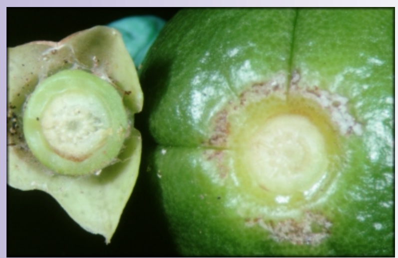
Don't spray yet!







Monitor Navels twice weekly from petal fall to calyx closure



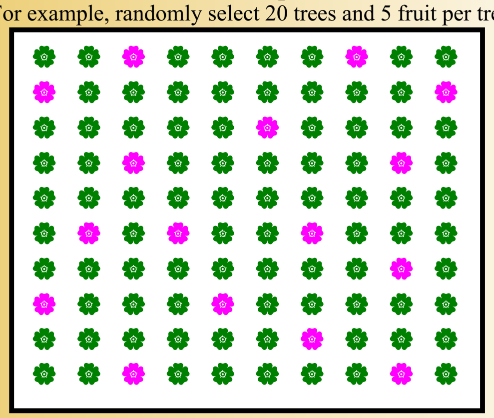
Use a 10 hand lens to look under the calyx for KCT larvae

Continue to monitor weekly for KCT larvae after calyx closure until January

The key: look under the calyces of at least 100 fruit per block.

For example, randomly select 20 trees and 5 fruit per tree

Spray when you see KCT larvae on 5% of fruit



It is important to achieve good spray coverage at tree tops where damage is often highest

SARDI — SOUTH AUSTRALIAN RESEARCH AND DEVELOPMENT INSTITUTE

HRDC — Horticultural Research & Development Corporation

RIVERLINK — Linking Horticultural Centres in Sunraysia - Riverland

AGRICULTURE VICTORIA — A business of the Department of Natural Resources and Environment

Victoria — The Place To Be

Who does the damage?

Larvae of Kelly's citrus thrips (KCT) feed under citrus fruit calyces causing a 'halo' of scurfing.

The high risk period is from petal fall to calyx closure, although this can extend through to January

Kelly's citrus thrips larvae

- Range in colour from pale yellow to bright orange
- 1 mm long
- 10 hand lens will aid identification
- found under calyces, remnant petals and between touching fruit

Don't confuse KCT larvae with…

Mealybug crawlers

- Often found under the calyx of citrus fruit
- Associated with sooty mould problems in citrus

Apple dimpling bug nymphs

- Found under the calyx of citrus fruit
- Considered beneficial in citrus
- Pest in apples, pears and nashi

Darryl Jackman[3], Mike Keller[3], Greg Baker[2], Alison MacGregor[1] Sheridan Purvis[1]
[1]Agriculture Victoria, SARDI, [3]University of Adelaide

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Identification of Thrips in Citrus

Adult (2mm)

Kelly's citrus thrips

Pezothrips kellyanus

- **Only thrips causing halo marks on citrus**
- **Found year round in citrus canopies**
- **Larvae white- yellow**
- **Adults are black**
- **Wings parallel and dark with a small clear band at top**
- **Does not live in covercrops or weeds of citrus orchards**

Larva (<1mm)

Other thrips found in citrus orchards THAT DO NOT cause damage:

Predatory thrips (2mm)

Haplothrips victoriensis

- Adults are black
- Wings overlap and are clear, creating a silver line along body
- Found in citrus canopy, covercrops and weeds

Flower thrips (silverbacks) (1mm)

Frankliniella schultzei

- Adults are small and black
- Wings are parallel and clear, creating two silver lines along body
- Found in citrus canopy, covercrops and weeds

Plague thrips (1mm)

Thrips imaginis

- Adults are small and pale yellow – brown in colour
- Found in citrus canopy, covercrops and weeds

- **When to identify:-**
  - Flowering to calyx closure.
- **Where:-**
  - On flowers, under calyx
  - Post calyx closure- lemons
  - On the outside of mature fruit (late in the season – e.g lengs navels and tangelos)

Prepared for Riverlink, by Sheridan Purvis, SARDI and Design by Gregory Moulds, NSW Agriculture.

Talking Thrips *in citrus*

Produced by Sheridan Purvis, SARDI

May 2002 Issue 1

This quarterly newsletter has been produced to keep you informed of the latest research on Kelly's citrus thrips (KCT) management. The research is funded by Horticulture Australia and involves scientists from SARDI and the University of Adelaide.

Due to the mild season things seem a little quiet on the KCT frontline in many growers orchards during 2001-02, but behind the scene researchers have been hard at work trying to find a weakness in the battle armour of KCT.

Get familiar with KCT

KCT is the little thrips pest that causes halo marking on citrus fruit, with which most of you are all too familiar. KCT larvae cause scarring as they feed on the developing fruitlet, punching open the rind cells to suck out the contents.

- KCT larvae are very tiny, use a 10× hand lens to see them clearly
- KCT larvae range in colour from pale yellow to orange.
- Adult KCT are small and black.

KCT larva KCT adult

There are three **other** species of thrips that can also be found in citrus orchards but **none of these cause any damage**, in fact one is a 'good bug' because it feeds on other insects. Hence correct identification is very important!

A4 colour laminated posters showing how to correctly identify and monitor for citrus thrips are available from Kym Thiel (0417 800 937), Peter Morrish (03 5021 1890) or Sheridan Purvis (08 8595 9100).

Is your block attractive to KCT – or not? And why?

Over the years pest scouts have noticed that some citrus blocks just never seem to have a problem with KCT. Why is this?

Research is underway at 11 Riverland citrus blocks to identify biological and environmental factors that influence KCT activity. The sites were selected based on their history of KCT pressure – either high or low. Each stage of KCT life cycle is monitored. Fruit samples are collected to record number of KCT adults and larvae in the tree canopy. Water-filled pan traps are set under the dripline of the tree to catch KCT larvae as they drop to the ground to pupate, and sticky emergence traps are also set under the tree dripline to record the number of adult KCT emerging from the ground. Weather data, covercrop composition and management and spray programs are also being recorded for each site.

This intense monitoring is designed to provide important information on trends in KCT abundance and their correlation with orchard environmental and management factors.

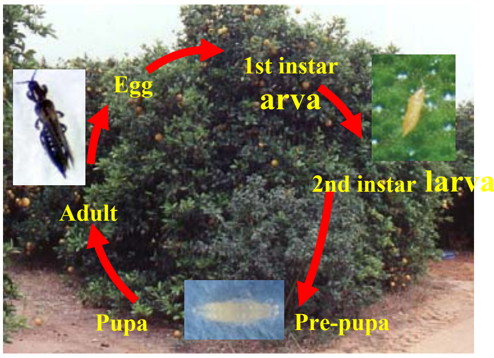
Taking the fight down under!

KCT pupate in the leaf litter and soil beneath the citrus canopy. Healthy soil abounds with small organisms, many of which are predacious and beneficial. Surely some of these critters must find inactive plump KCT pupae just too good to pass by!

To find out what is feeding on KCT pupae, we are compiling a list of soil dwelling predators at the 11 sites mentioned above. We set pitfall traps – simply plastic drinking cups set into the soil - under the dripline of the tree canopy. Arthropods moving around the orchard floor fall into a cup and are unable to climb out. From have identified a number of predacious species of ants, rove beetles, carabid beetles and spiders. Soil samples have also been collected and taken back to the laboratory where predatory mites are extracted for identification. We will then test which of these predators will feed on KCT pupae.

The effect of *metarhizium* fungi on pupating KCT has also been investigated with the assistance of Richard Milner from CSIRO in Canberra.

KCT life cycle



Are KCT developing resistance to chlorpyrifos?

Populations of KCT collected from Riverland orchards were found to have very low susceptibility to chlorpyrifos in laboratory experiments recently conducted at the Waite Institute. This is a major concern for citrus growers due to the limited range of insecticides available for KCT control. It is vitally important for all growers to implement a resistance management system for the long term control of KCT.

Effective spray timing and coverage is important because the number of sprays applied per season must be kept to a minimum. To do this, yet still produce clean fruit, a grower must regularly monitor their blocks for pests. A control spray should be applied as soon as the pest threshold is reached: for KCT this is 5-10% fruit infested. Give your spray the best chance to work by taking the time to hit the pest before it has a chance to build up numbers and ensure good coverage of the whole tree.

On the horizon – a new weapon in the fight against KCT

New insecticides for control of KCT have been trialed in both the laboratory and the field over the past 2 seasons. While damage assessments are yet to made this season, 1 or 2 treatments are showing some promise with control equal to and above that currently obtained by chlorpyrifos. This is great news for citrus growers as a new insecticide will not only achieve better KCT control but will also help manage KCT resistance to existing insecticides by alternating the use of different insecticide groups. Further research will be required to assess the impact of these new insecticides on other pests and beneficials in the citrus orchard.

Acknowledgements

The Project thank David Ingerson, Mr and Mrs Radomi, Joel Pickering, Peter Fechner, Graham McInness, Grant Brown, Mark McLean, Tim Whetstone and Yandilla Park Ltd. for generously providing us with access to their properties for our KCT field studies.

We also thank Peter Walker and the Wylie family for generously providing us with a KCT insecticide trial site for the past two seasons.

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Talking Thrips *in citrus*

Produced by Sheridan Purvis, SARDI

October 2002 Issue 2

This newsletter has been produced to keep you informed of the latest research on Kelly's citrus thrips (KCT) management. The research is funded by Horticulture Australia and involves scientists from SARDI and the University of Adelaide.

2001-02 Insecticide Trial Results

One of the aims of our project is to identify new insecticides and develop an insecticide management plan that:

a) provides good KCT control

Harvest damage assessments have now been completed for the 2001-02 Insecticide Field trial. Results are encouraging with one treatment obtaining significant control of KCT damage.

Plans are now in place to conduct rate and application trials to get Product X on the road to registration.

b) minimises resistance

Product X is from the neo-nicotinoid chemical group. To minimise insecticide resistance it is important that growers alternate the use of different chemical groups.

c) mininises negative impact on beneficials

Tests were conducted in the laboratory to determine the contact toxicity of Product X to the beneficial *Aphytis melinus.* Field rates were hand sprayed to run-off onto citrus foliage. Tests were conducted with leaves collected 0, 3, 7 and 13 days after treatment. The mortality of *A. melinus* was assessed after 24 hours exposure to treated leaves.

Unfortunately Product X proved to be significantly more toxic to *A. melinus* than chlorpyrifos, with over 80% mortality on 7 day old residues.

With the citrus industry in need of some relief from KCT, the planned field trials with Product X will go ahead, albeit with caution.

Are KCT developing resistance to chlorpyrifos?

Populations of KCT collected from Riverland orchards were found to have very low susceptibility to chlorpyrifos in laboratory experiments recently conducted at the Waite Institute. This is a major concern for citrus growers due to the limited range of insecticide groups available for KCT control.

Taking an alternative route!

The effect of *Metarhizium* (fungal pathogen) on pupating KCT has also been investigated with the assistance of Richard Milner from CSIRO in Canberra.

Laboratory tests have determined that isolate FI-1248 does kill KCT. Further tests in the lab have been initiated and subject to the results, field trials will be conducted later this season.

Population sampling at 5 Riverland sites

KCT populations are being monitored at 5 Riverland sites. Each stage of the KCT life cycle is monitored with pan traps (catching larvae dropping to ground), emergence traps (catching emerging adults) and flower/fruit samples (records the number of KCT adults and larvae in canopy). The aim of this long term study is to a) document seasonal patterns of infestation, b) estimate levels of pupal mortality and c) compare sites and determine whether infestations are affected by site-specific factors.

Soil-dwelling predators

Two sampling methods (pitfall traps and Berlese extraction funnels) have been used to record soil-dwelling predator species at each of the 5 Riverland sites. Potential predatory mites have been collected and reared in the laboratory. Darryl Barbour, Honours student (Uni of Adelaide), will use the mites in feeding arena experiments with KCT pupae.

Arboreal Mite Predators
Sticky bands placed on or around plant parts (primarily outer limbs) have been used at the 5 Riverland sites to determine potential predatory mite species in the citrus canopy.

We are currently attempting to collect and rear some of the arboreal predatory mites to test in feeding area experiments with KCT larvae.

Remember!
Effective spray timing and coverage is important because the number of sprays applied per season must be kept to a minimum. To do this, yet still produce clean fruit, a grower must regularly monitor their blocks for pests. A control spray should be applied as soon as the pest threshold is reached: for KCT this is 5-10% fruit infested. Give your spray the best chance to work by taking the time to hit the pest before it has a chance to build up numbers and ensure good coverage of the whole tree.

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Talking Thrips
in citrus

Produced by Sheridan Purvis, SARDI

June 2003, Issue 3

This newsletter has been produced to keep you informed of the latest research on Kelly's citrus thrips (KCT) management. Issue 3 highlights results from trials conducted over the past eight months. The research is funded by Horticulture Australia and involves scientists from SARDI and the University of Adelaide.

2002-03 Insecticide Trial Results

Actara™ (previously called Product X) Registration Update

It is anticipated that HAL-funded trial data combined with Syngenta trial data will be sufficient for product registration. Subject to residue results (nil residues allowable for US market) Syngenta expect registration by late 2004/early 2005.

This is good news with regard to insecticide resistance management. However, Actara™ is a broad-spectrum insecticide, which our laboratory trials have shown to be significantly more toxic to the beneficial *Aphytis melinus* than chlorpyrifos. Therefore Actara™ will need to be used strategically to avoid secondary pest problems.

Spinosad/Brella™ Trial

The "softer" insecticide spinosad (Success™, Dow Agrosciences), which has previously performed poorly, was re-trialled this season in combination with the Caltex petroleum oil Brella™. Results to date suggest spinosad performs substantially better when used with Brella™ (Figure 1). Harvest damage assessments are yet to be conducted.

Soil-applied Contact Insecticides

The strategic use of soil-applied contact insecticides to control pupating KCT is being explored. It is anticipated that this control method will be used only in high-risk areas, such as lemon orchards, in early spring to reduce the spring peak in KCT populations.

Insecticide Resistance

Our research has identified 10-250 fold reductions in organophosphate susceptibility in KCT populations in some Riverland orchards. To provide effective long-term control of KCT, insecticides from other chemical groups will need to be incorporated

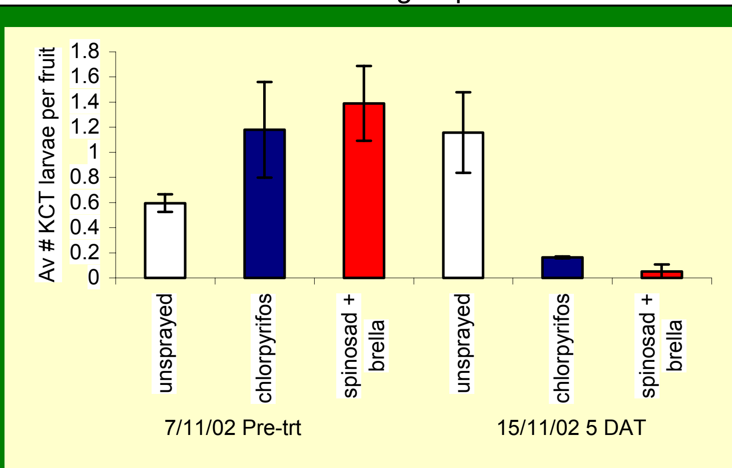

Figure 1. Spinosad + Brella™ Efficacy trial. KCT larvae density significantly reduced 5 days post treatment application in a Riverland navel orchard.

into an industry wide resistance management strategy. A brief questionnaire is included to help us assess KCT control practices and difficulties across the Riverland. Your cooperation in filling this in and returning in the pre-paid envelope will help us to help your industry.

KCT pupal survival in soil

KCT population data has been collected from 5 Riverland orchards over the past 2.5 years. Each stage of KCT life cycle was monitored. Initial analysis of data indicates a substantial difference in the survival of KCT pupae (soil-dwelling life stage) amongst the 5 orchards. Low pupal survival appears to be associated with low KCT pest status and high predatory mite incidence. Fieldwork has been initiated to further investigate the causes of low pupal survival.

Soil-Dwelling Predatory Mites

Over 40 species of predatory mites have been identified from soil samples collected from 8 Riverland orchards. Darryl Barbour (University of Adelaide, Honours student) has evaluated seven of these species in feeding trials using KCT pupae. Each species is able to develop and reproduce on a diet of KCT alone.

Figure 2. Predatory mite attacking KCT pupae in feeding trial.

Figure 3. Magnified image of slide-mounted predatory mite.

Suction Trap Study of KCT Movement

Suction traps were positioned at 6 sites in the Riverland to investigate the source of the spring peak in KCT populations observed in navel orchards. Two traps were each positioned within a citrus orchard, near citrus orchards (300m) and well away from citrus orchards (5-17km). Traps were operated 24/day during spring-early summer. The trapping data suggest that there is some localised movement (ie orchard to orchard), but there is no evidence of any large-scale long distance movement. That is, the KCT in any orchard in October-November originate within the orchard itself or its near neighbours.

Acknowledgements

The Project team thank Peter Fechner, Graham McInness, Grant Brown, Mark McLean, James Altmann and Yandilla Park for generously providing us with access to their properties for our KCT field studies. We also thank Peter Walker and Dennis Battams for each providing us with a KCT insecticide trial site.

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THE BIOLOGICAL CONTROL OF KELLY'S CITRUS THRIPS IN AUSTRALIAN CITRUS ORCHARDS

GJ Baker[1], MA Keller[2], P Crisp[2], S Purvis[1], D Jackman[1] and D Barbour[2]

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[2] *Plant and Pest Science, School of Agriculture and Wine, University of Adelaide, SA 5005.*

Introduction

Kelly's citrus thrips (KCT) (*Pezothrips kellyanus* (Bagnall) (Thysanoptera: Thripidae) emerged in the 1990's as a serious citrus pest in southern Australia (Figs. 1-3). Information about KCT biology and ecology has been scarce, and in Australia the management of KCT has been limited to the use of several insecticides.

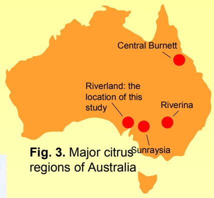

Fig. 3. Major citrus regions of Australia

Objectives

1. To identify soil-dwelling and arboreal natural enemies of KCT (a soil pupating species) in Australian citrus orchards.
2. To evaluate the potential of these natural enemies to regulate KCT populations in a commercial citrus IPM system.

Fig. 1. Adult KCT feeding on citrus flower.

Fig. 2. KCT scurfing damage on mature fruit.

Methods

One organic and two conventional-production navel orange orchards were chosen. The organic and one of the conventional (Conv. 1) orchards had a known history of low KCT abundance, and the other conventional orchard (Conv. 2) a history of high KCT abundance.

KCT abundance and pupal mortality were estimated using water traps and Tanglefoot® - coated ground traps placed under the canopy.

Soil-dwelling predators were sampled with pitfall traps and Berlese funnel extraction of soil-litter cores. Arboreal natural enemies were sampled by collecting plant samples, foliage beating and using sticky bands on twigs and fruits. KCT adults were dissected for the presence of nematodes.

Ground cover diversity and composition was quantified using the Levy Point quadrat method, and soil organic carbon content was measured using the modified Walkley-Black method.

Key Findings

1. KCT abundance and survival – KCT abundance is directly related to the survival of the soil-dwelling life-stages of KCT at the three study orchards (Fig. 4).

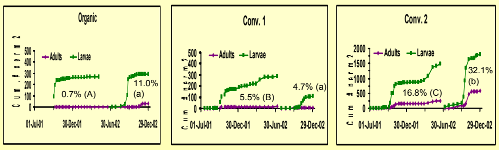

Fig. 4. The cumulative number of KCT 2nd instar larvae captured in water traps and KCT adults captured in sticky emergence traps placed under the canopy of the 3 study orchards, 2001-03. The mean % survival of KCT in the soil in each year at these 3 sites are presented. For each year, means followed by different letters are significantly different, ANOVA (*P*<0.05).

2. Soil-dwelling Predators – The abundance and diversity of predatory mites was greatest at the Organic orchard and least at Conv. 2 (Fig. 5).

Several of the soil-dwelling predatory mite species were tested in lab feeding trials, where they successfully developed and reproduced on a diet of KCT propupae.

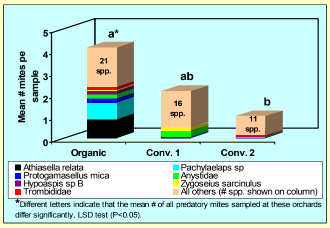

Fig. 5. The mean number of predatory mites per sample at the 3 study orchards, 2001-03. Several spp. (particularly *Athiasella relata* (Ologomasidae), *Pachylaelaps* sp. (Pachylaelapidae) and *Protogamasellus mica* (Ascidae)) were notably more abundant at the organic orchard.

Differences between the 3 study orchards in the abundance and species composition of soil-dwelling predatory insects and spiders were not significant (ANOVA, *P*>0.05).

None of the measures of ground-cover plant composition and diversity at the 3 orchards correlate with the observed differences in survival of soil-dwelling KCT and abundance of predatory mites.

Soil organic carbon (OC) content positively correlates with predator abundance and KCT mortality. (5.3, 2.5 and 1.5 g C/ kg soil at Organic, Conv. 1 and Conv. 2 respectively.)

3. Other Natural Enemies – Generalist predatory insects (mirids, chrysopids, coccinellids, *Haplothrips* sp.) and the eulophid parasitoid *Ceranisus menes* were present in each orchard at similar low densities.

No parasitic nematodes were found in >500 dissected KCT adults.

Arboreal predatory mites, which belonged to four families (Erythraediae, Phytoseiidae, Anystidae and Stigmaeidae), were more abundant at the organic orchard (Table 1).

Table 1. Mean # of arboreal predatory mites per sticky band sample. Means followed by same letter are nsd, Tukey HSD test (P>0.05).

Orchard	Mean # of predatory mites
Organic	33.0 a
Conv. 1	19.1 b
Conv. 2	11.8 b

Conclusions

There are complexes of soil-dwelling and arboreal mite species that are either demonstrated or likely predators of KCT in Australian citrus orchards. Negative correlations between the abundance of KCT and the predators, and between the survival of soil-dwelling KCT and the abundance of the soil predatory mites suggest a causal link between predatory mites and low thrips numbers.

Further research is underway to develop an effective IPM system which optimizes the contribution of these mite predators.

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