Effects of insect growth regulators on *Chilocorus nigritus* (Fabricius) (Coleoptera: Coccinellidae), a non-target natural enemy of citrus red scale, *Aonidiella aurantii* (Maskell) (Homoptera: Diaspididae), in southern Africa: evidence from laboratory and field trials

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Chilocorus nigritus (Fabricius) is one of the major coccinellid predators of the citrus pest *Aonidiella aurantii* (Maskell) in southern Africa. Laboratory and field experiments were carried out on eggs, larvae and adults of this ladybird to determine the effects of three insect growth regulators (IGRs) used against citrus pests in the region. Two chitin synthesis inhibitors, buprofezin and teflubenzuron, and a juvenile hormone analog, pyriproxyfen, were applied to *C. nigritus* populations at the recommended dosages. Mortality and development of egg and larval stages, as well as mortality and fecundity of the adults were recorded. Laboratory experiments indicated that, of the three IGRs tested, buprofezin was the most detrimental compound, especially to larval stages, irrespective of whether the larvae were fed IGR-treated *A. aurantii* directly or sprayed with IGRs. Immediate larval mortality from pyriproxyfen and teflubenzuron was not significantly different from the controls. None of the larvae that were fed with IGR-treated *A. aurantii* pupated. By contrast, larvae that had only been sprayed with IGRs pupated, but no adults emerged. Adult fecundity was not affected by exposure to IGRs, either in the laboratory or in the field, but all eggs exposed to IGRs failed to hatch. Although larvae developed to the adult stage in the field experiments, the IGRs' ovicidal activity and effects on immature stages still had a detrimental effect on *C. nigritus* population levels. As a result, spraying of IGRs is likely to impede *C. nigritus* population increases in citrus orchards. This emphasizes the need to avoid spraying during *C. nigritus* population increases should the use of IGRs be unavoidable. Insect growth regulator's impact on non-target species still requires further consideration, especially with the incorporation of these chemicals into integrated pest management programmes.

Key words: *Chilocorus nigritus*, insect growth regulators, buprofezin, pyriproxyfen, teflubenzuron, citrus, pest control, integrated pest management.

INTRODUCTION

Many species of Coccinellidae are beneficial predators of homopteran pests. In the citrus industry, coccinellids are of economic importance owing to their activity against these pests. Red scale, *Aonidiella aurantii* (Maskell) (Homoptera: Diaspididae) is a serious homopteran pest in citrus, especially in southern Africa (Samways 1988). Chemical control of red scale became ineffective during the 1970s when the it developed resistance to organophosphate compounds. A predator, *Chilocorus nigritus* (Fabricius) (Coleoptera: Coccinellidae), was observed to have a major impact on red scale populations, especially in the southern African lowveld where the levels of red scale infestation were often severe (Samways & Mapp 1983; Samways & Tate 1986).

Chilocorus nigritus is indigenous to India and the Far East (Ahmad 1970). In Pakistan, this beetle is effective against *Aspidiotus destructor* Signoret (Homoptera: Diaspididae), *A. aurantii* and *Quadraspiodiotus perniciosus* (Comstock) (Homoptera: Diaspididae). The beetle later became established in the Seychelles, Mauritius and East Africa, where it preyed on scale insects such as *Ischnaspis longirostis* (Signoret), *Chrysomphalus ficus* (Ashmead) and *Pinnaspis buxi* (Bouche) (Vezey-Fitzgerald 1941; Vezey-Fitzgerald 1953; Greathead & Pope 1977). This predator first appeared in the South African lowveld and later became

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established in the lowveld of Swaziland and Mozambique (Samways 1984).

Chilocorus nigritus appears to have been remarkably effective as a natural enemy of red scale populations, even on trees with twig die-back caused by high levels of red scale infestation. Under such circumstances, the predator is capable of clearing red scale and reducing it to economically acceptable thresholds. (Samways & Mapp 1983; Samways 1988). Control of *C. nigritus* is enhanced by its ability to feed on all sessile stages of the scale, including the gravid female, (Samways & Tate 1986; Samways & Wilson 1988).

Pesticides are known to have deleterious effects on non-target organisms including beneficial insect predators. These effects have resulted in unexpected repercussions associated with pest control in the agricultural industry, which is dependent on agrochemicals to maximize profits. Despite being promoted as innocuous to non-target organisms, various studies have indicated that insect growth regulators (IGRs) also have a negative impact on non-target, beneficial species (Mansour *et al.* 1993; Ragusa Di Chiara *et al.* 1993; Sauphanor *et al.* 1993; Gerling & Sinai 1994; Butaye & Degheele 1995). In particular, IGRs appeared to have a detrimental effect on beneficial Coccinellidae as indicated by studies involving *Cryptolaemus montrouzieri* Mulsant (Smith & Papacek 1990), *Rodolia cardinalis* (Mulsant) (Loia & Viggiani 1992), *Chilocorus bipustulatus* Linnaeus (Peleg 1983) and *Stethorus punctum* (Biddinger & Hull 1995). Furthermore, the use of pyriproxyfen on southern African citrus is suspected to have led to the decline of *C. nigritus* populations on the giant bamboo, *Dendrocalamus giganteus* Munro, used by the beetles as alternative habitats near citrus orchards (Hattingh & Tate 1995). This has serious implications for biological control of pests in citrus orchards.

The activity of predators in agroecosystems is far more effective in the control of economically damaging species than chemical control (Pyle *et al.* 1981). Maintenance of coccinellid populations in citrus orchards would thus contribute to reducing pesticide use, with significant economic implications (Bedford 1968; Hattingh 1995). *Chilocorus nigritus* is one of the most economically important Coccinellidae in the Swaziland lowveld, where IGRs are used. This study was carried out to identify which growth stages of *C. nigritus* were susceptible to IGRs used on citrus in Swaziland. Both laboratory and field studies were conducted to determine the effects of IGRs on adult fecundity, larval development and egg viability of eggs.

MATERIAL AND METHODS

Commercial formulations of three insect growth regulators were tested. These were buprofezin (Applaud, Nihon Nohyaku Co., Japan) at 30 g/100 l 25 % wettable powder, pyriproxyfen (Nemesis, Agrihold) at 30ml/100 l plus 300 ml mineral oil emulsifiable concentrate and teflubenzuron (Nomolt, Celamerck GmbH, Germany) at 20 ml/100 l SC. All three chemicals are registered for use against various citrus pests in Swaziland's citrus orchards. Distilled water was used as a control.

Rearing of host scale (Aspidiotus nerii) *and* Chilocorus nigritus

The Oleander scale, *Aspidiotus nerii* Bouche, was used as substitute prey for *C. nigritus*, collected from bamboo at the Inyoni Yami Swaziland Irrigation Scheme (Tunzini), The polyphagous Oleander scale was reared on potatoes (*Solanum tuberosum* L.) and butternuts (*Curcubita moschcata* Turnhalle).

The host vegetables were prepared by washing in a 2.5 % sodium hypochlorite solution followed by dipping in a 0.2 % solution of benomyl fungicide (Erichsen *et al.* 1991). After drying at room temperature for at least 24 hours, the vegetables were infested with the scale. Potatoes were used for the stock culture from which the butternuts were infested. Butternuts infested with mature scale were then placed in wooden boxes used for rearing *C. nigritus*. The boxes (500 × 500 × 800 mm) had netted sides, hard masonite bases and glass tops. Polyester fibre pads were provided as oviposition sites for the beetles. Water was provided daily by spraying the interior of the boxes with a micro-syringe. The oviposition pads of polyester fibre were removed three times a week and placed in hatcheries comprising cylindrical insect cages with clear sides and a netted top (380 × 200 mm).

Newly-emerged adults were transferred from the hatcheries into the rearing boxes containing healthy butternuts. Before each experiment, *C. nigritus* adults were removed from the rearing boxes and sexed according to the guidelines of Samways & Tate (1984). Immature stages were also selected according to the life stage required for

each experiment. Both adults and immature stages were used for the laboratory and field experiments.

Laboratory experiments

Laboratory experiments were carried out to identify the IGR-susceptible stages and to investigate the effects of the IGRs on the development of *C. nigritus*. Larvae and adults of *C. nigritus* were exposed to IGRs in two ways. First, *C. nigritus* were provided with prey that was treated with IGRs. Butternuts covered with various stages of *A. nerii* were dipped into either buprofezin, pyriproxyfen, teflubenzuron or distilled water for 10 seconds, then air-dried for at least three hours. The life stage to be tested was then placed on the butternuts which, in turn, were placed in netted cages (200 × 200 × 150 mm). Larvae were fed on the treated prey for three days or until they moulted, whichever occurred first, and then transferred to untreated prey for further feeding. Observations on mortality and development were made daily. Fecundity of adults over a 24-hour period was assessed at two and seven days after treatment. Second, *C. nigritus* were sprayed with IGRs. Individuals were placed in empty netted cages and sprayed with one of the IGRs and then air-dried for at least three hours. The beetles were then transferred onto untreated butternuts infested with scale. Observations on mortality, development and fecundity were made as described above.

Each treatment was repeated three times, with each replicate having at least nine larvae, depending on the availability of the life stage. Five pairs (male and female) of adults per replicate were used in the experiments that required adults.

Field experiments

Experiments were carried at three commercial estates in Swaziland, Tambuti Estate, Tambankulu Estate and Inyoni Yami Swaziland Irrigation Scheme (Tunzini), whenever the appropriate IGR was being sprayed. Five pairs of adults were placed in a 200 × 200 × 150 mm wooden-framed, nylon-mesh-covered cage together with a butternut. Before each orchard was sprayed, four transect lines were measured from the edges of the orchard along the four cardinal points. Along each transect line, five intervals based on the log base two scale were measured, *i.e.* 2, 4, 8, 16m and 32 m.

Two cages were then placed on citrus trees at each interval, although those at 2 m and 4 m were usually placed on tall weedy plants since they were usually within the space that separated orchards. In addition, four cages with adults, three cages with egg pads and two cages with various stages of *C. nigritus* on butternuts were randomly placed within the experimental orchard. Cages with larvae were also placed at the 2 m and 32 m intervals.

A full cover spray of buprofezin (0.4 kg/2000 l plus 3.5 l/2000 l Sunspray) was carried out at Inyoni Yami Swaziland Irrigation Schemes, using J8 spray guns. Pyriproxyfen (600 ml/2000 l plus oil 6 l/2000 l) was sprayed at Tambuti Estate, using an Eagle sprayer also at full cover. Spraying was carried out in the morning when there was no wind, to minimize drift. Wind conditions were carefully monitored and spraying was postponed if conditions were unsuitable. *Chilocorus nigritus* was initially collected from the unsprayed, control orchard at Tambankulu Estate for rearing.

The cages were left in the field for 5–6 hours after spraying and then taken to the laboratory where they were separated according to their respective orchard placements. The IGRs were sprayed at different times so that insects exposed to different treatments were never simultaneously housed in the same room. Controls from an unsprayed orchard were taken to a separate laboratory.

Observations on adult and larval mortality, oviposition and larval development were made daily, and the results were recorded at 24 hours, 48 hours and one week after spraying.

Statistical analyses

The proportions of larvae that moulted, died or pupated were arcsin-transformed, while adult oviposition was $\log(x + 1)$ transformed before the data were analysed using the SPSS statistical package.

RESULTS

Laboratory experiments

Chilocorus nigritus *larvae fed prey treated with IGRs*

After 24 hours, there were no significant differences ($P > 0.05$) in larval mortality, either where the IGR was ingested ($P = 0.101$) or life-stage treated ($P = 0.194$). After 48 hours, however, larval

mortality was significantly higher ($P < 0.05$) in those that had ingested buprofezin-treated prey ($P = 0.005$) and these differences were more pronounced after a week (Table 1). All the first-instar larvae that had ingested buprofezin-treated scale died during their first moult. Although the first moult commenced, with either the head or body cuticle splitting, the next-instar larva was unable to emerge from the old cuticle and eventually died. Mortality of first instar larvae that had ingested the IGRs was significantly higher than that of fourth instar larvae, one week after treatment (Tukey HSD, $P = 0.05$). Pyriproxyfen had the least effect on larvae after one week (Table 1), and only first instar larvae suffered mortality at this time. Although first, second and third instar larvae that had ingested teflubenzuron-treated scale suffered some mortality, none of the fourth instar larvae had died one week after treatment.

Despite the survival and successful moults of many of the larvae, all larvae that had ingested IGR-treated scale failed to pupate. This is in contrast to the control ($P = 0.000$), where 94.44 ± 7.86 % of the larvae pupated. In the controls, fourth instar larvae had a higher rate of successful pupation than the younger life stages, although the differences were not significant (Tukey HSD, $P = 0.05$) (Table 1).

A two-way ANOVA indicated a significant interaction between the IGR used for treatment and the life stage exposed ($P = 0.000$). This is reflected in Table 1 which reveals that first instar larvae suffered a higher mortality than fourth instar larvae seven days after treatment (Tukey HSD, $P = 0.05$).

Chilocorus nigritus *larvae sprayed with IGRs*

When larvae were sprayed with IGRs and then transferred to uncontaminated scale insects, there were significant differences in mortality after 48 hours depending upon the IGR sprayed ($P = 0.004$) (Table 2). In particular, buprofezin-sprayed larvae suffered significantly higher mortality than the teflubenzuron-sprayed larvae and the controls, and these differences were more apparent after seven days.

Larval development up to pupation was not affected by the life stages treated ($P = 0.960$), but all IGR treatments caused significantly lower pupation relative to the controls ($P = 0.000$) (Table 2). The buprofezin-treated group had the lowest proportion of pupation (7.76 ± 13.93 %) compared to the pyriproxyfen (44.08 ± 17.81 %), teflubenzuron (65.77 ± 14.99 %) and control (94.44 ± 7.86 %) groups (Tukey HSD, $P = 0.05$).

Pupae originating from larvae treated with buprofezin and teflubenzuron appeared normal but no adults emerged during the period of observation. By contrast, pupae developing from larvae treated with pyriproxyfen usually showed some deformities, where the wings spread out from the pupal case, and none of these deformed pupae produced adults. With regard to larval mortality,

Table 1. Mean (±1 S.D.) percentage mortality and pupation of *Chilocorus nigritus* larvae that were fed with scale insects treated with the three insect growth regulators in the laboratory.

IGR	Larval stage treated	% Mortality at indicated periods after treatment		% Pupation
		2 days	7 days	
Buprofezin	1st	5.14 ± 4.81^{a}	$100.00 \pm 0.00^{a,c}$	0.00 ± 0.00^{b}
	2nd/3rd	17.36 ± 23.23^{a}	60.56 ± 34.17^{a}	0.00 ± 0.00^{b}
	4th	10.56 ± 12.95^{a}	$2.22 \pm 3.85^{a,d}$	0.00 ± 0.00^{b}
Pyriproxyfen	1st	0.00 ± 0.00^{b}	$6.667 \pm 11.55^{b,c}$	0.00 ± 0.00^{b}
	2nd/3rd	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}
	4th	0.00 ± 0.00^{b}	$0.00 \pm 0.00^{b,d}$	0.00 ± 0.00^{b}
Teflubenzuron	1st	$18.52 \pm 6.42^{a,b}$	$18.52 \pm 6.42^{b,c}$	0.00 ± 0.00^{b}
	2nd/3rd	$0.00 \pm 0.00^{a,b}$	13.67 ± 7.56^{b}	0.00 ± 0.00^{b}
	4th	$0.00 \pm 0.00^{a,b}$	$0.00 \pm 0.00^{b,d}$	0.00 ± 0.00^{b}
Control	1st	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}	88.89 ± 9.62^{a}
	2nd/3rd	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}	97.22 ± 3.93^{a}
	4th	0.00 ± 0.00^{b}	0.00 ± 0.00^{b}	100.0 ± 0.00^{a}

Means compared by one-way ANOVA; those followed by the same letter (IGR, instar) are not significantly different ($P > 0.05$; Tukey HSD multiple range test).

Table 2. Mean (±1 S.D.) percentage mortality and pupation of *Chilocorus nigritus* larvae that were sprayed with the three insect growth regulators in the laboratory.

IGR used	Larval stage treated	% Mortality at indicated periods after treatment		% Pupation
		2 days	7 days	
Buprofezin	1st	28.57 ± 14.29^{a}	61.92 ± 35.95^{a}	14.43 ± 24.74^{c}
	2nd/3rd	$25.18 \pm 7.04^{a,c}$	$82.04 \pm 5.36^{a,d}$	8.98 ± 2.68^{c}
	4th	$2.38 \pm 4.12^{a,d}$	$7.94 \pm 8.36^{a,e}$	0.00 ± 0.00^{c}
Pyriproxyfen	1st	0.00 ± 0.00^{a}	5.56 ± 9.62^{b}	38.89 ± 25.46^{b}
	2nd/3rd	$29.17 \pm 29.46^{a,c}$	$50.00 \pm 58.93^{b,d}$	41.67 ± 0.00^{b}
	4th	$5.16 \pm 4.51^{a,d}$	$5.16 \pm 4.51^{b,e}$	50.39 ± 14.10^{b}
Teflubenzuron	1st	5.56 ± 9.62^{b}	11.11 ± 9.62^{c}	67.41 ± 17.78^{b}
	2nd/3rd	$7.78 \pm 8.39^{b,c}$	$18.33 \pm 21.28^{c,d}$	63.52 ± 23.81^{b}
	4th	$0.00 \pm 0.00^{b,d}$	$0.00 \pm 0.00^{c,e}$	62.69 ± 11.25^{b}
Control	1st	0.00 ± 0.00^{b}	0.00 ± 0.00^{c}	88.89 ± 9.62^{a}
	2nd/3rd	0.00 ± 0.00^{b}	0.00 ± 0.00^{c}	97.22 ± 3.91^{a}
	4th	0.00 ± 0.00^{b}	0.00 ± 0.00^{c}	100.00 ± 0.00^{a}

Means compared by one-way ANOVA; those followed by the same letter (IGR, instar) are not significantly different ($P > 0.05$; Tukey HSD multiple range test).

there was no significant interaction between the life stage treated and IGR used ($P = 0.051$), seven days after treatment.

IGR effects on C. nigritus *adults*

Adult mortality was not observed in either experiment, indicating that adult survival is not adversely affected by any of the IGRs. Furthermore, there were no significant differences in oviposition success (over a 24-hour period) between the IGR treatments, seven days after treatment, regardless of whether the adults were exposed through ingestion of treated scale ($P = 0.134$) or sprayed ($P = 0.084$), and the controls (Table 3). Oviposition was higher seven days after treatment in those exposed through ingestion of treated scale ($P = 0.027$). When the adults were sprayed, there was no significant difference in oviposition 48 hours and one week after treatment ($P = 0.188$) (Table 3).

Field experiments

None of the IGRs was directly toxic to adult *C. nigritus*. Adult mortality was usually due to drowning through inundation by the high volumes of liquid during spraying. This was observed in individuals placed at the centre of the experimental orchard and those placed in close proximity to the orchard edges, *e.g.* at 2 m. The viability of eggs produced by the experimental female *C. nigritus* was not investigated owing to the lack of equipment.

There were no significant differences in oviposition relative to the direction of the transects, or whether the cages at the centre of the orchard were included ($P = 0.888$) or excluded ($P = 0.821$). There were, however, significant differences in oviposition relative to the distance from the orchard edge ($P = 0.002$) (Table 4). Adults exposed at 2 m displayed a significantly lower oviposition rate than those exposed at 16 m and 32 m from the orchard edge (Tukey HSD, $P = 0.05$). When the cages at the centre of the orchard were excluded, oviposition at 2 m was still significantly lower than that at 8, 16 and 32 m (Tukey HSD, $P = 0.05$). There were significant differences in the oviposition rate when comparing the IGRs. At two days after treatment, oviposition in the control groups was significantly higher than that in the buprofezin and pyriproxyfen-treated groups ($P = 0.000$).

There was no interaction between the IGR used and the distance from the orchard sprayed, whether the cages at the orchard centre were included ($P = 0.608$) or excluded ($P = 0.740$). There was also no significant difference in oviposition between the two intervals ($P = 0.163$). None of the eggs exposed to buprofezin and pyriproxyfen hatched, while 80.09 ± 3.20 % of those in the control group hatched. Eggs exposed to IGRs changed from their normal golden-brown colour to black.

Owing to limitations in availability, larvae were

Table 3. Mean (±1 S.D.) number of eggs deposited per female, over a 24-hour period, after the respective treatment in the laboratory.

IGR used	Adults fed treated scale		Adults only sprayed	
	2 days	7 days	2 days	7 days
Buprofezin	0.56 ± 0.34[a]	2.64 ± 1.23[a]	1.61 ± 1.43[a]	2.69 ± 0.38[a]
Pyriproxyfen	2.83 ± 0.58[a]	3.31 ± 0.92[a]	2.42 ± 2.32[a]	4.63 ± 1.46[a]
Teflubenzuron	0.56 ± 0.57[a]	3.61 ± 0.65[a]	1.45 ± 1.93[a]	3.22 ± 0.49[a]
Control	0.92 ± 0.30[a]	1.11 ± 0.34[a]	0.92 ± 0.30[a]	1.11 ± 0.34[a]

Means compared by one-way ANOVA; those followed by the same letter are not significantly different ($P > 0.05$; Tukey HSD multiple range test).

Table 4. Mean (±1 S.D.) number of eggs laid per female, over a 24-hour period, after exposure to insect growth regulators during field sprays.

IGR used	Distance from orchard edge (m)	Eggs per female at times indicated	
		2 days	7 days
Buprofezin	2	0.42 ± 0.08[b,c]	0.00 ± 0.00[a,c]
	4	0.67 ± 0.61[b]	1.13 ± 1.24[a]
	8	0.88 ± 1.10[b]	1.21 ± 1.46[a,d]
	16	2.33 ± 1.18[b,d]	1.96 ± 1.57[a,d]
	32	2.17 ± 1.06[b,d]	2.58 ± 1.85[a,d]
Pyriproxyfen	2	0.00 ± 0.00[b,c]	0.50 ± 1.00[a,c]
	4	0.88 ± 1.12[b]	3.21 ± 2.18[a]
	8	1.71 ± 1.25[b]	3.13 ± 2.30[a,d]
	16	1.67 ± 1.42[b,d]	4.21 ± 3.57[a,d]
	32	1.13 ± 0.37[b,d]	2.88 ± 2.29[a,d]
Control	2	2.13 ± 1.51[a]	1.79 ± 0.92[a]
	4	2.08 ± 1.35[a]	2.17 ± 0.49[a]
	8	3.83 ± 1.58[a]	3.83 ± 0.69[a]
	16	4.04 ± 1.47[a]	3.04 ± 0.71[a]
	32	2.83 ± 0.86[a]	2.50 ± 0.94[a]

Means compared by one-way ANOVA; those followed by the same letter (IGR, distance) are not significantly different ($P > 0.05$; Tukey HSD multiple range test).

placed only at the centre of the orchard and at 2 m and 32 m from the orchard edge. The percentage of larvae that pupated was higher in the controls than in the treated groups ($P = 0.003$) (Table 5). In addition, where both IGR treatments were considered, higher proportions of larvae exposed at 32 m pupated, compared to those at 2 m and the orchard centre ($P = 0.036$). Approximately 57.89 % of larvae exposed to buprofezin developed into apparently normal pupae, but all failed to produce adults, while 15.79 % formed deformed pupae and only 26.32 % emerged as adults. In larvae treated with pyriproxyfen, the pupae were also deformed and did not produce adults during the period of observation, while other fourth instar larvae never pupated. Pupal deformities were determined by the emergence of wings through the pupal case, in which the wings were smaller than normal.

DISCUSSION

Chitin synthesis occurs actively during larval development (Boness 1983; Ishaaya 1990) and IGRs could consequently have an impact on larval stages, with adults seldom being adversely affected. No adult mortality due to IGR activity was observed in this study. Effects on adults were thus investigated in terms of their fecundity after exposure to IGRs.

Despite the initial significant differences in

Table 5. Mean (±1 S.D.) percentage pupation of larvae exposed to insect growth regulators during field sprays.

Distance from orchard edge (m)	% Larval pupation after exposure to IGRs		
	Buprofezin	Pyriproxyfen	Control
Centre	7.41 ± 12.83[a]	6.11 ± 6.73[a]	81.81 ± 12.85[a]
2	0.00 ± 0.00[a]	12.50 ± 12.50[a]	90.00 ± 0.00[a]
32	53.13 ± 27.72[b]	46.86 ± 15.73[b]	90.00 ± 7.35[b]

Means compared by one-way ANOVA; those followed by the same letter are not significantly different ($P > 0.05$; Tukey HSD multiple range test).

fecundity between exposed females and the control after two days, there were no significant differences after seven days. This suggested that the IGRs did not have a significant impact on the overall fecundity of the exposed females, confirming observations by Hattingh & Tate (1995) for the same species, Peleg (1983) for *C. bipustulatus* and Ascher *et al.* (1986) for *Carpophilus hemipterus* (Linnaeus).

Although egg viability was not determined owing to various constraints, other studies have indicated that egg viability is severely affected by exposure to IGRs. Hatching of eggs laid by *C. bipustulatus* females exposed to methoprene, diflubenzuron and fenoxycarb was totally inhibited, with similar results being obtained when *C. hemipterus* was exposed to diflubenzuron, chlorfluazuron, XRD-473 and teflubenzuron (Peleg 1983; Ascher *et al.* 1986). Although egg viability was restored when the females were transferred to a clean environment, continual exposure to IGRs during field applications would still have a detrimental effect on the predator populations. The disruption of normal population increases would render the predators incapable of controlling pest populations.

Although there were significant differences in fecundity relative to distance from the orchard edges, this was unlikely to have been due to IGR exposure. The females at the centre of the orchard and at 2 m from the edge were drenched during spraying. The slight decline in egg production may have been caused by the disruption of suitable environmental conditions experienced during spraying. Mating and oviposition would thus be delayed until the individuals recovered.

The three IGRs tested had varying detrimental effects on *C. nigritus* larvae. Since larval development involves moulting and associated chitin synthesis, the effects of these chemicals would probably be most pronounced during these developmental stages. Buprofezin activity was apparent at the end of the life stage treated, with the younger stages (*i.e.* first-instar larvae) being the most susceptible. In another study, fenoxycarb was observed to have a greater detrimental effect on early-instar larvae than late instar larvae of *Stethorus punctum* (LeConte) (Biddinger & Hull 1995).

Buprofezin acts through the inhibition of chitin synthesis as well as disturbance of hormone balance (Nagata 1986; Kobayashi *et al.* 1989; Konno 1990). This results in the inhibition of ecdysis, and mortality results from the inability of larvae to emerge from the old cuticle, as observed in this study. Development of larvae in the pyriproxyfen and teflubenzuron treatments was not significantly different from the controls both in laboratory and field experiments. Teflubenzuron, methoprene and fenoxycarb also did not affect larval development in *R. cardinalis* and *C. bipustulatus*, even when the larvae preyed on treated scale (Peleg 1983; Loia & Viggiani 1992).

When larvae were fed IGR-treated scale, none pupated despite having moulted successfully, particularly as in the pyriproxyfen and teflubenzuron treatments. In addition to the high retention of IGRs in larval tissue and lower detoxification, late-instar larvae and pupae were extremely sensitive to IGR activity. The presence of IGRs in the insect's body, even in minute quantities, would consequently inhibit further development during the late developmental stages (Staal 1975; Ishaaya 1990). Peleg (1983) also observed that, despite *C. bipustulatus* larvae having moulted normally until the fourth instar, the duration of the fourth instar was extended by 2–3 weeks, after which all larvae died before pupation. *Coccinella septempuncata* larvae also failed to complete their development after exposure to diofenolan. The inability of coccinellid larvae to complete their development after exposure to IGRs has been widely reported (Sechser 1994).

Insect growth regulators function as stomach poisons and would thus have maximum effect on predators when ingested from contaminated prey, rather than by direct contact. This was confirmed by the higher mortality and failure to pupate in individuals that had preyed on IGR-treated scale compared to those that were sprayed with the IGRs. In orchard situations, the less-mobile larvae are more likely to feed on contaminated prey. The predator populations would thus be affected by IGRs during the immature stages through the disruption of larval moulting and inhibition of pupation. Predator populations would consequently decline owing to the lack of progeny (Kramer *et al.* 1981).

Insect growth regulators have the greatest detrimental effect during the initial spray periods when predators are in direct contact with the chemicals. Predators are, however, capable of resuming their development and their egg viability if they can move to an uncontaminated environment (Hattingh & Tate 1995; Peleg 1983). This was manifest in *C. nigritus* larvae that developed to pupation after temporary exposure in the field and laboratory. Movement of adults to uncontaminated habitats occurs naturally in the field and *C. nigritus* colonizes bamboo stands where other scale insects are available as alternative prey (Samways 1984). Movement between sprayed orchards and uncontaminated environments would be particularly beneficial if *C. nigritus* could feed on either red scale or alternative scale species and was able to adapt to different prey species. If, however, *C. nigritus* preferred scale species in orchards it would be continually exposed to contaminated prey and populations would decline.

All three IGRs had strong ovicidal effects and all exposed eggs failed to hatch. According to Masner *et al.* (1987), the ovicidal activity of IGRs functions through the inhibition of embryonic development in young eggs. Ovicidal effects may also occur through trans-ovarial transmission. Consequently, eggs laid by treated females also mostly fail to hatch and, in the few that do hatch, the larvae do not survive beyond the early instars (Staal 1975; Ascher *et al.* 1986; Yasui *et al.* 1987; Ishaaya & Horowitz 1992).

The IGR effects in the field were still apparent at 32 m, which was the furthest distance assessed in this study. Larvae exposed at this distance had significantly lower pupation rates (53 % for buprofezin and 47 % for pyriproxyfen), than in the controls (90 %). The effect of these chemicals was further compounded by the formation of abnormal pupae and the lower percentage of adult emergence.

Although adults were not affected by the IGRs, their reproductive success was adversely influenced as a result of non-viable eggs (Ascher *et al.* 1986; Peleg 1983; Hattingh & Tate 1995). The resulting decline in the population's reproductive potential, together with the negative effects of IGRs on larval development and egg viability, would ultimately have a negative impact on *C. nigritus* populations. Lower predator populations would be unable to effectively control pest populations, which would tend to recover much more quickly from adverse conditions. Even when scale populations are at low levels, predator presence remains necessary to ensure a regulatory effect on the pest populations (Samways 1984).

The persistence of these chemicals in the field also requires consideration. Hattingh & Tate (1995) observed that pyriproxyfen residues had adverse effects on female *C. nigritus*, even 131 days after spraying. Despite having a shorter residual activity (Hattingh & Tate 1995), the initial devastating effects of buprofezin observed in this study would still have a negative impact on predator population levels and contribute to the predator's inefficiency in controlling scale pests. Insect growth regulator persistence requires consideration since these chemicals are sprayed in spring and early summer, when predator populations increase. Such population increases are likely to be hampered by the presence of IGRs, even in their residual form. In the field environment, buprofezin was the least detrimental since exposed larvae were able to develop to the reproductive adult stage, while those exposed to pyriproxyfen failed to do so. Although teflubenzuron was not tested in the field, the laboratory experiments suggested that it was the least detrimental of the three compounds.

This study indicated that larval and egg stages are the worst affected by IGRs, especially the early instars. Consequently, coccinellid population increases in the orchards are likely to be impeded, causing reduced rates of predator activity. These impacts are aggravated by the residual activity of IGRs. Should IGR spraying be unavoidable in orchards with high coccinellid activity, it would be advisable to prevent the spray coinciding with

periods of maximum larval and egg development.

Chilocorus nigritus is a valuable natural enemy of red scale in southern Africa and sustainable population levels within citrus agroecosystems are of considerable importance. Careful monitoring of IGR activity and effects on natural enemies is an essential prerequisite in the agrochemical and agricultural industries.

ACKNOWLEDGEMENTS

We thank the technical managers and senior pest control scouts at all three estates.

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Accepted 26 October 1999