

**EFFECTS OF THE INSECT GROWTH REGULATOR
PYRIPROXYFEN ON IMMATURE STAGES OF SUNN
PEST, *EURYGASTER INTEGRICEPS* PUTON
(HETEROPTERA: SCUTELLERIDAE)**

M. Mojaver* and A. R. Bandani*

* Plant Protection Department, Faculty of Agriculture, University of Tehran, Karaj, IRAN.
E-mail: abandani@ut.ac.ir

[Mojaver, M. & Bandani, A. R. 2010. Effects of the insect growth regulator pyriproxyfen on immature stages of Sunn Pest, *Eurygaster integriceps* Puton (Heteroptera: Scutelleridae). Munis Entomology & Zoology, 5 (1): 187-197]

ABSTRACT: The effect of insect growth regulator (pyriproxyfen) on immature stages (eggs and nymphs) of Sunn pest, *Eurygaster integriceps* Puton (Heteroptera: Scutelleridae), which is of great importance in wheat and barley fields as nymphal and adult stages, was determined. When one day old eggs were treated with concentrations of 0.0, 0.1, 10, 100 and 1000 ppm, the egg hatchability were 96.90, 92.38, 98.4, 93.21 and 55.23 %, respectively. Similar results obtained when 3 day-old eggs were treated. However, when 5 day-old eggs were treated, no significant differences in egg hatchability between treatments observed ($F = 1.51$, $P > 0.01$). Adult weight was significantly influenced by treatment of nymphal instars. Treatments of first nymphal instars caused significant differences in adult weight between control and treatments ($F = 25.63$, $P < 0.01$). Adult weights were 102.5, 86.14, 69.47, 56.84, 62.11, and 64.56 mg when first instar nymphs were treated with concentrations of 0.0, 0.1, 10, 100 and 1000 ppm pyriproxyfen, respectively. Reduction of adult weight was significantly more when the nymphs were treated with high concentrations of pyriproxyfen. Nymphal development time was not significantly affected by pyriproxyfen treatment ($P > 0.1$). Adult emergence, sex ratio, nymphal survival rate and adults abnormalities were also affected by pyriproxyfen treatments of nymphs.

KEY WORDS: *Eurygaster integriceps*, immature stages, pyriproxyfen

Modern insecticide research started almost 65 years ago with the chlorinated hydrocarbons, followed shortly by the organophosphates, methylcarbamates and botanicals e.g. pyrethroids and their analogs. The use of these conventional organic insecticides to control insect pests has given rise to problems of the proliferation of resistance and accumulation of residue in the environment with adverse ecological effects (Hoffmann and Lorenz, 1997). In the search for safer insecticide technologies, *i.e.*, more selective modes of action and reduced risks for non-target organisms and the environment, progress has been made in the last two decades with the development of natural and synthetic compounds capable of interfering with the processes of growth, development and metamorphosis of the target insects (Smet et al., 1990; Oberlander et al.; 1978, 1997).

Since the early 1970s, numerous analogs of JH (juvenoids) have been tested for insecticidal activity (Sehna, 1976). Most of the early analogs resemble JH in their basic terpenoid structure. The first juvenoids were farnesol and farnesal isolated from insects themselves (Dhadialla et al. 1998). The 'paper factor' (Slama and Williams, 1965), now called juvabione, represents a group of hormone mimics present in a variety of plants where they may function as defensive mechanisms against herbivorous insects (Dhadialla et al. 2005). Two very active JH analogs, methoprene and hydroxy-methoprene lack the epoxide function present in JH. More recently, several highly active compounds with less apparent similarity to JH (aromatic non-terpenoidal JH analogs) like fenoxycarb, pyriproxyfen and

diofenolan have been synthesized (Dhadialla et al. 1998; Dhadialla et al. 2005). These chemicals have been called insect growth regulators (IGR) or third-generation insecticides (Williams, 1967). IGRs differ widely from the commonly used insecticides, as they exert their insecticidal effects through their influence on development, metamorphosis and reproduction of the target insects by disrupting the normal activity of the endocrine system (Smet et al., 1990; Oberlander et al., 1997).

In the European corn borer, *Ostrinia nubilalis*, application of fenoxycarb during the second to fourth larval instars had no effect on the duration of these instars. However, the duration of the resulting fifth instars increased significantly (Gadenne et al., 1990). Applications of fenoxycarb in the fifth instar produced different effects, depending upon the dose and the timing of application, which resulted in production of supernumerary or permanent larvae, or of larval-pupal intermediates (Gadenne et al., 1990). Li and Chen (2001) observed significant effects of fenoxycarb on all immature stages (eggs, three larval instars and pupae) of *Chrysoperla rufilabris* when tested at three concentrations of (0.1, 1.0 and 10.0 mg [AI]/l). Fenoxycarb had ovicidal effect, lethal effect on larvae and pupae. Also, fenoxycarb significantly delayed the developmental times from the stage treated to adult emergence for all immatures of *C. rufilabris* that successfully developed to adults by 3.2-4.6, 2.3-3.0, 2.1-2.8, and 4.6-7.5 days when egg, first, second and third instars were treated, respectively (Li and Chen, 2001).

Hatakoshi et al. (1991) found that pyriproxyfen was much more potent in inducing supernumerary larvae than methoprene and JH I when injected into last larval-stage of *Spodoptera litura*. Treatment of *C. fumiferana* larvae with fenoxycarb resulted in larval-pupal intermediates and production of deformed pupae (Mulye and Gordon, 1989; Hicks and Gordon, 1992). Related morphogenetic effects have been observed with fenoxycarb for *Heliothis virescens* (Mauchamp et al. 1989). Application of fenoxycarb to the fifth instar of the German cockroach, *Blattella germanica*, not only induced morphological deformities but also induced sterility in adults. The sterility seems to have been transferred from treated males mated to untreated females, which suggests effects on sperm (King and Bennett, 1990). Similar sterility effects were also obtained for *C. fumiferana* (Hicks and Gordon, 1992). Topical application of fenoxycarb suppressed egg production by queens of the red imported fire ant, *Solenopsis invicta* (Banks et al. 1988), and reduced both egg production and hatching in the California fivespined ips, *Ips paraconfusus* (Chen & Borden, 1989). Pyriproxyfen decreased ecdysteroid titer in the hemolymph of the mealworm (*Tenebrio molitor*) when applied to the notum of the newly ecdysed pupae.

In addition, the treatment induced an increase of the protein in the hemolymph of the treated species (Aribi et al., 2005). Supernumerary-molt nymphs with 1-3 extra molts were found when the first three nymphal instars of turnip aphid, *Lipaphis erysimi* were exposed to juvenile hormone analog, pyriproxyfen, and all these nymphs died prematurely. The longevity of all pyriproxyfen-treated *L. erysimi* nymphs and adults and the fecundity of treated adults were reduced by 50% (Liu & Chen, 2001).

Since, there are no published reports on the effects of pyriproxyfen on the Sunn pest, the aim of the current study was to determine the effect of pyriproxyfen on immature stages of Sunn pest, *Eurygaster integriceps* Puton (Heteroptera: Scutelleridae) that is of great importance in wheat and barley fields as nymphal and adult stages in the wide area of the Near and Middle East, West and Central Asia. There is hope that the insect control strategies based on the insect endocrine system will help to overcome residual toxicity and development

of resistant strains for conventional insecticides especially of organophosphorus insecticide (e.g. fenitrothion) which are used extensively in order to control this insect.

Studying the effects of pyriproxyfen on the critical life stages of Sunn pest will help in their proper management. The critical life stages of Sunn pest that are at risk due to spraying of pyriproxyfen are the eggs (embryonic development), nymphal instars and the adults. Therefore, in this study the effects of pyriproxyfen on the hatchability of the insect's eggs, insects' developmental time, adults' weight, adults' emergence, adults' sex ratio, and survival rate of nymphal stages were assessed.

MATERIALS AND METHODS

Insects

The insects were collected before oviposition from Mash-had wheat farms in Mash-had province, Iran. The colony (Adult insects) was maintained and reared on wheat plants or wheat kernels (nymphs) in the laboratory at 25 ± 2 °C, 65 % relative humidity and a photoperiod of 14 h light: 10 h dark (LD 14:10). All tests were run at the mentioned condition.

Insecticide

Pyriproxyfen (registered as ADMIRAL® 10 % EC, Sumitomo Chemical Co.) were used in bioassays.

Egg bioassays

The insect rearing boxes were investigated daily and newly laid eggs (0-24 h) were chosen and left in plastic Petri dishes (9.0 cm diam.) until they developed to the desired age. Three different batches of 1.0 (0-24h), 3.0, and 5.0 day-old eggs were selected and used for egg bioassays. In this assay, five concentrations of 0.0 (Control), 0.1, 10, 100, and 1000 ppm of pyriproxyfen were prepared in distilled water. For each egg batch one assay was made. Each assay consisted of five treatments (five different concentrations of pyriproxyfen) and each treatment (insecticide concentration) had five replications. In each replication 50 eggs were used. The whole experiment was repeated twice.

The selected eggs were dipped in the pyriproxyfen solutions for 5 s. The treated eggs were placed in a Petri dishes lined with a Whatman No. 1 filter paper for 2 h to air dry. Then, the eggs were transferred to a rearing box and their hatching was recorded daily.

Residual bioassay

Pyriproxyfen was dissolved in acetone to give six concentrations of 0.01, 0.1, 10, 100, 1000 ppm.

Petri dishes (9 cm diam.) lined with Whatman No. 1 filter paper and 1000 μ l of each concentration were applied to the filter paper followed by air drying for 2 hour at room temperature. Controls were treated with acetone alone.

For this assay, for each nymphal stadium one assay was made. Since Sunn pest have five nymphal stages, five different assays were run. To obtain desired stages of nymphs, eggs with the same age were separated and allowed to hatch in the rearing box. After the eggs hatched, the nymphs were reared and fed with 12 h-soaked wheat kernels until they developed to the desired instars.

Nymphs of 0-12 h after hatching were considered first instar and nymphs of 0-12 h after first, second, third, and fourth ecdysis were considered second, third, fourth, and fifth instars, respectively. As mentioned before, for each nymphal instar one assay was made. Each assay consisted of six treatments (six different concentrations of pyriproxyfen) and each treatment was replicated six times using

15 nymphs per replicate. The whole experiment was repeated twice. Mortality (survival rate) was recorded daily until nymphs moult to the adults.

Effect of pyriproxyfen on insect development

One assay as described in the previous section (residual assay) was set up to test the effect of pyriproxyfen on insect growth and development. The parameters recorded were developmental time (the period between eclosion from egg to adult emergence for the first instar nymphs and the period between nymphal stages and adult emergence for the other nymphal stages), adults' weight (the weights of individual emerging adult), adult sex ratio (the numbers of females emerged relative to the numbers of adults emerged), percentage of adult emergence (the numbers of emerging adults relative to the initial cohort size) and percentage of adult abnormalities (the numbers of abnormal adults emerged relative to the numbers of emerged adults). Abnormal adults were defined as adults with some degree of deformation in antennae, legs, wings, mouthparts (beak).

Data analysis

Data was submitted to analysis of variance (ANOVA) and mean comparison was made using Duncan test in a completely randomized design. ANOVA was used to evaluate treatment effects on nymphal developmental time, survival rate, adults' weight, adults' emergence, sex ratio (percent female emergence), and adults' abnormalities.

RESULTS

Effects of pyriproxyfen on eggs hatchability (embryonic development)

The effect of pyriproxyfen on different stages of embryonic development of Sunn pest is shown in table 1. There were significant differences in 1 day-old eggs treatments ($F = 14.75$, $P < 0.01$) (Table 1). In one day-old egg treatment, hatchability in concentrations of 0.0, 0.1, 10, and 100 ppm were 96.90, 92.38, 98.4 and 93.21%, respectively. There were no differences between egg hatchability in concentrations of 0.1, 10, and 100 ppm and that in control (Table 1). Survival rate was significantly lower when the nymphs were treated with the highest concentration. When one day-old egg was treated with concentration of 1000 ppm, only 55.23 % of eggs were survived. Similar results obtained when 3 day-old eggs were treated with different concentrations of pyriproxyfen. Similar to treated 1 day-old eggs, survival rates of 3 day-old eggs were significantly different among concentrations ($F = 16.1$, $P < 0.01$). Survival rate of 3 day-old eggs when treated with concentrations of 1000 ppm was 71.42 % which was significantly different from the other concentrations used. However, when 5 day-old eggs were treated, no significant differences between treatments were observed ($F = 1.51$, $P > 0.01$). When 5 day-old eggs treated with concentrations of 0.0, 0.1, 10, 100, and 1000 ppm, percentage of egg hatching were 100, 97.14, 95.37, 100, and 92.91, respectively. Even at a high dose (1000 ppm), the percentage of egg hatching (92.91%) was not significantly different from the other concentrations.

Effect of pyriproxyfen on insect developmental time

As can be seen from table 2, there were no significant differences in the developmental duration between treatments ($P > 0.1$). The developmental duration to adults in treated first instar nymphs with concentrations of 0.0, 0.01, 0.1, 10, 100, and 1000 ppm of pyriproxyfen were 26.5, 28, 29, 27.5, 30, and 28.5 days, respectively (Table 2). Thus, when first instar nymphs were treated, there were no significant effect on the subsequent development of the first instar to adults ($F = 2.36$, $P > 0.1$). However, developmental duration was longest at high concentrations. Similar results were obtained when second, third, fourth and fifth

instars were treated. Generally, the developmental times of treated nymphs in high doses (100 and 1000 ppm) were delayed slightly. The duration of nymphal instars when first instar nymphs were treated with concentrations of 100 and 1000 ppm were 30 and 28.5 days, while that in control was 26.5 days (table 2).

Effect of pyriproxyfen on survival rate

When first instar nymphs were treated, percentage of mortality was significantly different among the five concentrations used ($P < 0.01$). Similar results were obtained when nymphs of the other instars were treated.

Survival rate was significantly lower when the nymphs were treated with the highest dose (1000 ppm) (Table 3). Mortality was no more than 12% in controls, while mortality at highest dose against first, second, third, fourth and fifth instar nymphs were 52.03, 51.92, 66.06, 38.85, and 59.21%, respectively (Table 3).

Effect of pyriproxyfen on adults' weight

When all five nymphal instars were treated, there were significant differences in adult weight among treatments (Table 4). Generally, adults in the controls weighed consistently more than those in pyriproxyfen treatments. Treatments of first nymphal instars caused significant differences between control and treatments ($F = 25.63$, $P < 0.01$). Reduction of adult weight was significantly more when the nymphs were treated with high concentrations of pyriproxyfen (Table 4). When first instar nymphs were treated with concentrations of 0.0 (control) and 1000 ppm, adults weight were 102.5 and 64.56 mg, respectively. Adult weight reductions were 16 and 38% when first instar nymphs were treated with concentrations of 0.01 and 1000 ppm, respectively. Younger nymphs (first, second and third instar nymphs) were more affected than older nymphs. For example treatments of younger nymphs with different concentrations of pyriproxyfen caused significant differences in adult weight between control and treatments ($P < 0.01$), whereas treatment of older nymphs (fourth and fifth instar nymphs) did not produce significant differences between control and treatments ($P > 0.01$).

First, second, third, and fourth instar nymphs treated with concentration of 1000 ppm adults' weight were 64.56, 60.17, 63.83, 72.34, and 80 mg, respectively (Table 4). As a result, higher nymphal stages less reduction of adult weight observed. For example, when first instar nymphs were treated with high concentration of pyriproxyfen (1000 ppm) adult weight was 64.56 mg while adult weight which obtained from treatment of fifth instar with the same concentration of pyriproxyfen was 80.32 mg.

Effect of pyriproxyfen on adults' emergence and their sex ratios

Adult emergence was affected by pyriproxyfen treatments and as pyriproxyfen concentrations increased the percentage of adult emergence decreased (Table 5). Significant differences were found between treatments and control when first, second, third, fourth and fifth instar nymphs were treated ($P < 0.01$). In comparison with the control the lowest adult emergence was observed when fifth instar nymphs were treated with a concentration of 1000 ppm that the percentage of adult emergence was about 46%. Whereas, nymphs treated with a concentration of 1000 ppm in the first instar, percentage of adult emergence was about 53%.

Adult sex ratios were affected by nymphal treatments (Table 6). Treatment of nymphal stages with pyriproxyfen caused a significant difference from 1♂:1♀ in each case.

When first instar nymphs were treated with concentrations of 0.01 and 1000 ppm, percentage of female individuals in the populations were 52.78 and 18.06 %, respectively ($F = 10.46$, $P < 0.01$) (Table 6). The effect of pyriproxyfen on adult

sex ratio was more on younger instars than older instars. For example, when first and fifth instars were treated with concentrations of 1000 ppm, percentages of adult females were 18.06 and 43.08, respectively.

Effect of pyriproxyfen on the emergence of deformed adults

Pyriproxyfen had strong effect on production of abnormal adults (Table 7). Treatment of fifth instar nymphs with different concentration of pyriproxyfen caused significant differences in production of deformed adults between treatments and control ($F = 43.41$, $P < 0.01$). For example when first and fifth instar nymphs were treated with 0.01 and 1000 ppm, percentage of deformed adults were 46.78 and 83.97, respectively.

DISCUSSION

The presented data showed that application of pyriproxyfen to eggs and nymphs of Sunn pest (*Eurygaster integriceps*) resulted in increased mortality and slight prolonged development of nymphs. Pyriproxyfen showed significant ovicidal effect when younger eggs were treated depending on concentration used. For example, when first, second and third instar nymphs were treated at 1000 ppm survival rates were 55.23, 71.42 and 92.91%, respectively. Similar results have been reported when fenoxycarb tested on eggs of *Chrysoperla rufilabris* (Neuroptera: Chrysopidae) (Liu & Chen, 2001) or topical application of fenoxycarb to adult insects of California fivespined ips, *Ips paraconfusus*, reduced both egg production by females and egg hatching (Chen & Borden, 1989).

Liu & Chen (2001) reported egg hatching rate of 66.7% when they applied a concentration of 10 mg (AI L⁻¹) of fenoxycarb. Bhargava and Urs (1993) reported mortality effects on the eggs of rice moth (*Corcyra cephalonica*) exposed to various doses of hydroprene. Among the three age groups of eggs that they exposed to hydroprene, the hatching percentage was highly reduced in the freshly laid eggs (0-12 h old) compared to older eggs. Also, embryonic effects of fenoxycarb have been observed in the eggs of the eastern spruce budworm (*Choristoneura fumiferana*) (Hicks & Gordon, 1992), and the cat flea (*Ctenocephalides felis*) (Marchiondo et al., 1990). It has been reported that juvenile hormone analogs (JHAs) are more effective at the beginning stage of metamorphosis and embryogenesis in insects, such as freshly ecdysed last larval instars, freshly ecdysed pupal instars, and freshly deposited eggs (Dhadialla et al., 1998; Tunaz & Uygun, 2004). Thus embryogenesis is disrupted when young eggs are treated with JHAs. Eggs exposed to fenoxycarb and other juvenile hormone analogs show disruption of the blastoderm with associated cellular and organelle disruption (Dhadialla et al., 1998).

Application of pyriproxyfen to different stages (early and late nymphal instars) of Sunn pest nymphs did not significantly affect nymphal developmental time although slight increase in developmental time was observed when younger instars were treated with high concentrations of pyriproxyfen. The number of days taken for first nymphal instar to emerge as adults in control was 26.5 days, which was the shortest time, while days taken for first instar treatments with 100 and 1000 ppm were 30 and 28.5 days, respectively. So, the longest development time was observed when first nymphal instar treated with the highest concentrations of pyriproxyfen.

In the European corn borer, *O. nubilalis*, application of fenoxycarb during the second to fourth larval instars had no effect on the duration of these instars, while the duration of the resulting fifth instars increased significantly (Gadenne et al., 1990).

Apart from developmental time, which was not strongly affected by the IGR, the other parameters including percentage of adult emergence, adult weight, adult sex ratio, and deformed adults were strongly affected by pyriproxyfen. As reported in the literature (Koehler & Patterson, 1991; Dhadialla et al., 1998; Kostyukovsky et al., 2000).

Adult weights in Sunn pest were strongly affected by pyriproxyfen. For example in control the adult insect weigh more than 100 mg while first nymphal instars, which treatment with highest dose, weigh about 64.56 mg. Weight is the main feature of the adult insects that influence its reproductive competitiveness (Slansky & Scriber, 1985). Weight is an indicator of the amount of energy and nutrients stored in the body which can influence mate-seeking, dispersal flights and fecundity. Presumably, as a consequence of weight importance, insect larvae or nymphs have evolved a means to evaluate their body weight prior to making the neurohormonal decision to go to the next stage (Nijhout, 1994; Nation, 2002).

Pyriproxyfen affects the hormonal balance in insects and results in a strong suppression of embryogenesis, metamorphosis, and adult formation. Accumulated nymphal mortality was as high as 66% in the treated nymphs. Highest mortality (66 and 59 %) occurred in the fourth and fifth nymphal instars and lowest mortality occurred in the third nymphal instars (36.85%) followed by second (51.92%) and first nymphal instars (52.03%). Results of the current study showed that pyriproxyfen can cause direct mortality as well as strong sublethal effects on treated nymphs. These effects are in accordance with the response of *Aphis gossypii* to pyriproxyfen (Wood & Godrey, 1998), *Myzus persicae* to pyriproxyfen (Hatakoshi et al., 1991), *Lipaphis erysimi* to pyriproxyfen (Liu & Chen, 2001), hydroprene and methoprene (Sidhu & Arora, 1990), stored product insects such as *Tribolium castaneum* and *Sitophilus oryzae* to pyriproxyfen (Kostyukovsky et al., 2000), and *Hyposoter didymator* to pyriproxyfen (Schneider et al., 2004).

Treatment of fifth instar nymphs with different concentrations of pyriproxyfen caused significant differences in production of deformed adults e.g. highest dose produced 83.97 % of deformed adults. Abnormalities effect was more than any other effects observed. These results showed that pyriproxyfen has a more potent juvenilizing effect than the other effects on Sunn pest. Similar results were reported by Singh (1992) and Liu & Chen (2001) who found that pyriproxyfen causes some degree of abnormalities in treated *Lipaphis erysimi*. In insects the principal hormones involved in the life processes of insects include neurohormones (neuropeptides), ecdysteroids (molting hormones) and the sesquiterpenoid juvenile hormones (JHs). In general ecdysone (hydroxylated steroid) is involved in molting and juvenile hormone (sesquiterpene) is involved in maintaining the insects in current form (Status quo) (Hoffmann & Lorenz, 1997; Gade et al., 1997; Goodman & Granger, 2005). In insects with incomplete metamorphosis such as Sunn pest at the time of the final molt, JH is absent and the adult emerges. Therefore, persistence of JH or juvenile hormone analogs (JHA) during that time, depending upon the dose and time of application, give rise to abnormal adults.

In conclusion it should be mentioned that pyriproxyfen could cause direct mortality (lethal effects) on eggs and nymphs and sub-lethal effects such as reduction of the insect weight, disruption of sex ratio, production of abnormal adults and to some extent interference in the nymphal longevity. All these effects were observed in laboratory conditions. Further investigations of the effect of pyriproxyfen on Sunn pest population should be conducted on a larger scale under field conditions.

ACKNOWLEDGEMENT

This work was funded by University of Tehran grant number 31303.

LITERATURE CITED

- Banks, W. A., Williams, D. F. & Lofgren, C. S.** 1988. Effectiveness of fenoxycarb for control of the red imported fire ants (Hymenoptera: Formicidae). *Journal of Economic Entomology*, 81: 83–87.
- Bhargava, M. C. & Urs, K. C. D.** 1993. Ovicidal effect of three insect growth regulators on *Corcyra cephalonica*. *Indian Journal of Plant Protection*, 21: 195–197.
- Chen, N. M. & Borden, J. H.** 1989. Adverse effects of fenoxycarb on reproduction by the California five-spined ips, *Ips paraconfusus* Lanier (Coleoptera: Scolytidae). *Canadian Entomologist*, 121: 1059–68.
- Dhadialla, T. S., Carlson, G. R. & Le, D. P.** 1998. New insecticides with ecdysteroidal and juvenile hormone activity. *Annual Review of Entomology*, 43: 545–69.
- Gade, G., Hoffmann, K. H. & Spring, J. H.** 1997. Hormonal regulation in insects: facts, gaps, and future directions. *Physiological Review*, 77: 963–1032.
- Gadenne, C., Grenier, S., Mauchamp, B. & Plantevin, G.** 1990. Effects of a juvenile hormone mimetic, fenoxycarb, on postembryonic development of the European corn borer, *Ostrinia nubilalis* Hbn. *Experientia*, 46: 744–47.
- Granett, J. & Wesoloh, R. M.** 1975. Dimilin toxicity to the gypsy moth larval parasitoid, *Apanteles melanoscelus*. *Journal of Economic Entomology*, 68: 577–580.
- Hatakoshi, M., Shono, Y., Yamamoto, H. & Hirano, M.** 1991. Effects of juvenile hormone analog pyriproxyfen on *Myzus persicae* and *Unaspis yanonensis*. *Applied Entomology and Zoology*, 26: 412–414.
- Hicks, B. J. & Gordon, R.** 1992. Effects of the juvenile hormone analog fenoxycarb on various developmental stages of the eastern spruce budworm, *Choristoneura fumiferana* (Clemens) (Lepidoptera: Tortricidae). *Canadian Entomologist*, 124: 117–23.
- Hoffmann, K. H. & Lorenz, M. W.** 1997. The role of ecdysteroids and juvenile hormones in insect reproduction. *Trends in Comparative Biochemistry and Physiology*, 3: 1–8.
- King, J. E. & Bennett, G. W.** 1990. Comparative sterilizing and ovicidal activity of fenoxycarb and hydroprene in adults and oothecae of the German cockroach (Dictyoptera: Blattellidae). *Journal of Medical Entomology*, 27: 642–45.
- Kostyukovsky, M., Chen, B., Atsmi, S. & Shaaya, E.** 2000. Biological activity of two juvenoids and two ecdysteroids against three stored product insects. *Insect Biochemistry and Molecular Biology*, 30: 891–897.
- Marchiondo, A. A., Riner, J. L., Sonenshine, D. E., Rowe, K. F. & Slusser, J. H.** 1990. Ovicidal and larvicidal modes of action of fenoxycarb against the cat flea (Siphonaptera: Pulicidae). *Journal of Medical Entomology*, 27: 913–21.
- Mauchamp, B., Malosse, C. & Saroglia, P.** 1989. Biological effects and metabolism of fenoxycarb after treatment of the fourth and the fifth instars of the tobacco budworm, *Heliothis virescens* F. *Pesticide Science*, 26: 283–301.
- Mulye, H. & Gordon, R.** 1989. Effects of selected juvenile hormone analogs on sixth-instar larvae of the eastern spruce budworm, *Choristoneura fumiferana* Clemens (Lepidoptera: Tortricidae). *Canadian Entomologist*, 121: 1271–72.
- Nation, J. L.** 2002. *Insect Physiology and Biochemistry*. CRC Press, USA.
- Nijhout, H. F.** 1994. *Insect Hormones*. Princeton, NJ: Princeton Univ. Press.

Schneider, M. I., Smagghe, G., Pineda, S. & Viñuela, E. 2004. Action of insect growth regulator insecticides and spinosad on life history parameters and absorption in third-instar larvae of the endoparasitoid *Hyposoter didymator*. *Biological Control*, 31: 189–198.

Sehnal, F. 1976. Action of juvenoids on different groups of insects. pp. 301-321. In: Gilbert LI [ed.] *The Juvenile Hormones*. Plenum Press, New York, NY.

Sidhu, H. S. & Arora, R. 1990. Toxic and demorphogenetic effects of juvenile hormone analogues against alate mustard aphid, *Lipaphis erysimi* (Kalt). *Journal of Research Punjab Agricultural University*, 27: 65–71.

Slama, K. & Williams, C. M. 1965. Juvenile hormone activity for the bug *Pyrrhocoris apterus*. *Proceedings of National Academy of Science of USA*, 54: 411-414.

Williams, C. M. 1967. Third-generation pesticides. *Science of America*, 217: 13-17.

Wood, J. P. & Godfrey, L. D. 1998. Effects of whitefly insect growth regulators Knack and Applaud on cotton aphid reproduction and survival. In: Dugger P, Richter D (eds), *Proceedings of Beltwide Cotton Conference*, San Diego, CA. National Cotton Council, Memphis, TN, pp. 1278–1281.

Table 1. The effect of different concentrations of Pyriproxyfen on hatchability of different ages of egg (1, 3, and 5 day-old eggs) of the Sunn pest.

Concentration (PPM)	Percentage of eggs hatchability (Means \pm se)		
	1 day-old egg treatment	3 day-old egg treatment	5 day-old egg treatment
0.0 (control)	96.90 \pm 1.76 ^a	100 \pm 0.0 ^a	100 \pm 0.0 ^a
0.1	92.38 \pm 4 ^a	100 \pm 5.06 ^a	97.14 \pm 2.86 ^a
10	98.0 \pm 4.1 ^a	97.14 \pm 2.8 ^a	95.37 \pm 4.8 ^a
100	93.21 \pm 4.17 ^a	92.86 \pm 3.3 ^a	100 \pm 0 ^a
1000	55.23 \pm 4.2 ^a	71.42 \pm 1.75 ^b	92.91 \pm 1.2 ^a
F	16.115	14.750	1.516
P	< 0.01	< 0.01	> 0.01

Means in the same column followed by the same letters do not differ significantly at 0.01 (Duncan test).

Table 2. Effects of pyriproxyfen on the duration of nymphal period of Sunn pest when nymphs treated at different developmental stages.

Concentration (PPM)	Stages treated and developmental time to adult (Mean \pm se)				
	1st instar	2nd instar	3rd instar	4th instar	5th instar
0.0 (control)	26.5 \pm 0.5 ^a	26 \pm 1 ^a	20 \pm 1 ^a	15 \pm 1 ^a	10 \pm 0.5 ^a
0.01	28 \pm 1 ^{ab}	26.5 \pm 1.5 ^a	20.5 \pm 1.5 ^a	14.5 \pm 0.5 ^a	11.5 \pm 0 ^b
0.1	29 \pm 1 ^{ab}	28.5 \pm 0.5 ^a	22 \pm 1 ^a	16 \pm 1 ^a	10.5 \pm 0.5 ^{ab}
10	27.5 \pm 0.5 ^{ab}	26 \pm 0 ^a	20 \pm 1 ^a	16.5 \pm 0.5 ^a	10 \pm 0 ^a
100	30 \pm 1 ^b	26 \pm 1 ^a	21 \pm 1 ^a	15.5 \pm 1.5 ^a	10.5 \pm 0.5 ^{ab}
1000	28.5 \pm 0.5 ^{ab}	26 \pm 0 ^a	21.5 \pm 0.5 ^a	16 \pm 1 ^a	10 \pm 0 ^a
F	2.360	1.333	0.615	0.565	2.733
P	> 0.1	> 0.1	> 0.1	> 0.1	> 0.1

Means in the same column followed by the same letters do not differ significantly at 0.01 (Duncan test).

Table 3. Accumulated mortalities of Sunn pest nymphs when five different nymphal instars treated with five concentrations of pyriproxyfen and water (control).

Concentration (PPM)	Stages treated and percentage mortality (Mean \pm se)				
	1st instar	2nd instar	3rd instar	4th instar	5th instar
0.0 (control)	11.39 \pm 0.6	9.46 \pm 0.11	12.47 \pm 2.03	11.95 \pm 1.51	5.67 \pm 0.91
0.01	38.31 \pm 1.16	55.75 \pm 4.91	17.70 \pm 3.43	54.81 \pm 18.98	16.66 \pm 2.38
0.1	28.94 \pm 1.67	37.42 \pm 1.71	19.29 \pm 0.7	65.90 \pm 2.28	25.82 \pm 2.75
10	51.9 \pm 5.25	47.22 \pm 2.78	25.68 \pm 7.67	64.5 \pm 0.87	33.78 \pm 1.75
100	56.15 \pm 0.98	39.11 \pm 7.56	25.92 \pm 2.43	74.82 \pm 3.43	30.17 \pm 0.59
1000	52.03 \pm 6.6	51.92 \pm 1.92	36.85 \pm 9.60	66.06 \pm 4.53	59.21 \pm 2.69

Table 4. Effects of pyriproxyfen on the adults weight of Sunn pest when nymphs treated at different developmental stages.

Concentration (PPM)	Stages treated and adult weight (mg) (Mean \pm se)				
	1st instar	2nd instar	3rd instar	4th instar	5th instar
0.0 (control)	102.5 \pm 1.8 ^a	97.20 \pm 1.2 ^a	94.75 \pm 2.8 ^a	104.03 \pm 3 ^a	104.1 \pm 1.2 ^a
0.01	86.14 \pm 6.7 ^b	65.85 \pm 2.1 ^b	75.88 \pm 0.3 ^b	89.73 \pm 9.6 ^{ab}	99.90 \pm 0.4 ^{ab}
0.1	69.47 \pm 1 ^c	64.25 \pm 1.7 ^b	71.50 \pm 3.5 ^{bc}	79.89 \pm 4.4 ^b	94.02 \pm 2 ^{abc}
10	56.84 \pm 0.4 ^d	53.65 \pm 2.6 ^c	55.06 \pm 3.3 ^d	76.43 \pm 2.7 ^b	86.30 \pm 1.8 ^{cd}
100	62.11 \pm 4.3 ^{cd}	59.31 \pm 5.2 ^{bc}	65.83 \pm 2.8 ^{bc}	78.24 \pm 3.9 ^b	88.44 \pm 4.8 ^{bcd}
1000	64.56 \pm 10.9 ^{cd}	60.17 \pm 1.1 ^{bc}	63.83 \pm 1.3 ^{cd}	72.34 \pm 5.5 ^b	80.32 \pm 6.5 ^d
F	25.636	26.683	20.410	5.042	5.805
P	0.0006	0.0005	0.0011	0.0369	0.0269

Means in the same column followed by the same letters do not differ significantly at 0.01 (Duncan test).

Table 5. Effects of pyriproxyfen on the percentage of adults emergence of Sunn pest when nymphs treated at different developmental stages.

Concentration (PPM)	Stages treated and percentage of adult emergence (Mean \pm se)				
	1st instar	2nd instar	3rd instar	4th instar	5th instar
0.0 (control)	59.7 \pm 0.9a	58 \pm 1.5a	62.18 \pm 9.2a	70.4 \pm 0.1a	94.32 \pm 0.9a
0.01	26.19 \pm 1.1bc	31.34 \pm 2.7b	35.63 \pm 1.44b	66.03 \pm 4.5ab	83.33 \pm 2.3b
0.1	27.38 \pm 10b	16.19 \pm 3.5c	15.47 \pm 1.1b	66.33 \pm 2.7ab	74.17 \pm 2.7c
10	16.36 \pm 1.2bc	15.97 \pm 0.5c	11.41 \pm 0.83bc	53.28 \pm 6.25bc	67.21 \pm 1.7c
100	12.56 \pm 0.4c	12.01 \pm 2.6c	7.93 \pm 2.4c	48.76 \pm 4.82c	69.12 \pm 0.1c
1000	13.18 \pm 5bc	9.61 \pm 2.4c	8.20 \pm 2.7c	41.45 \pm 6.1c	40.78 \pm 2.69d
F	18.278	56.277	8.268	6.292	80.568
P	0.0014	0.0001	0.0115	0.0223	0.000

Means in the same column followed by the same letters do not differ significantly at 0.01 (Duncan test).

Table 6. Effects of pyriproxyfen on the sex ratio (% female) of Sunn pest when nymphs treated at different developmental stages.

Concentration (PPM)	Stages treated and sex ratio(%female)				
	1st instar	2nd instar	3rd instar	4th instar	5th instar
0.0 (control)	57.17±1.1a	43.74±0.2a	46.83±0.9a	66.38±0.2a	53.40±0.6a
0.01	52.78±2.2a	34.23±4.2ab	29.28±0.7a	52.32±7.7b	47.72±2.2ab
0.1	24.69±3.5b	31.73±6.7ab	28.57±1.1b	45.56±2.7bc	42.72±2.7b
10	27.93±0.8b	24.44±2.2bc	25.00±1b	39.50bc±5.5bc	43.75±2b
100	22.14±6.6b	24.36±2.9bc	22.50±2.5bc	32.92±0.4c	42.22±2.2b
1000	18.06±5b	9.090±3.2c	19.64±5.3bc	34.15±9.1c	43.08±1.9b
F	10.462	5.517	15.232	9.902	3.587
P	0.0063	0.0302	0.0023	0.0073	0.0757

Means in the same column followed by the same letters do not differ significantly at 0.01 (Duncan test).

Table 7. Percentage of deformed adults of the Sunn pest when fifth instar nymphs treated with different concentration of pyriproxyfen.

Concentration (PPM)	Percentage of deformed adults after 5th instar treatment
0.0 (control)	0.0±0a
0.01	46.78±6.804b
0.1	54.76±4.77bc
10	66.74±5.89c
100	58.22±5.17.8bc
1000	83.97±1.94d
F	43.415
P	0.0002

Means in the same column followed by the same letters do not differ significantly at 0.01 (Duncan test).