# Morphogenetic Effects of Precocene II in the Brown Planthopper Nilaparvata lugens Stal. (Homoptera: Delphacidae)

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(Received on 16 October 1989; Accepted on 11 November 1989)

Newly moulted third, fourth and fifth instar nymphs of Nilaparvata lugens treated with a single dose of 1  $\mu$ g, 3  $\mu$ g and 5  $\mu$ g of the anti-allatin, precocene II (PII), result in a high rate of mortality in the treated as well as in the subsequent instars. PII treatments also result in the prolongation of nymphal periods. PII treatments of third and fourth instar nymphs induced precocious metamorphosis. In the fifth instar development stage, the PII treated nymphs, moult into adultoids possessing many morphological features of the adult. The significance of these findings is discussed.

Key Words: Nilaparvata lugens, Precocene II, Precocious metamorphosis

#### Introduction

Precocenes (Precocene I and II) isolated from the plant *Ageratum houstonianum* induce symptoms of juvenile hormone (JH) deficiency in certain insects especially in Hemiptera (Bowers et al. 1976, Masner et al. 1979, Farag & Varjas 1981, Hales & Mittler 1981), Orthoptera (Pener & Orshan 1977, Nemec et al. 1978, Pener et al. 1978, 1981, Chenevert et al. 1978, 1981, Unnithan et al. 1980) and in a few species of Lepidoptera (Kiguchi 1982, Sam Mathai & Nair 1983, 1984a, b, Santha & Nair 1986, Santha et al. 1987). The precocenes act on corpora allata causing atrophy and necrosis of the glands esulting in cessation of JH biosynthesis (Unnithan et al. 1977, 1980, Pener et al. 1978, Schooneveld 1979, Jair et al. 1981). Precocene-induced effects include recocious metamorphosis, sterilization, inhibition of

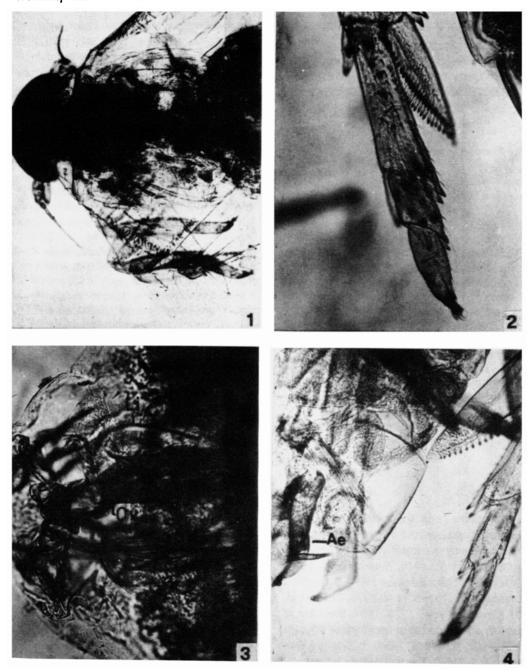
pheromone production, disturbances of embryogenesis, feeding inhibition and toxicity (Bowers 1985). Studies with precocenes have demonstrated their usefulness in basic (to evaluate the role of JH) and in applied (as insect control agents) research.

Nilaparvata lugens (Homoptera: Delphacidae) is a major pest of rice throughout Asia, Pacific regions and Australia. In this article we demonstrate the morphogenetic effects of precocene II, in this insect, after treatments of third, fourth and fifth instar nymphs.

## Materials and Methods

Rearing and staging of nymphs

The third, fourth (penultimate) and fifth (last) instar



Figures 1-4 1 Partially expanded wings with trichoid sensillae of the adultoid; 2, Ill-developed three segmented hind tarsi of the adultoid; 3, Ill-developed female external genitalia of the adultoid (Ov. Ovipositor; Ll, Lateral lobes); 4, Incompletely developed male external genitalia of the adultoid (Ae, Aedeagus)

Table 1	Effects of	PII on the	duration of	the instars	in <b>Nilaparvata</b>	lugens
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Treated	Dose	n ')	No. of Survivors	Duration of the instar (days $\pm$ S.D.)		
instar	(μg/cm²)			Third instar	Fourth instar	Fifth instar
Third	1	34	17	2.8 <u>+</u> 0.9	3.7 <u>+</u> 0.5	3.5 <u>+</u> 0.7*
	3	40	25	2.5 <u>+</u> 0.6	3.4 <u>+</u> 0.1	3.6 <u>+</u> 0.7
	5	25	0	0	0	0
Fourth	1	40	36	_	3.1 <u>+</u> 0.9	4.1 <u>+</u> 1.2
	3	55	19	_	3.7 <u>+</u> 1.1	5.7 <u>+</u> 1.7
	5	25	0	_	0	0
Fifth	1	45	32	_	_	4.1 <u>+</u> 1.3
	3	38	27	_	_	5.3 <u>+</u> 1.4
	5	40	26		_	4.2 <u>+</u> 1.3
Control	_	20	20	2 <u>+</u> 0	2.1 <u>+</u> 0.4	3.0 <u>+</u> 0
		Each				

All the effects in the duration of instars in PII treated insects are significant at the P < 0.001 level or better except one (\*) which is significant at the P < 0.05 level (Student's t-test) when compared to controls.

Table 2 Morphogenetic effects of PII in Nilaparvata lugens

Treated	Dose	n	No. of	% of nymphs showing		
instar	(μg/cm <sup>2</sup> )		survivors	Precocious metamorphosis	Normal metamorphosis	
Third	1	34	17	0	100	
	3	40	25	60	40	
	5	25	0	0	0	
Fourth	1	40	37	15	85	
	3	55	19	84.2	15.8	
	5	25	0	0	0	
Fifth	1	45	32	_	100	
	3	38	27		100	
	5	40	26		100	
Control	_	20	20	0	100	
		Each				

nymphs were isolated from a stock colony of *Nilaparvata lugens* reared and maintained in the laboratory following the methods described by Medrano and Heinrichs (1985). The insects were provided with one-month old rice seedlings of the susceptible variety T(N)1. The insects were reared under 12L:12D photoperiodic regime at  $27 \pm 2^{\circ}C$  and  $57 \pm 3\%$  r.h. Under these laboratory conditions, the total nymphal period was about 12 to 13 days and consisted of five instars. The fifth instar moulted into the adults. The age of the nymph is abbreviated to day n where day 0 indicates the day of ecdysis to this developmental stage.

#### Treatment of animals

Precocene II (PII; 6, 7-dimethoxy-2, 2-dimethyl chromene, Aldrich Chemical Co., West Germany) was dissolved in acetone and diluted to obtain various concentrations. Topical application of nymphs of N. lugens with PII was highly toxic and none of the insects survived. Hence nymphs of N. lugens were treated by the contact method with a residue of PII. In this method. nymphs were confined to 4.6 × 1.6 cm Petri dish coated with PII residue at a concentration 1  $\mu$ g, 3  $\mu$ g or 5  $\mu$ g/cm<sup>2</sup>. The inner surfaces of the glass Petri dish were coated with acetone solution of the compound which provide after evaporation of the solvent a continuous layer of PII residue. At a time 15 Nos. of nymphs were exposed to PII for 1 hr. The nymphs which were kept as controls were released to Petri dishes previously coated with acetone for the same period.

PII treated and control nymphs were released into small test cages. The test cage was made in the following manner. T(N)1 plants were grown singly in earthern pots. Each seedling was drawn through a hole made in the centre of a wooden plank (15 × 15 cm) which rests on the rim of the pot. The tiller of each plant was again drawn through a perforation made in the centre of an inverted transparent plastic cup (7 cm height and 6 cm dia.). The nymphs were transferred to these small cages with the help of an aspirator. Observations were made daily to see whether there was any moulting by the nymphs. On the day of moulting the number of exuviae were counted and removed. The number of dead nymphs as well as those undergoing precocious or normal metamorphosis were recorded.

### Results and Discussion

The results of this study reveal that PII treatments of N. lugens nymphs by contact method result in high rate of mortality (table 1). Mortality is observed not only in the treated stage but also during the subsequent development of these nymphs. Precocenes are toxic substances which may induce a high rate of mortality in treated insects at sufficiently high doses. Similar high rate of mortality after treatment with precocenes have been observed in Locusta migratoria (Pederson 1978, Pener et al. 1978, 1981), Oncopeltus fasciatus (Unnithan & Nair 1979) Acyrthosiphon pisum (Mackauer et al. 1979), Schistocerca gregaria (Chenevert et al. 1980, Unnithan et al. 1980), Dysdercus cingulatus (Farag & Varjas 1981), Diploptera punctata (Feyereisen et al. 1981). Quadraspidiotus perniciosus (Darvas et al. 1985) and Brachycaudus schwartzi (Darwish et al. 1986).

Treatments of nymphs with different doses of Pll result in a significant (P < 0.001, t-test) prolongation of intermoult period (table 1). Delay in moulting has been frequently observed in insects treated with precocenes (Masner et al. 1979, Pener et al. 1981, Sam Mathai and Nair 1983, 1984a, b). These delays are often attributed to direct/indirect effects of precocenes on prothoracic glands which produce the moulting hormone.

In N. lugens, exposure of third and fourth instar nymphs with 3  $\mu$ g PII/cm<sup>2</sup> results in a large proportion of them moulting into non-viable precocious adults (table 2). The prothetelic forms are distinguished by many adult characters. The wings are partially expanded and membraneous. The wings of these adultoids possess distinct venation and the trichoid sensillae characteristic of adults (figure 1). The hind legs of these adultoids have three segmented tarsi (figure 2) whereas in the normal fifth instar the tarsi have only two segments. The external genitalia (lateral lobes and ovipositor in female; aedeagus in male) are also prothetelic (figures 3, 4). Presence of white patches in the anterior segments of the abdomen characteristic of nymphal instars is lacking in these adultoids. Thus these results demonstrate that N. lugens is sensitive to the well known morphogenetic effects of PII. The surprising feature of the study is that precocious metamorphosis could be induced even after treatments of penultimate nymphal instar of N. lugens. Intervention

of an intercalary instar after the treated stage is not essential for the differentiation of adult characters, as has been frequently observed in other hemimetabolous insects sensitive to precocenes.

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# Acknowledgements

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