Proc. Indian natn. Sci. Acad. B48, No. 4, pp. 481–485 (1982)

Effect of Beta-ecdysone Ingestion on 5th and 6th Instar Larvae of the Armyworm, *Spodoptera litura* (Fabr.) (Lepidoptera: Noctuidae)

SHAKIL AHMAD and MUMTAZ A KHAN
Insect Physiology Laboratory, Department of Zoology, Aligarh Muslim University, Aligarh 202001, UP

(Received 26 February 1982; after revision 12 April 1982)

The 5th and 6th instar larvae of the armyworm, *Spodoptera litura* (Lepidoptera: Noctuidae) were separately allowed to ingest 0.5, 1.0, 2.0, 4.0, and 6.0 μg of beta-ecdysone (moulting hormone) to observe its effect on longevity, moulting, metamorphosis, fecundity and fertility. The longevity of the treated larvae of either instar was unaffected except by the ingestion of 6·0 μg and about 5% 6th larvae following the ingestion of this dose moulted to supernumerary instar. Further, ingestion of 6.0 μg dose by both the treated instars produced abnormal pupae but ingestion of 2.0 and 4.0 μg dose by 6th instar larvae also resulted in pupal malformation. The maximum percentage of reduction in fecundity and fertility of the females emerging from the treated larvae of 5th instar was 60.54% and 32.96% respectively whereas the corresponding values were 89.95% and 60.33% in case of the females emerging from the treated larvae of 6th instar.

Key Words: *Spodoptera litura*, **Beta-ecdysone, Longevity, Moulting, Metamorphosis, Fecundity, Fertility**

Introduction

The insect moulting hormone (ecdysone) is known to control moulting during growth. Besides natural ecdysones in the form of alpha- and beta-isomers several ecdysoids including that of plant tissues (phytoecdysone) have been synthesized. The exogenous application of these ecdysones or their analogues results in moulting disorders, suppression of metamorphosis, inhibition in ovarian development, oogenesis etc., in several insects (Engelmann 1959, Bowers 1968, Robbins et al. 1968, 1970, Earle et al. 1970, Wright & Kaplanis 1970, Wright et al. 1971, Kaplanis et al. 1971, Engelmann 1971, Zdarek & Slama 1972, Walker & Thompson 1973 and Garcia et al. 1979).

In the present investigation the effect of different doses of the beta-ecdysone-fed larvae of *Spodoptera litura*, which are polyphagous on several crops, was observed as regards the longevity, moulting, metamorphosis, fecundity and fertility of this species, with a view to controlling the larvae of *S. litura*.

Materials and Methods

From a stock culture of stages of *S. litura* maintained at 30±1°C and 70–80 percent

Table 1 *Showing the disturbances at larval-larval, larval-pupal moulting and absence of emergence following the ingestion of different doses of beta-ecdysone by 5th instar larvae of* Spodoptera litura

Doses (μg)	No. of treated larvae	Mortality at the same stage (No. of larvae)	Mortality at larval-larval moult (No. of larvae)	Mortality at 6th instar stage (No. of larvae)	Mortality at larval-pupal moult (No. of larvae)	Absence of emergence in pupae		Total loss before emergence
						Normal	Abnormal	
Control	100	—	—	—	—	—	—	—
0.5	100	5	—	2	1	—	—	8
1.0	100	5	1	3	2	—	—	11
2.0	100	7	3	4	4	—	—	18
4.0	100	8	2	6	6	—	—	22
6.0	100	10	3	8	7	4	2	34

Table 2 *Showing disturbances at larval-pupal moult, larval mortality during unsuccessful moult to extra instar and number of supernumerary instars formed following the ingestion of different doses of beta-ecdysone by 6th instar larvae of* Spodoptera litura

Doses (μg)	No. of treated larvae	Mortality at the same stage (No. of larvae)	Mortality at larval extra instar moult (No. of larvae)	No. of super-numerary larvae formed	Mortality at larval-pupal moult (No. of larvae)	Absence of emergence in pupae		Total loss before emergence
						Normal	Abnormal	
Control	100	4	—	—	1	—	—	5
0.5	100	4	—	—	5	—	—	9
1.0	100	8	—	—	6	4	—	18
2.0	100	12	4	—	8	7	3	34
4.0	100	15	5	—	14	11	2	47
6.0	100	20	10	5	15	12	3	65

relative humidity newly moulted larvae of 5th and 6th instars were isolated and starved overnight. Doses of beta-ecdysone ranging from 0.5, 1.0, 2.0, 4.0 and 6.0 μg were tested against the individuals of either intsar. Each dose was sprayed on 100 pieces of castor leaves, measuring 5 × 5 cm each and air-dried. Then 100 larvae of either instar were confined with these treated leaf pieces. After complete ingestion of a treated leaf piece by each larva, all larvae fed on one dose were transferred to a separate rearing jar and provided with untreated castor leaves for feeding. The treated larvae by each dose and also the normal larvae (control) were then maintained for observation on their longevity, moulting and metamorphosis. To observe the effect on fecundity and fertility, 6 female moths emerged from the treated larvae were paired with equal number of normal males. Number of eggs laid/female was noted. They were allowed to hatch for the subsequent observation on fertility.

Results and Discussion

Following the ingestion of 6.0 μg beta-ecdysone by each larva of 5th and 6th instar the larval longevity of these instars was reduced by 10–15 hr and 24–36 hr respectively. It was also noted that the longevity of 6th instar larvae, which moulted from 5th instar larvae ingesting 4.0 and 6.0 μg ecdysone was also reduced by 8–10 hr and 15–20 hr respectively. At the lower doses the longevity of either instar generally remained unaffected (table 1 & 2). It thus appears that the steroid beta-ecdysone is neither digested nor hydrolysed in the digestive tract, but is absorbed by the midgut and transported to the haemolymph to increase the concentration of this hormone which is already in circulation. Ingestion of 6.0 μg beta-ecdysone may thus result to enhance the titre of this hormone to such a level which accelerates the moulting process.

Both 5th and 6th instar larvae suffered heavy mortality following the ingestion of either of the selected doses of beta ecdysone (table 1 & 2). Thus by ingesting 6.0 μg ecdysone the percentage of larval mortality in 5th and 6th instars was 28 and 50 respectively. It is well known that the titre of moulting hormone gradually falls as metamorphosis approaches. Thus at 5th and 6th larval stage if the titre of the hormone is enhanced in the haemolymph it will result in abnormal conditions and one of the conditions is toxicity in the body leading to mortality. This is further supported by the fact that the rate of mortality in the larvae treated at 6th instar is higher than those of the 5th instar and also that mortality increases with higher doses of ingested hormone.

Five percent of 6th instar larvae transformed into supernumerary larvae (average length =4.80 cm; width=0.70 cm; weight=0.82 g; as compared to normal 6th instar larva: average length=4.30 cm; width=0.60 cm and weight=0.635 g) following the ingestion of 6.0 μg. But by feeding on lower doses such transformation was less. It is also well known fact that the exogenous increase in juvenile hormone concentration in the fully grown larvae results in formation of a supernumerary larval instar. However, frequent occurrence of an extra larval instar by ingestion of 6.0 μg beta-ecdysone at 6th larval instar is another abnormal feature due to effect of stronger concentration of this hormone either directly or indirectly through further possible secretion of juvenile hormone at this stage. In case of 5th instar larvae, pupal malformation occurred only after the ingestion of 6.0 μg ecdysone (2%) (table 1), whereas with regard to 6th instar larvae, it was prevalent by the ingestion of 2.0, 4.0 and 6.0 μg ecdysone to the level of 3%, 2% and 3% respectively (table 2). The adult emergence was reduced to 66 and 35% in 5th and 6th instar larvae respectively following the ingestion of 6.0 μg. Similar reduction

Table 3 *Showing fecundity and fertility in the female of* Spodoptera litura *emerged form 5th and 6th instar larvae ingesting different doses of beta-ecdysone*

Doses (μg)	5th instar		6th instar	
	Fecundity Mean±SE	Fertility Mean±SE	Fecundity Mean±SE	Fertility Mean±SE
Control	2738.16±39.21	2461.33±45.13	2475.16±13.71	2154.66±25.51
0.5	2260.66±19.63	2061.16±21·63	1811.66±363.77	1370.5±277.72
1.0	2178.00±39.66	1814.33±29.21	1655.83±28.03	1389.83±25.80
2.0	1780.33±19.58	1540.66±37.73	1223.16±26.37	947.66±16.00
4.0	1411.66±21.77	1109. 5±19.50	764. 5±154.80	424. 0±88.23
6·0	1080. 5±11.61	615. 5±20.68	248.83±113.37	66. 5±32.39

in the yield of adults (at 3 ppm conc.) and complete elimination of the formation of adults (at 10 ppm conc.) occurred in case of *Anthonomous grandis* when a trihydroxy 6-keto steriod was added to the larval diet (Earle et al. 1970).

The fecundity and fertility of the females emerging from the treated 5th or 6th instar larvae decreased with the ingestion of increasing doses of beta-ecdysone. Thus, the ingestion of 6.0 μg by the 5th and 6th instar larvae reduced the fecundity in the corresponding females by 60.54% ($t=364.32$, $p<0.05$) and 89.95% ($t=309.2$, $p<0.05$) respectively. Similarly, the reduction in fertility was 32.96% ($t=356.33$, $p<0.05$) and 60.33% ($t=429.66$, $p<0.05$) respectively (table 3). The observations were statistically significant.

Robbins et al. (1968) explained that beta-ecdysone inhibited ovarian maturation and egg production in the housefly, *Musca domestica*, by antagonising the gonadotropic activity. Beta-ecdysone prevented lipid synthesis necessary for vitellogenesis (Wright et al. 1971). Earlier Wright and Kaplanis (1970) observed that in newly-emerged stable flies, *Stomoxyx calcitrans*, ovarian growth was inhibited due to the effect of the same hormone. Thus, the reduction in fecundity and fertility of female *S. litura* is the result of prolonged effect of ingestion of beta-ecdysone at the advanced larval stages. The presence of this hormone in the pupal stage would also inhibit the ovarian growth. Thus, it may be suggested that *S. litura* may be controlled successfully by the application of this hormone at the last larval instar (i.e., 6th instar) which is extremely susceptible to produce the maximum reduction in fecundity and fertility.

Acknowledgements

Authors are grateful to Professor S M Alam, former Head, Department of Zoology, Aligarh Muslim University, who encouraged and provided facilities for this investigation. Thanks are also due to the Indian Council of Agricultural Research, New Delhi, for financial assistance.

References

Bowers W S 1968 Juvenile hormone: Activity of natural and synthetic synergists; *Science* **161** 895–897

Earle N W, Padovani I, Thompson M J and Robbins W E 1970 Inhibition of larval development and egg production in the bollweevil following ingestion of ecdysone analogue; *J. econ. Ent.* **63** 1064–1069

Engelmann F 1959 Uber die Wirkung implantierter Prothoraxdrusen im adulten Weibchen von Leucopheaea maderae; *Z. vergl. Physiol.* **41** 456–470

____ 1971 Endocrine control of insect reproduction, a possible basis for insect control; *Acta Phytopath. Acad. Sci.* Hungary **6** 211–217

Garcia M L M, Mello R P and Garcia E S 1979 Ecdysone, juvenile hormone and oogenesis in *Rhodnius prolixus*; *J. Insect Physiol.* **25** 695–700

Kaplanis J N, Thompson M J and Robbins W E 1971 The effects of ecdysone and analogues on ovarian development and reproduction in the housefly *Musca domestica* (L.); *Proc. 13th Int.Congr. Ent.*, Moscow, 1968 **1** 393

Robbins W E, Kaplanis J N, Thompson M J, Shortino T J, Cohen C F and Joyner S C 1968 Ecdysones and analogues: Effects on development and reproduction in insects; *Science* **161** 1158–1160

____, ____, ____, ____, and ____ 1970 Ecdysones and synthetic analogues: Moulting hormone activity and inhibitive effects on insect growth, metamorphosis and reproduction; *Steroids* **16** 105–125

Wright J E and Kaplanis J N 1970 Ecdysones and ecdysone analogues: Effects on fecundity of the stable fly *Stomoxyx calcitrans*; *Ann. ent. Soc. Am.* **63** 622–623

____ Chamberlain W R and Barret C C 1971 Ovarian maturation in stable flies: Inhibition by 20-hydroxy ecdysone; *Science* **172** 1247–1248

____ and Thompson M J 1973 22, 25-bisdeoxyecdysone: Pathological effects on the Mexican bean beetle and synergism with juvenile hormone activity; *J. econ. Ent.* **66** 64–67

Zdarek J and Slama K 1972 Supernumerary larval instars in Cyclorrhaphcus Diptera; *Biol. Bull.* **142** 350–357