

mined by the method of Lowry et al.<sup>13</sup>. Data were initially evaluated by a 1-way analysis of variance and differences between means were determined using the Student-Newman-Keuls' test<sup>14</sup>.

**Results and discussion.** NE and DA concentrations in various brain regions of rats subjected to SCGX or a sham operation are summarized in the table. Control values for NE and DA contents of the posterior pituitary are in agreement with those reported earlier by Saavedra et al.<sup>8</sup> but are substantially lower than those reported by Holzbauer et al.<sup>9</sup>. SCGX did not alter the dopamine concentration in any of the brain regions analyzed. The NE content of the median eminence was slightly, but not significantly, reduced by SCGX. On the other hand, the NE concentration was significantly reduced in the posterior pituitary and completely depleted from the pineal gland. These results suggest that NE in the pineal gland is contained exclusively in sympathetic nerves which originate in the superior cervical ganglia, while approximately, one-third of the NE in the posterior pituitary is contained in terminals of these peripheral nerves.

Histochemical fluorescent studies have revealed a rich network of fine fluorescent varicose catecholaminergic nerve fibres distributed throughout the neurointermediate lobe of the rat<sup>4</sup>. In addition, fibres of coarser varicosities are located around the larger blood vessels in the neural lobe. Some of these coarser fibres disappear while the network of fine fluorescent fibres is unaltered by bilateral SCGX. The results of the present investigation suggest that the coarser fluorescent fibres contain NE since only this amine declines following sympathectomy. Björklund et al.<sup>4</sup> originally proposed that both NE and DA nerve fibres in the posterior pituitary were of central origin, while only a

few fibres in this tissue were of sympathetic origin. In a subsequent report<sup>5</sup>, however, it was stated that 'noradrenergic innervation of the neurointermediate lobe is probably exclusively of peripheral sympathetic origin'. The results summarized in the table are consistent with the original proposal and suggest that the posterior pituitary contains both NE and DA nerves of central origin, with DA nerves predominating.

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## Precocene-induced metamorphosis in the desert locust *Schistocerca gregaria*<sup>1</sup>

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**Summary.** 50–100% of the 2nd, 3rd and 4th instar nymphs of *Schistocerca gregaria* exposed to precocene 2 by contact method metamorphosed precociously from the instar treated. The corpora allata of the precocious adults were degenerate. Topical application of a juvenile hormone analogue (ZR-512) induced in the precocious adults the colouration characteristic of sexually mature adults.

Precocene 2 extracted from the plant *Ageratum houstonianum* induced precocious metamorphosis in the milkweed bug *Oncopeltus fasciatus*<sup>4,5</sup>. Further studies showed that in *O. fasciatus* precocene inhibited the growth of the corpus allatum (CA)<sup>6</sup> and induced extreme degeneration of the parenchyma cells of the CA<sup>7,8</sup>. Recently Schooneveld<sup>9</sup> observed similar effects in the CA of precocene-treated 4th instar nymphs of *Locusta migratoria migratorioides*. Although topical application of precocene 2 induced precocious metamorphosis in *L. migratoria*<sup>10,11</sup>, this method of precocene application was ineffective in inducing precocious metamorphosis in the desert locust, *Schistocerca gregaria*. On the contrary when we exposed *S. gregaria* nymphs to precocene by contact method<sup>5</sup> inside Petri dishes coated with precocene a high proportion of the treated nymphs metamorphosed precociously. The results are presented in this paper.

**Materials and methods.** Precocene 2(6,7-dimethoxy-2,2-dimethyl chromene) was obtained from Zoecon Corpora-

tion, Palo Alto, Ca. and Aldrich Chemical Co., Milwaukee, Wisc. Various concentrations of it were made in spectrograde acetone. Groups of 6–10 newly-moulted 2nd, 3rd and 4th instar nymphs were exposed to concentrations of precocene residue (10–25 µg/cm<sup>2</sup>) for 24 h in Petri dishes (15×2 cm) at 35 °C (table). The control insects were also kept for 24 h at 35 °C in Petri dishes which were previously treated with acetone only. After treatment the control and precocene-treated insects were transferred to rearing cages and fed regularly on wheat bran and fresh wheat blades and maintained at 35 °C, 16 h photophase and 20–30% relative humidity. For light microscopy CA of 1-day-old normal female adults and of precocious adults from precocene-treated 4th instar nymphs were fixed in paraformaldehyde-glutaraldehyde mixture and embedded in Spurr low viscosity medium<sup>7</sup>. Semithin sections were examined after staining in toluidine blue.

In another series of experiments 1-month-old female and male precocious adults from precocene-treated 3rd instar nymphs were treated topically on the abdomen with 2 doses

Effects of precocene 2 on metamorphosis of 2nd, 3rd and 4th instar nymphs of *S. gregaria*

Nymphal instar and age in h	Number of insects	Dose $\mu\text{g}/\text{cm}^2$	Mortality <sup>a</sup>	Days in treated instar	Number of precocious adults <sup>b</sup>	Number of normal adults <sup>b</sup>
II < 4	11	Control	2	4-5	0	9
II < 4	32	15	12	7-8	20	0
III < 24	20	Control	0	4-6	0	20
III < 24	6	10	0	7-8	4	2
III < 24	63	15	5	7-8	54	4
IV < 18	20	Control	0	5-6	0	20
IV < 18	20	25	0	9-10	10	10

<sup>a</sup> Mortality occurred either during treatment or at subsequent stages of development. <sup>b</sup> The average body lengths of precocious adults from 2nd, 3rd, and 4th instars were 21, 32 and 44 mm respectively, whereas that of normal adults was 62 mm.

of 100  $\mu\text{g}$  each of a juvenile hormone analogue (JHA) (ZR-512) 24 h apart. The control precocious adults received only 5  $\mu\text{l}$  of acetone each time. All insects were returned to cages and provided with food as described earlier. After 3 weeks the ovaries from the control and JHA-treated females were dissected out in 50% ethanol and lengths of the terminal oocytes were recorded.

**Results.** Precocene-induced premature metamorphosis occurred at the 1st moult after precocene treatment (table). The size of the precocious adults varied with the instar treated (table). Precocene-induced premature adult differentiation was accompanied by a delay in moulting in those insects that metamorphosed prematurely (table).

There was no mortality of the precocious adults during the period of this study, which lasted for about 80-90 days. Microscopic examination of the CA revealed that most of its parenchyma cells had degenerated in the precocious adults and that they were replaced by connective tissue from the outer sheath (figures 1 and 2). The normal-looking female adults obtained from precocene treated 4th instar nymphs (table) were neither sterile nor had atrophied CA. These laid eggs, however, the fecundity of these females was not measured.

The precocious adults did not develop the colouration characteristic of sexually mature adults even under crowded conditions. Topical application of JHA induced especially in the males deep yellow colouration characteristic of sexually mature males. Examination of the ovaries revealed that most of the terminal oocytes of JHA treated insects were yellowish in colour and had an average length of  $4.7 \pm 1.1$  mm. In the control insects the terminal oocytes were arrested in the previtellogenic stage and had an average length of  $1.8 \pm 0.2$  mm. Comparison of the means

showed that the differences in the length of the terminal oocytes of the 2 groups were significantly different from each other  $p < 0.001$ .

**Discussion.** The results of this study substantiate the earlier suggestion<sup>7</sup> that premature adult differentiation is apparently due to the degeneration of the CA. That precocene impairs with the functional integrity of the CA is evident also from our observation that precocious adults did not develop the colouration characteristic of sexually mature adults, which is believed to be under the influence of the CA<sup>12,13</sup>. This effect was reversed by treatment with a JHA. Although Nemec et al.<sup>11</sup> observed that precocene treatment of *L. migratoria* nymphs by contact method was less effective in inducing precocious metamorphosis than topical application, it appears that *S. gregaria* is extremely sensitive to contact method and almost insensitive to topical application. It is pertinent to mention that Nemec et al.<sup>11</sup> exposed *L. migratoria* nymphs to precocene 2 residue on filter paper. It is possible that the uptake of precocene by the nymphs from the filter paper may not be as uniform as from glass surface. The reason why *S. gregaria* is insensitive to topical applications of precocene 2 cannot be ascertained from our studies. Other species of *Schistocerca* such as *S. vega* is also insensitive to precocene 2 when applied topically (G.B. Staal, personal communication). Since we have been able to induce precocious metamorphosis in *O. fasciatus* by fumigation with precocene (unpublished) it is possible that the high sensitivity of *S. gregaria* nymphs to contact method inside Petri dishes is due to the rapid uptake of precocene fumigant via the tracheal system to the critical organ(s), whereas topically applied precocene may be rapidly metabolised before it reaches the critical organ(s), probably the CA.

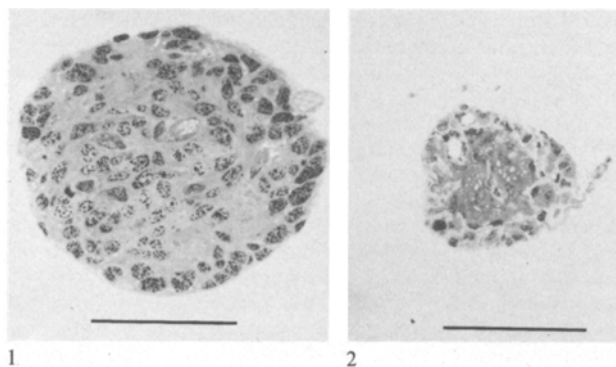


Fig. 1. Semithin section of the corpus allatum of 1-day-old female adult *S. gregaria* stained with toluidine blue. Scale 100  $\mu\text{m}$ .

Fig. 2. Semithin section of the corpus allatum of 1-day-old female precocious adult from 4th instar nymph of *S. gregaria* showing degeneration of the cells. Scale 100  $\mu\text{m}$ .

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