

Effects of Three Juvenile Hormone Analogs on the Female German Cockroach, *Blattella germanica* (L.) (Dictyoptera: Blattellidae)¹

The strategy of insect control by insect hormone analogs lies in the interference with the embryonic and post-embryonic development, and with the reproductive physiology of the adult. Some work on this has been reported for a few important insect pests of field crops, stored products and livestock^{2,3}. So far as is known, no such work has been done on the German cockroach, which is a common household pest in the USA, and whose control continues to be a pressing problem. The present paper describes the effects of 3 juvenile hormone analogs on some aspects of metamorphosis and reproductive physiology of the female German cockroach.

Materials and methods. Three juvenile hormone analogs (JHA), viz., Ro 20-3600⁴ R-20458⁵ and farnesyl methyl ether (FME), were tested on the last instar (6th) female nymphs of German cockroach, *Blattella germanica* (L.). The analogs were used as undiluted technical grades for topical application (2–4 μ l/nymph) and injection (0.33 μ l/nymph), and diluted in acetone for treating topically the food pellets⁶ (8–10 μ l/g). Presence or absence of mating in emerging adults was tested by introducing unmated males that matured in isolation. The internal reproductive structures were studied in freshly killed insects.

Results and discussion. All 3 methods of treatment produced essentially similar effects in various individuals, viz., loss of capacity to molt, death in the process of molting, and various external and internal abnormalities in the emerging adults.

Several treated nymphs showed abnormal prolongation of the 6th stadium up to 2 $\frac{1}{2}$ months, as against the normal period of about 10 days. These affected nymphs

¹ Paper of the Journal Series, New Jersey Agric. Exp. Station, Rutgers–The State University of New Jersey, New Brunswick, New Jersey 08903, USA.

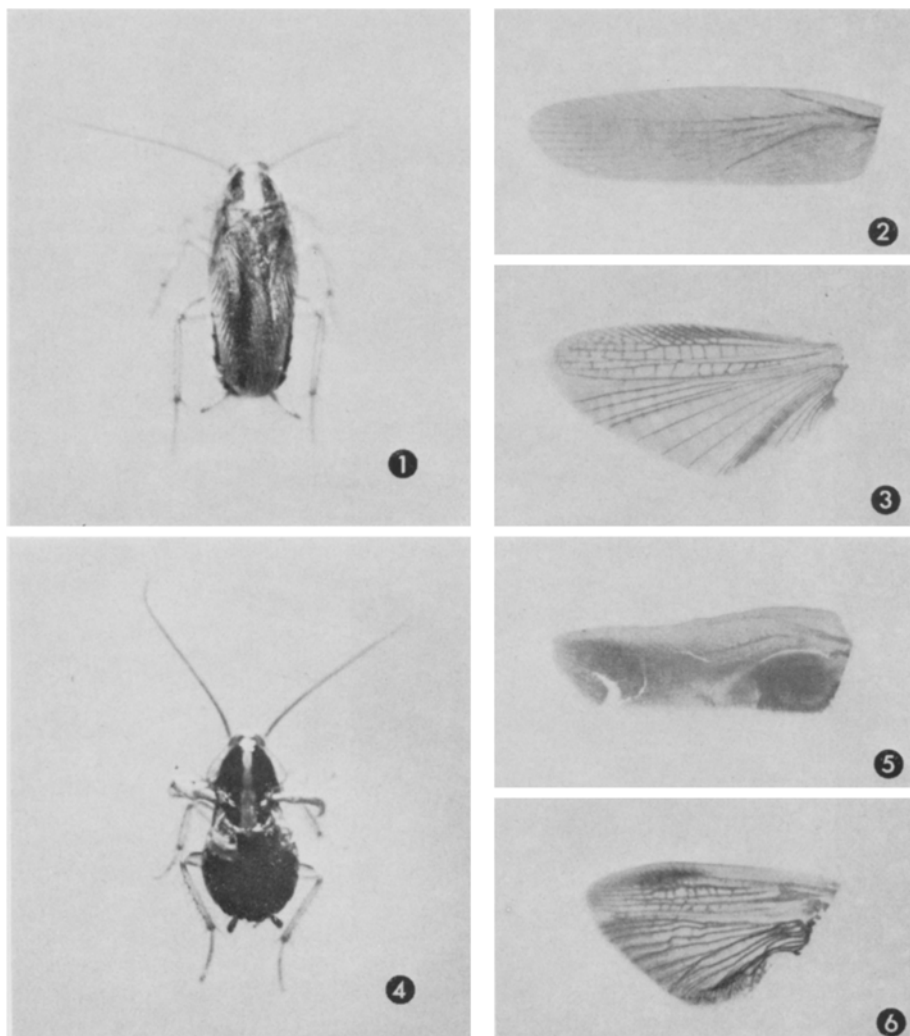
² H. A. SCHNEIDERMAN, A. KRISHNAKUMARAN, P. J. BRYANT and F. SEHNAL, Agric. Sci. Rev. Coop. State Res. Serv. U. S. Dept. Agric. 8, 13 (1970).

³ J. J. MENN and M. BEROZA, *Insect Juvenile Hormones, Chemistry and Action* (Academic Press, New York 1972).

⁴ 6,7-epoxy-3,7-dimethyl-1-(3,4-methylenedioxy)phenoxy-2-nonene. Generous gift from Hoffmann-La Roche, Nutley, New Jersey.

⁵ 1-(4-ethylphenoxy)-6,7-epoxy-3,7-dimethyl-2-octene. Generous gift from Stauffer Chemical Co., Richmond, California.

⁶ Little Friskies cat food, Carnation Co., Los Angeles, California.



Figures 1–6. 1. Normal adult female. 2. Normal front wing. 3. Normal hind wing. 4. Treated adult female (topically with R-20458 2 μ l/nymph). 5. Affected front wing. 6. Affected hind wing.

eventually died without metamorphosis. This failure to molt is generally assumed to be due to the absence of endogenous molting hormone, but it is likely that in the present case the JHA superseded the molting hormone both in quantity and effect. A mutually antagonistic action of these two hormones on nucleic acid synthesis was offered as tentative explanation by SOCHA and SEHNAL⁷ for the induced abnormalities in *Tenebrio molitor*. In our present study we have not seen any extra (supernumerary) molting, as reported by STOCK and O'FARRELL⁸, who observed 'an attempted adult molt' and 'the persistence of the prothoracic gland in the molting adult individuals' when the 6th instar nymphs were treated with FME at a critical period. Dissection of one 56-day-old treated nymph from our study showed evidence of oocyte development, even though metamorphosis did not occur. Metamorphosis and oocyte development are thus independently controlled by the juvenile hormone through some regulatory mechanism.

One of the external abnormalities is the deformity of wings (Figures 2, 3, 5 and 6). The wings of the treated adults were greatly reduced in size, curved, and only slightly larger than the nymphal wing pads. The venation

and the regional wing topography are apparently unaffected. Microscopic examination of the deformed wings revealed that the difficulties in expansion and flattening of the wings are the primary causes for the wing deformity. WHITE and GREGORY⁹ attributed such a situation in the cabbage aphid, *Brevicoryne brassicae*, to the abnormal quantities of microtubules in the wing cells.

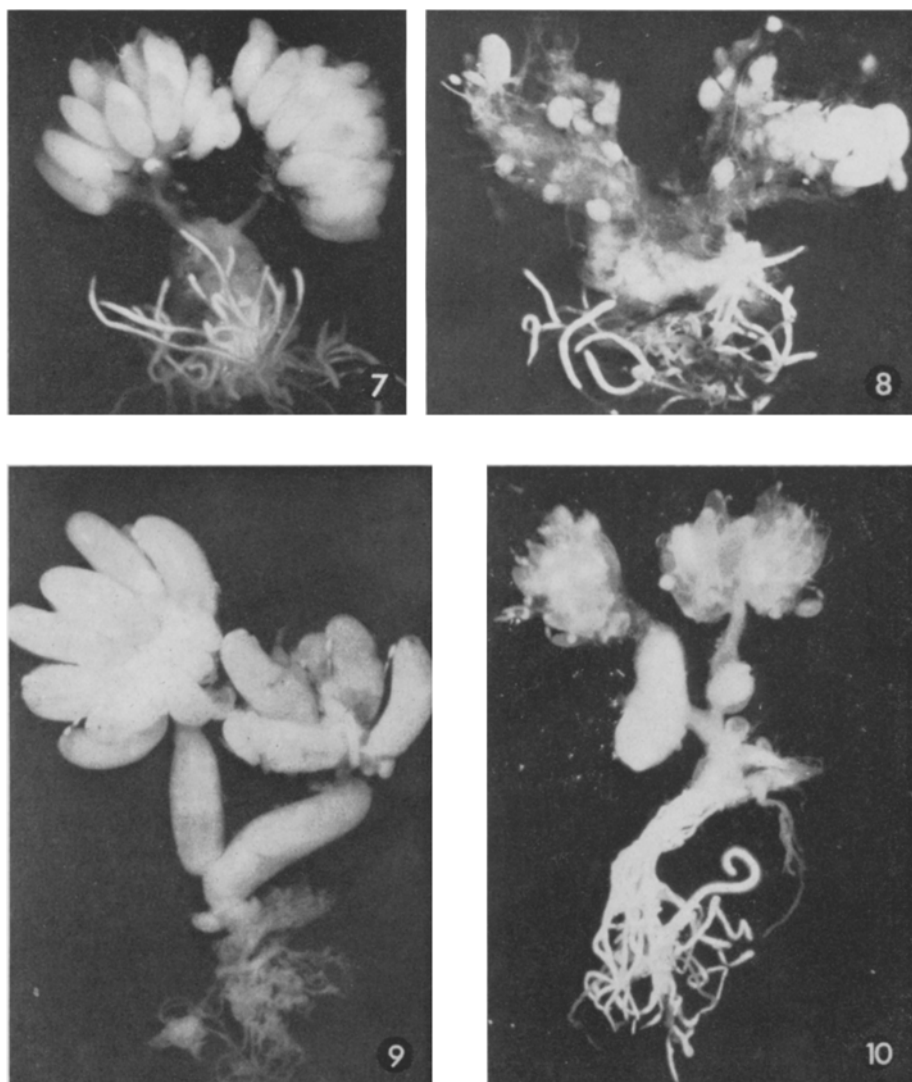
The most striking external feature of the treated females is the dark body colour. The normal cuticular colour pattern is hardly traceable in the general darkening of the body, including the head and appendages (Figures 1 and 4). STOCK and O'FARRELL⁸ also observed very dark coloration of the German cockroach adults that resulted from nymphs treated with FME during a 'critical stage' of the last instar. WHITE¹⁰ observed that the

⁷ R. SOCHA and F. SEHNAL, J. Insect Physiol. 19, 1449 (1973).

⁸ A. STOCK and A. F. O'FARRELL, 14th Int. Congr. Entomol., Canberra (Australia), August 1972 (mimeographed circular).

⁹ D. F. WHITE and J. M. GREGORY, J. Insect Physiol. 18, 1599 (1972).

¹⁰ D. F. WHITE, J. Insect Physiol. 14, 901 (1968).



Figures 7-10. Internal reproductive system of adult female. 7. Normal adult. 8-10. Treated adults (topically with Ro 20-3600 4 μ l/nymph).

'juvenilized adults' of the cabbage aphid, *B. brassicae*, had only partially pigmented wings. BAGLEY and BAUERNFEIND¹¹, in their field tests with JHA, found the larvae of the tobacco hornworm, *Manduca sexta*, to be lacking in melanization, and this effect has been independently used by TRUMAN et al.¹² in developing an ultrasensitive bioassay for juvenile hormone.

It is important to note that the same JHA (Ro 20-3600) caused excessive melanization in the German cockroach, but prevented melanization in the tobacco hornworm larvae. Whenever colour patterns are affected, it seems that the exogenous JHA either induces or prevents the cuticular pigment synthesis, depending on the insect species and age.

Excessive darkening of the cuticle is generally assumed to be due to the deposition of melanin pigment. However, the darkening due to sclerotization cannot be totally disregarded¹³. The exact relationship between sclerotization and melanization is not known at present, although tyrosine injected into the insect body during the later part of ecdysis is primarily incorporated into the melanized areas of *Gryllus bimaculatus*¹⁴ and *Schistocerca gregaria*¹⁵. The brain, corpora allata and the prothoracic glands are variously implicated in pigment synthesis and colour changes, depending on the insect species, sex, age, and environmental stimuli, the epidermis being the 'target organ' for hormone¹⁶⁻¹⁸. In *Periplaneta americana*, an adult cuticular tanning hormone is known to be released from the terminal abdominal ganglion¹⁹, and is comparable to the 'Bursicon' of blow flies²⁰⁻²³. Experiments are in progress to find out how the exogenous JHA could alter the normal pigment synthesis and the bursicon-mediated cuticular processes.

In insects, the juvenile hormone controls vitellogenin (female-specific protein) synthesis, yolk deposition, accessory sex gland activity, and sex pheromone production^{24, 25}. In the JHA-treated adult females, development of ovarioles, oviducts and accessory sex glands is affected in varying degrees in individual adults. In some, the entire ovariole is attenuated, while in others the individual oocytes are atrophied or hypertrophied. The latter effects, unlike in a normal ovary (Figure 7), resulted in a highly irregular appearance of ovarioles within the same ovary (Figures 8 and 10). In some adults, the oocytes are uniformly overdeveloped (Figure 9). The mean length of the terminal (= basal) oocyte was 2.2 mm as compared to 1.3 mm of the control insect (ca. 70% increase). Here, the growth of the oocytes apparently proceeded irreversibly beyond a critical stage. Hypertrophy of the lateral oviducts (as well as of the calyx) was observed in some cases (Figure 8), and in some, the oocytes were clogged in the lateral oviducts (Figure 10).

The left accessory sex gland in the adult female cockroach produces a structural protein and the precursor of a phenolic tanning agent, viz., protocatechuic acid glucoside, while the right accessory sex gland secretes the glucosidase. From these secretions, the ootheca is synthesized inside the genital chamber²⁶⁻²⁹. The development of these glands in the treated females was inhibited in some cases, and in a few others hypertrophy resulted. In the latter situation, the tubules were not only larger but also appeared more whitish and dense. A few treated females tended to oviposit naked masses of eggs, a condition suggestive of malfunction in the formation of oothecae. ROHDENDORF and SEHNAL^{30, 31} recently reported several JHA-induced effects in *Thermobia domestica* and considered loss of control over cell growth and multiplication as some of the probable causes.

The influence of extremely low titer of JH during metamorphosis (which is typical of a normal insect) on

the reproductive organs is not known at present. It appears that, like the cuticular epithelium, these organs are relieved of the JH influence at metamorphosis until the JH titer builds up again in the adult. The diversity of abnormalities observed among the treated females in our present study can be partly attributed to the age difference between the individual nymphs at the time of treatment. However, variations within the same organ, such as the hypertrophy and atrophy of the ovarioles within the same ovary, need further research for a clear explanation.

Of great practical importance is the fact that the treated females lost the capacity to mate, regardless of the extent of abnormalities in the internal and external structures. Unmated normal males responded with typical mating behaviour when paired with the treated females, suggesting that the females were attractive to the males. However, the females were quite unresponsive to the males, and no mating occurred. Experiments of ROTH and BARTH³² indicated that, while corpora allata of the female cockroach may control extrinsically the expression of her receptivity, they are not directly involved in producing a state of sexual receptivity, and that the neurosecretory system may control the receptivity. It appears, therefore, that the exogenous JHA is in some way acting against the neurosecretions, leading to the loss of sexual receptivity in the treated females.

Zusammenfassung. Vergleichende Beschreibung der die Reifung und die Fortpflanzung verzögernden bzw. verhindernden Effekte von drei Juvenilhormonanaloga bei *Blattella germanica*.

Y. T. DAS and A. P. GUPTA³³

Department of Entomology & Economic Zoology,
Rutgers University, New Brunswick (New Jersey 08903,
USA), 26 February 1974.

¹¹ R. W. BAGLEY and J. C. BAUERNFEIND, in *Insect Juvenile Hormones* (Eds. J. J. MENN and M. BEROZA; Academic Press, New York 1972), p. 113.

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¹⁸ S. FUZEAU-BRAESCH, *A. Rev. Ent.* 17, 403 (1972).

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²² G. FRAENKEL and C. HSIAO, *Science* 138, 27 (1962).

²³ G. FRAENKEL and C. HSIAO, *J. Insect Physiol.* 11, 513 (1965).

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²⁵ W. J. BELL and R. H. BARTH JR., *J. Insect Physiol.* 16, 2303 (1970).

²⁶ J. H. WILLIS and P. C. J. BRUNET, *J. exp. Biol.* 44, 363 (1966).

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²⁸ M. ZALOKAR, *J. Insect Physiol.* 14, 1177 (1968).

²⁹ E. SHAYYA and D. BODENSTEIN, *J. exp. Zool.* 170, 281 (1969).

³⁰ E. B. ROHDENDORF and F. SEHNAL, *Experientia* 28, 1099 (1972).

³¹ E. B. ROHDENDORF and F. SEHNAL, *J. Insect Physiol.* 19, 37 (1973).

³² L. M. ROTH and R. H. BARTH JR., *J. Insect Physiol.* 10, 965 (1964).

³³ We thank Miss DEBBIE A. WAKSMUNDZKI for her able secretarial help.