Material and methods. Female immature rats, weighing 50 g, were used in the present experiments. A solution of estradiol-17β in 5% ethanol-saline was injected into the jugular vein under ether anesthesia, using a dosage of 30 μg/100 g b.wt. The control rats were similarly injected with equal amounts of the vehicle. The propranolol-treated animals were i.p. injected with 50 μg DL-propranolol (in saline)/100 g b.wt 20 min prior to the estrogen or vehicle injection.

The animals were killed 6 h after estrogen (or vehicle) administration and the uteri excized. The right uterine horn was used for biochemical studies and the left uterine horn was fixed in neutral formalin for subsequent histological studies[17].

The following parameters were measured for each animal: uterine wet wt, DNA[25], RNA[26], protein[27] and glycogen[28] content, and total number of uterine eosinophils[16]. The increases in uterine wet wt, RNA per unit of DNA, protein per unit of DNA and glycogen per unit of DNA were expressed as percent change over the controls. The uterine eosinophilia were expressed as the total number of eosinophils in the uterus.

Results. DL-Propranolol, injected i.p. 20 min prior to the estrogen injection, does not block the estrogen-induced uterine eosinophilia, the uterine wet wt response or the estrogen-induced increases in uterine RNA and protein contents ($p < 0.001$, $p < 0.01$, $p < 0.025$ and $p < 0.05$ respectively as compared to controls without estrogen injection) (table). The differences in glycogen content between estrogen and estrogen+propranolol-treated animals, as well as those between control and estrogen-treated animals, are not statistically significant ($p > 0.05$) (table).

Discussion. Our results show that a pretreatment with propranolol does not block the estrogen-induced uterine eosinophilia, the water imbibition effect nor the increases in uterine RNA and protein contents. It was previously shown that a similar pretreatment with propranolol suppresses the estrogen-induced increase in uterine cAMP[3,7]. Therefore, it can be assumed that the estrogen-induced uterine eosinophilia and the water imbibition effect (proposed by one of us to be mediated by the eosinophil receptor system[13,15,17]), and the estrogen-induced increases in uterine RNA and protein contents (generally considered to be mediated by the cytosol-nuclear estrogen receptor system[18–20]) are independent of the estrogen-induced increase in uterine cAMP content.

The differences in glycogen content between estrogen and estrogen+propranolol-treated animals were not statistically significant in our experiments ($p > 0.05$). It was, however, previously shown that exogenously administered cAMP produces an estradiol-like induction of several glycogenolytic enzymes[8,9] and that theophylline, a drug that promotes cAMP accumulation by inhibiting phosphodiesterase, potentiates the action of submaximal doses of estradiol on several uterine glycogenolytic enzymes[29]. This suggests that cAMP is involved in this estrogenic effect, probably as a separate mechanism of estrogen action. Our previous studies with cortisol-treated animals have also demonstrated the independence of the glycogen effect from estrogen-induced uterine eosinophilia[30]; and our experiments with estradiol and estriol have suggested the possibility that the glycogen effect is independent of the cytosol-nuclear estrogen receptor system[31].

Our previous studies have shown that cortisol drastically decreases the estrogen-induced uterine eosinophilia and water imbibition responses[30]. It was suggested that the cortisol-induced blood eosinopenia limits the number of eosinophils entering the uterus after estrogen administration, thereby limiting all estrogen responses assumed to be dependent on the eosinophil estrogen receptor system (i.e. water imbibition effect)[30]. Alternatively, the lysosome membrane-stabilizing properties of cortisol could account for this antiestrogenic effect of cortisol[32]. Our present results permit us to discard the latter possibility since propranolol, a drug with lysosome membrane-stabilizing properties similar to cortisol[11], failed to inhibit both the uterine eosinophilia and the water imbibition estrogenic effects.

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Reversal of metamorphosis in mealy bugs treated with juvenile hormone-active insect growth regulator[1]

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Summary. Anal pore-plates are a typical adult characteristic of male mealy bugs. Nymphs treated with a juvenile hormone-active insect growth regulator moult several times into intermediary forms between nymph and adult. The number of the anal pores is reduced during each of these supernumerary moults.

The postembryonic development of the mealy bugs is a rather complicated process. The first 2 larvel stages of both sexes are similar; in the females even the 3rd and 4th instars – the latter may be regarded as being the neotenic adult – still retain the same basic form. The male larvae of the 3rd and 4th instars, called pronymph and nymph, develop the wing buds as the first adult characteristics. The ventral part of the anal segment remains, however, still smooth as in younger instars (figure 1). The nymph moults into the adult male which is, besides the fully developed wings, characterized by 2 plates on the ventral side of the anal segment with numerous pores (80–100) (figure 2).

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The juvenile hormone (JH) and the JH-active insect growth regulators (IGRs) inhibit metamorphosis and often ecdysis in many other insects. The mealy bugs and several other species of scale insects were found to be very sensitive[3]. The deviations from the normal scheme of development observed on the treated larvae are the subject of the present paper.

Materials and methods. Citrus mealy bugs (*Planoccocus citri*) were reared on the potato tubers[4], dipped into an aqueous emulsion of 6,7-epoxy-3-ethyl-1-(p-ethyl-phenoxy)-7-methylnonane, cis/trans mixture (a constituent of epofenonane)[5]. The material for microscopy was treated with lactic acid and mounted in lactophenol medium.

Results and discussion. The development of the population of citrus mealy bugs reared on potatoes treated with

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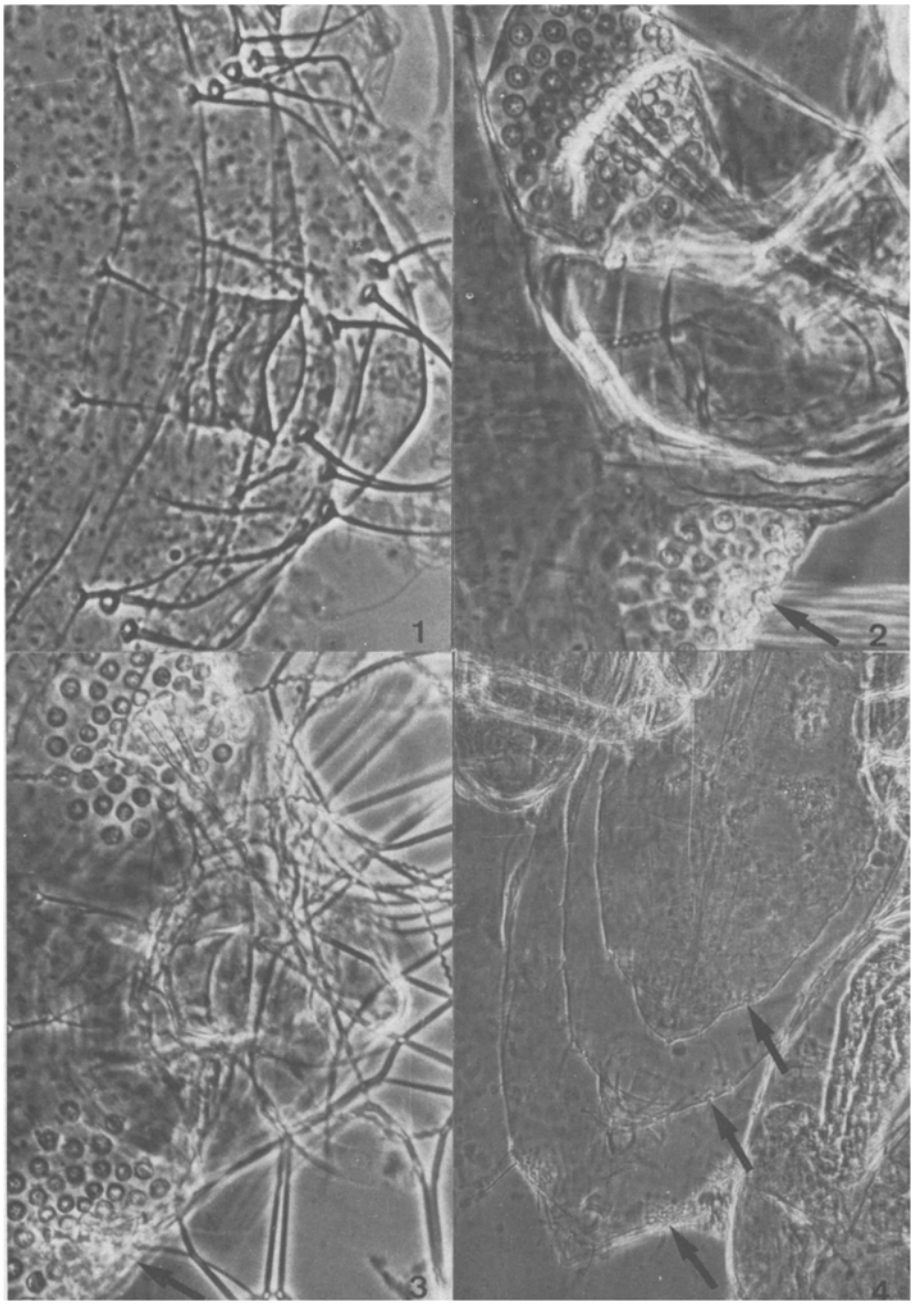

Fig. 1–4. Tip of the ventral part of the male abdomen of citrus mealy bug. Lactophenol. The anal plates with pores are indicated by arrow.
Fig. 1. Nymph, untreated. ×740.
Fig. 2. Male adult, untreated. ×740.
Fig. 3. Intermediate nymph-adult, JH-active IGR treatment. ×740.
Fig. 4. 3 successive nymphal-adult intermediates ecdysing without eclosion, JH-active IGR treatment. Note the anal plates of the oldest outermost skin of intermediate I with numerous pores and the plates of the innermost skin of intermediate III with very few pores. ×185.

0.001% emulsion of the IGR was completely stopped. Many larvae died already in the first ecdysis, and those of the male larvae, which passed through, proceeded successfully only to the nymphal stage but failed to metamorphose into adults.

The affected nymphs died mostly in ecdysis but some of them succeeded actually in moulting into intermediary forms between the supernumerary nymphs and adults (intermediates I) characterized by nymphal-like short wing buds and adult-like anal plates at the abdominal tip covered with pores (figure 3). The intermediates started 1–2 additional ecdyses which were never finished with eclosion. The lactophenol whole mounts often revealed, underneath the skin of the intermediates I, 1–2 cuticular layers of intermediates II and III. Thus the morphogenetic changes occurring during 2–3 supernumerary instars could be studied on the same animal (figure 4). The most striking feature was the reduction in the number of pores on the anal plates in the succession from the intermediate I (uppermost skin) with about half the number (35–50) to the intermediate III (the very inner skin) with only few pores left (0–3). Simultaneously, also the tip of the abdomen is getting more blunt in each successive stage.

The anal pore plates of the male mealy bugs are the adult characteristic which differentiates during the nymphal metamorphosis instar. The reduction of the number of these pores in 2–3 successive nymphal-adult intermediary instars is a new example of reversal of an adult structure induced by a JH-active substance in an intact insect. This observation is consistent with the results of experiments which proved that the adult epidermis has the capacity to secrete juvenile cuticle when transplanted into or connected with the larval milieu[6–11]. JH appears to be responsible for these changes as JH-active IGRs applied to *Tenebrio* pupae cause a 'larvalization' of the cuticle[12]. All these results prove that JH and JH-active IGRs possess an intrinsic morphogenetic activity which is exerted directly on the target tissues. Many parts of the epidermis remain capable of reverting from the more differentiated type back to the larval pattern under the influence of JH; the other parts are early committed irrevocably to lay down imaginal structures[13].

The JH-induced reversal of metamorphosis can be well explained by an activation of specific genes of conservative characteristics[14]. Another concept which views the JH as a repressor of the new genetic information, the status quo concept[15,16], leaves no room for any such reversion of the once attained degree of differentiation. Apart from its direct morphogenetic effect, JH seems to influence the metamorphosis also indirectly by reducing the titre of the moulting and differentiation hormone ecdysone in the blood[17]. This dual role of JH explains the inhibition of ecdysis occurring parallel to the morphogenetic changes in mealy bugs and many other insects treated with JH-active IGRs. On the other hand, the ecdysone inhibiting role of JH, separated from the morphogenetic activity, is demonstrated by the death of the young larvae in the first ecdysis in mealy bugs[3], scale insects[18,19], roaches[20] and other insects[21] treated with JH-active IGRs.

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Juvenile hormone: Evidence for a role in the feeding rhythm of *Oncopeltus fasciatus* (Dallas)

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Summary. The diurnal feeding rhythm of female milkweed bugs was damped when fed nonhost seeds. Juvenoid treatment partially restored the normal rhythm. Precocene II-treated milkweed seed-fed females fed arrhythmicly during the light phase without reduced total feeding activity. This effect was largely prevented by simultaneous treatment with JH III.

Little is known about the role of hormones in insect circadian rhythms. Possible neuroendocrine coupling in the circadian locomotor rhythms of a cockroach[2] and a cricket[3], as well as for spermatophore production in crickets[4] has been proposed. However, it has been pointed out[5,6] that the evidence is in no instance entirely consistent nor complete and hence needs further study. Truman and Riddiford's[7] demonstration of the involvement of a diffusable brain hormone in the circadian eclosion behaviour of silkmoths thus stands as the only undisputed example of involvement of a hormone in an insect behavioural circadian rhythm.

During the first week after eclosion, adult females of the large milkweed bug, *Oncopeltus fasciatus* (Dallas), gradually

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