munoreactive for PP. Since secretory granules are active in the proteolytic processing of the peptide and precursor it is possible that the proportions of co-existing peptides will vary so that one peptide may occur at times to the exclusion of the other. Ultrastructural characterization of these cells is in progress.

Cells which were immunoreactive for glucagon and somatostatin occurred to a similar extent in foetal and adult tissue. An ultrastructural study on the baboon endocrine pancreas has revealed that D cells contain a few A type granules amongst their normal secretory granules[9]. These granules have been found to be immunoreactive for glucagon (fig. 4). In foetal tissue PYY and PP occurred mainly in separate populations of cells with only an occasional overlap of activity.

Absorption of antisera with their respective homologous antigens prevented immunolabelling and all method controls were negative.

Discussion. The antiserum to PP was specifically raised against the C terminal hexapeptide and was shown, using immunoblotting and absorption studies, not to cross react with peptide YY (PYY), NPY or any known gastrointestinal hormone. Using this specific antiserum together with the absorbed antisera it was possible to view the results with a degree of confidence in their specificity. A slight cross reactivity between antiserum to glucagon and somatostatin antigen and between antiserum to somatostatin and glucagon antigen had been indicated in immunoblotting tests. Absorption of these antisera with their heterologous antigens eliminated this problem.

A possible simultaneous occurrence of multiple messengers in certain cell systems has been suggested and disputed for some years[10]. The co-existence of regulatory peptides can either be anticipated, in that it reflects the proteolysis of a known percursor, or it can be unpredictable. The co-existence of PYY and glucagon would be an example of truly unrelated peptides. It has been suggested that PYY and PP share antigenic determinants and that this might explain the reports of the co-existence of glucagon and PP. PYY has been found to co-exist in the large majority, if not all, of glucagon/glicentin containing cells in the mammalian colon[11] and in foetal cells from the pancreas and gut of many species from fish to primate[12]. The antiserum to PP used in our study does not cross react, however, with PYY. Our finding of cells in which glucagon and PP co-exist in both adult and foetus could be yet another case of co-existence of truly unrelated peptides and to our knowledge is the first report of any such cells in the adult primate pancreas.

It is tempting to join Kaung[2–4] in his speculation that at some time in vertebrate evolution, the two peptides were handled simultaneously in their production and that these glucagon-PP cells represent a primitive cell type still existing in small numbers in the rat, the mouse and in the baboon. Further investigation may well reveal their presence in other species. The detection in some D cells of secretory granules, which have the appearance of and are immunoreactive for glucagon, is interesting in that D cells have been described in the past as modified A cells.

Before a definitive identification of cells producing multiple peptide hormones is possible a demonstration of their synthesis is essential[10]. In situ hybridization is virtually the only way that this can be done in such a mixed population of cells and results from the application of this technique will be presented in the near future.

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Differences in the stimulation by calcium ionophore of juvenile hormone III release from corpora allata of solitarious and gregarious *Locusta migratoria*

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Summary. Incubation of the calcium ionophore A23187 resulted in an increase in the median rate of juvenile hormone III release by corpora allata (CA) of both gregarious and solitarious adult *Locusta migratoria* females at 3, 5 and 8 days after fledging. At all 3 datapoints, the enhancement of release rates was highly significant for CA from gregarious females but not significant for CA from solitarious females.
Key words. Juvenile hormone; corpora allata; locusts; *Locusta*; phase polymorphism; ionophore.

Locust species are characterized by the ability to exist in two phases (solitarious and gregarious) which differ in their biology. In *Locusta migratoria*, the differences in reproductive physiology between the two phases are particularly profound. Solitarious females are more fecund and more fertile[1, 2] and the first wave of oocytes matures much more swiftly in solitarious females[3].

Differences in corpus allatum (CA) activity and juvenile hormone (JH) titre between solitarious and gregarious locusts have been considered to be causative of many of the phenomena associated with phase polymorphism. It has been proposed that the solitarious phase is neotenous and is characterized by a high JH titre[4]. Bioassays of JH titre[5, 6] appeared to confirm this proposal. However, a study using

radioimmunoassay techniques [7] did not reveal consistent differences in JH titres between solitarious and gregarious *L. migratoria* larvae and adults. Moreover, it has not been possible to show differences in the rates of JH release from CA of gregarious and solitarious *L. migratoria* earlier than day 8 after fledging, whilst significant differences in the rate of oocyte maturation were detectable much earlier in the first gonotrophic cycle [3].

Assays of CA activity in locusts generally reveal very considerable variability in rates of JH biosynthesis and release [3, 8, 9] between individuals of the same sex and age, irrespective of phase. It has been argued [3] that, as has been suggested for *Nauphoeta cinerea* [10] and *Leptinotarsa decemlineata* [11], this variability may reflect a complex, possibly pulsatile, pattern of JH release from the CA.

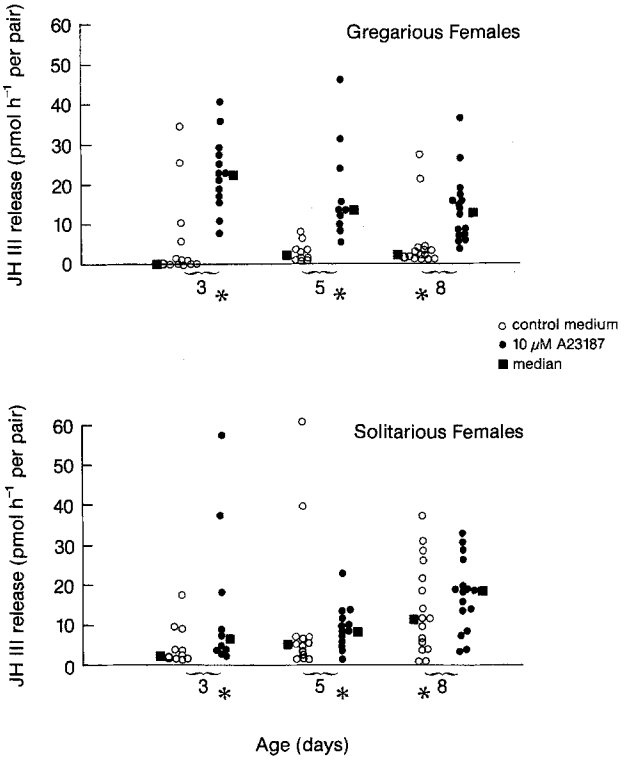
Calcium has been shown to play a role as a regulator of hormone production in cells of the prothoracic glands of *Manduca sexta* [13] and in the CA of *Diploptera punctata* [14]. Furthermore it has been found [12] that treatment in vitro of CA of *L. migratoria* with the calcium ionophore A23187 results in a dramatic stimulation of JH biosynthesis and release. This finding suggested a new approach to the investigation of CA activity in gregarious and solitarious *L. migratoria*, specifically, a comparison of the capacity of the CA of locusts of the two phases to be stimulated by ionophore treatment.

Materials and methods. Rearing conditions for *L. migratoria* were as described previously [3]. Gregarious females were raised in crowded conditions in 60-l cages. Solitarious females (which were from stocks which had been raised isolated for at least two generations) were raised isolated in 1.6 litre cages. All solitarious females were virgin at the time of JH assay.

JH III release in vitro was assayed as described earlier [15 - 17]. Incubations had a total duration of 180 min. However, separate determinations of JH release by each pair of CA were carried out using the medium in which the glands had been incubated for the first 90 min of the assay and that in which they had been incubated for the second 90 min. Incubation in the first 90 min was in standard TC 199 (GIBCO) (L-methionine concentration, 300 µM; calcium concentration, 1.3 mM), incubation in the second 90-min period was in standard TC 199 plus 10 µM A23187 in 0.1% DMSO. It has been shown [12] that 0.1% DMSO alone does not stimulate the rate of JH III release by CA from *L. migratoria* adults and that the stimulation due to A23187 manifests itself within 90 min of addition of the ionophore. As before [12], the effect of the ionophore on JH III release was assessed by comparing the rates of release during incubation with A23187 with those during the initial incubation in TC 199 alone. The significance of differences in rates was determined using Mann Whitney U-tests [3, 12]. In control medium, JH III release from *L. migratoria* CA has been shown to be linear for up to 6 h [8].

Results and discussion. Results are shown in the figure. It can be seen that in all cases, incubation in ionophore resulted in an increase in the median rate of JH III release. The magnitude of the increase in rates of release varied, however, according to phase. For CA from gregarious females at all datapoints, the release rates during incubation in the ionophore ('stimulated' rates) were very significantly higher ($p < 0.002$ in all cases) than rates during the initial incubation in control medium ('initial' rates). In contrast, the enhancement was not significant at any point for CA from solitarious females. Control incubations showed that 0.1% DMSO alone did not significantly enhance JH III release in females of either phase at any datapoint (data not shown). It was noticable that the median unstimulated ('initial') rates of JH release for the gregarious females at all datapoints were lower than median rates measured previously [3] for gregarious females of the same age. This was particularly apparent at day 8 after fledging. Nonetheless, the distribution of values for individual pairs of CA, at least at days 3 and 8, was typical of that found previously in locusts [8, 21] – i.e., a non-normal distribution with a small subset of CA showing high release rates and a larger group showing considerably lower rates. In gregarious adult female *L. migratoria*, higher rates of JH biosynthesis and release are generally associated with rapid vitellogenic growth of basal oocytes and occur towards the end of the first gonotrophic cycle [9]. At the beginning of the cycle, when there is little growth of the basal oocytes, rates are in general very low [3]. It is also known that the time to sexual maturation (and oocyte production) can vary considerably between groups of gregarious females [1]. We suggest that the low 'initial' rates of JH release found for the gregarious females in this study may reflect this variability, with this particular group of gregarious females being characterized by an extended period of sexual maturation and little basal oocyte growth [21].

Ionophore treatment of CA from gregarious females resulted, at all datapoints, in a marked stimulation of JH release. However, the degree of stimulation was particularly profound at 3 days (when, in general, the 'initial' rates of release were very low) and indeed, the median release rate of stimulated CA was higher at day 3 than at days 5 or 8. Interestingly, it has also been shown [18] that it is possible to stimulate JH release from CA of newly emerged adult gregarious *L. migratoria* by incubation with farnesoic acid – these rates of release are broadly comparable to those observed for gregar-



Rates of JH III release at 3 points during the first gonotrophic cycle by pairs of CA from gregarious and solitarious adult female *L. migratoria* during sequential 90 min incubation in, first, control medium (open circles) and, second, 10 µM A23187 (closed circles). Median values are also shown. For data from gregarious females, rates of release at each age by control and ionophore-treated CA are significantly different ($p < 0.002$). Rates of release by CA from gregarious and solitarious females are also significantly different ($p < 0.05$) on day 3 (A23187-treated), day 5 (A23187-treated) and day 8 (control untreated) only, as indicated by the asterisk.

ious day-3 females in the present study following treatment with A23187. It has been proposed [19], that exogenous farnesoic acid stimulation of *L. migratoria* CA is equivalent to that by a putative allatotropin [20], the latter possibly being involved in activating rate-limiting steps in the biosynthesis of JH prior to farnesoic acid synthesis. However, the mechanisms of stimulation in both cases remain to be defined, although both allatotropin- and A23187-stimulation probably involve enhancement of rate-limiting steps. Nonetheless, JH biosynthesis by CA from young adult females of *L. migratoria* can be stimulated, as the occurrence of a proportion of CA showing high release rates at this age would suggest; CA from these young females may therefore possess the potential to biosynthesize JH at high rates, although this potential is apparently not expressed continuously. Indeed, it has been suggested [18, 19] that JH production in adult *L. migratoria* is a function of two independent variables – stimulation of the CA by allatotropic substances (effective on a short-term basis) and the development, during early adult life, of responsiveness to allatotropin. The present data indicate that, in the case of gregarious females, the ability to respond to calcium ionophore has developed by day 3 of adult life. In solitarious females, the median rate of JH release in the presence of A23187 was higher than that during initial incubation in control medium at all datapoints. In no case, however, was the difference between 'initial' and 'stimulated' rates significant. Nonetheless, initial median rates of JH release were higher in solitarious than in gregarious females. This suggests that incubation in ionophore resulted in a significant elevation in release rates by CA of gregarious animals only because 'initial' rates were lower in these animals. It should be noted, however, that 'initial' rates were only significantly higher in solitarious females on day 8 (fig.) whereas 'stimulated' rates were significantly higher in gregarious animals on days 3 and 5.

In conclusion, treatment of CA with A23187 significantly stimulates JH III biosynthesis in CA from gregarious but not solitarious *L. migratoria* on days 3, 5 and 8. This difference between the phases reflects a higher 'stimulated' rate of biosynthesis in gregarious females on days 3 and 5 and a higher normal rate of biosynthesis in solitarious females on day 8. The cause of this difference between the phases may be complex and may reflect the interaction of more than one factor.

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Effect of dexamethasone on Fc γ receptor expression in foetal and neonatal rat gut

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Summary. When injected into 12-day-old suckling rats, dexamethasone caused a precocious disappearance of Fc γ receptors from enterocytes of the proximal small intestine. However, dexamethasone appeared to be necessary for the maintenance or production of such receptors in foetal rat gut cultured in vitro.
Key words. Fc γ receptor; rat gut; dexamethasone; IgG binding; enterocyte.

Transmission of maternal IgG across the gut of the suckling rat is effected in part by Fc γ receptors present on enterocytes of the duodenum and jejunum but absent from the ileum [1]. These receptors bind IgG optimally at pH 6.0 as shown by in vitro assays [2–6], but do not bind IgG at pH 7.2–7.4 and thus are well adapted to the pH prevailing in the lumen of the proximal small intestine [7]. Following binding, receptor-mediated endocytic processes effect the transcytosis of intact IgG across the enterocyte [8]. In vivo, antibody transport ceases after 21 days [9] and this is correlated with an absence of detectable Fc γ receptors on enterocytes obtained from rat gut beyond this age [2, 4, 10, 11]. Antibody transport also takes place across the gut of the late foetal rat [12] and we have recently shown that Fc γ receptors first become detectable on enterocytes in the most proximal region of the small intestine of the 20–21-day-old foetus [11]. Glucocorticoids have been implicated in the termination of Fc γ receptor expression, since in the 21-day-old neonatal rat there is a rise in the level of free plasma corticosterone [13] and injection of cortisone acetate into neonatal rats advances the time of closure of the