partially restored the rhythm on the nonhost diets, primarily by increasing feeding during the late photophase. Precocene II caused an arrhythmic feeding pattern (figure, C), without reducing total feeding activity (table), while simultaneous treatment with the juvenoid or JH III largely restored the normal rhythm (figure, D). Furthermore, when JH-III-treatment was discontinued after the 6th day, the rhythm was essentially eliminated between the 12th and the 30th day (figure, C), indicating that JH was not just involved in the initiation of the rhythm.

Discussion. One must consider the possibility that the feeding rhythm may be influenced by the oviposition rhythm, as the period of maximal oviposition coincides with the minimal feeding period during the photophase[8]. Since JH is also essential for ovarian maturation[12], JH deficiency might induce photophase feeding arrhythmicity indirectly by eliminating oviposition behaviour. However, a pronounced rhythmicity was evident in males and in virgin milkweed-fed females although they oviposited a mean of only 1.9 times during the 30-day-period. Also, as previously observed[8], cyclic feeding is initiated before oviposition begins, indicating that the 2 cycles are not obligatorily coupled.

Apparently, JH does not act as a proximal stimulus for development of the rhythm, as juvenoid treatment of young females did not cause its precocious appearance (unpublished data). Rhythm development is characterized by a reduction in feeding activity during times other than the peak evening hours which change relatively little[13]. This suggests a cyclic inhibitory influence is mainly responsible for the rhythmicity. This conclusion is in agreement with the marked increase in feeding activity during the afternoon (7 and 11 h) induced by precocene II treatment. Ironically, the results of the nonhost diet treatments, in themselves, suggest just the opposite: That the juvenoid accentuated rhythmicity primarily by stimulating peak evening feeding (figure, A). However, in view of the total evidence, the following interpretation seems more likely: The normal level of total feeding activity observed in precocene II-treated females (table) suggests that JH is not involved in the stimulatory effect of milkweed seeds on feeding activity. Apparently, juvenoids can largely or completely substitute for this specific seed effect in females when on nonhost diets, perhaps by some indirect means, such as stimulation of ovarian maturation. Thus, the more normal feeding activity pattern seen in juvenoid-treated females on nonhost diets appears to be due both to a general stimulatory effect on feeding and to the enhancement of a cyclic inhibitory influence.

It appears that the juvenoid treatments may not have completely prevented precocene II or dietary-induced rhythm damping, as in all 4 comparisons the 7/15 h feeding activity was lower than for the control juvenoid-treated females. This may well be due to a failure to restore a completely normal diurnal JH titre fluctuation.

While these experiments do not distinguish between a permissive vs. a regulatory role for JH in the feeding rhythm and do not exclude the possibility of an indirect mode of action, they do provide the first encouraging evidence that JH may be an essential component of some repetitive insect behavioural rhythms.

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Gastrin: Obligatory intermediate in the postprandial mobilization of gastric histamine in the rat[1]

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Summary. In unoperated fasted rats, feeding raised the serum gastrin concentration, reduced the gastric mucosal histamine content and activated the gastric histidine decarboxylase. The reduction of gastric histamine and activation of histidine decarboxylase was induced also by the injection of pentagastrin. In antrectomized rats, feeding failed to produce these effects. Injection of pentagastrin, however, still lowered gastric histamine and activated gastric histidine decarboxylase. Thus, antral gastrin seems to be an obligatory mediator of the postprandial activation of histidine decarboxylase and mobilization of histamine.

Histamine in the rat stomach is predominantly located in endocrine-like cells of the oxyntic mucosa[2-4]. Ultrastructurally, these cells comprise 2 types, distinguishable from each other by the morphology of their secretory granules[3,4]. These cell populations seem to respond to gastrin in the following 2 ways: 1. Gastrin mobilizes histamine and activates synthesis of the histamine-forming enzyme[4,5]. 2. Gastrin exerts trophic control of at least 1 of the 2 histamine-storing endocrine-like cell types[4].

Feeding after a period of fasting activates gastric histidine decarboxylase and elicits a marked but short-lasting reduction of the gastric histamine content[6]. This reduction in histamine content is thought to reflect mobilization of histamine[6]. It has been claimed that the effects of feeding on the histamine-storing cells are the result of direct vagal excitation in conjunction with the postprandial increase in the serum gastrin concentration[6,7].

In a series of studies, it has been demonstrated, however, that gastrin is the major intermediate in the activation of histidine decarboxylase, and that the contribution of

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the vagi is insignificant[4, 8–11]. This has been established in experiments using antrectomized rats, i.e. animals in which the endogenous gastrin stores have been almost completely removed. In the present study, we show that also the postprandial release of histamine from these cells requires gastrin and that in the absence of antral gastrin vagal excitation induced by feeding is without effect.

Methods. Adult male Sprague-Dawley or Wistar rats, weighing 150–200 g, were used. Radical antrectomy was performed on 30 Wistar rats by resection of the distal half of the glandular stomach (the pyloric gland area together with the adjacent portion of the oxyntic gland area) and the duodenal bulb[11]. Great care was taken to eliminate the entire lesser curvature, including the limiting ridge between the glandular and nonglandular mucosa at the cardia. The nervous and vascular supply to the remaining part of the stomach was spared. Gastrointestinal continuity was reestablished by gastro-duodenostomy end-to-end. The rats were allowed to recover for 2–4 weeks after surgery before being used in experiments. All rats were fed a standard diet of food pellets and tap water. Before the experiments, the rats were kept in individual cages with wire mesh bottoms and deprived of food but not water for 48 h. One group of rats remained fasted, another group received food for 1 h, a 3rd group received pentagastrin s.c. All animals were killed by exsanguination under diethyl ether anaesthesia. Blood was drawn from the abdominal aorta. Serum was lyophilized and stored in the deep freeze (– 25 °C) until analysis. The concentration of gastrin in serum was determined radioimmunochemically[12]. For details of measurement of gastrin in rat serum, see[5]. The oxyntic mucosa was scraped off the stomach wall and homogenized in ice-cold 0.1 M phosphate buffer, pH 6.9–7.0, to a final concentration of 100 mg (wet wt) per ml. The histamine content of these extracts was measured fluorometrically[13] and the histidine decarboxylase activity was determined radiometrically as described in detail elsewhere[13].

Results and discussion. The results of feeding following a period of fasting are summarized in figure 1. In unoperated fasted rats, feeding raised the serum gastrin concentration, reduced the gastric mucosal histamine concentration and activated the histidine decarboxylase. In the antrectomized rats, feeding did not elicit these effects (figure 1). In unoperated fasted rats, pentagastrin reduced gastric histamine and activated gastric histidine decar-

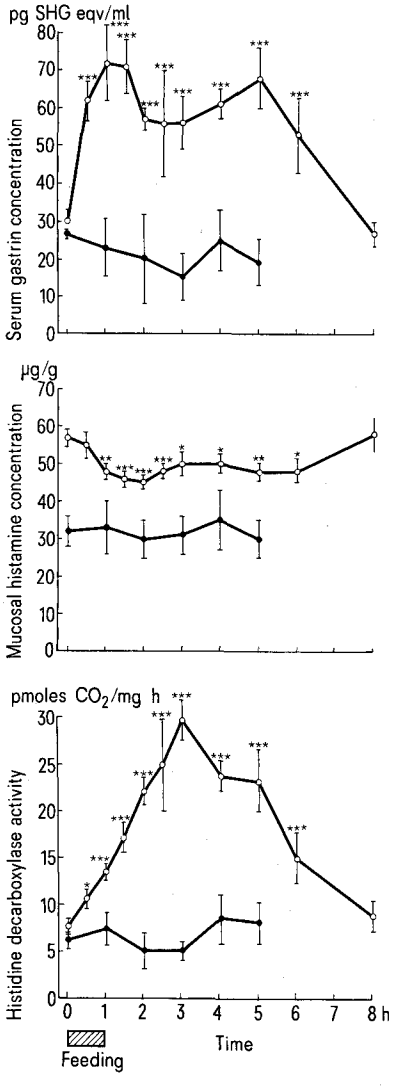

Fig. 1. Effect of feeding on serum gastrin concentration (upper panel), gastric mucosal histamine concentration (middle panel) and histidine decarboxylase activity (lower panel) in unoperated rats (○) and in antrectomized rats (●). The values are the mean of duplicate determinations on 4–40 animals. Vertical bars give SEM. Statistical significance for the difference between fasted rats and fed rats was assessed by Student's t-test and represented as * $0.01 < p < 0.05$, ** $0.001 < p < 0.01$ and *** $p < 0.001$.

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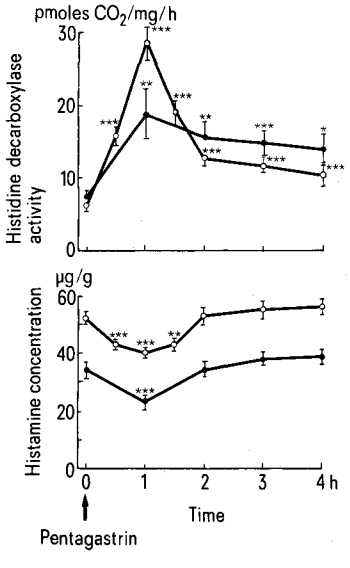

Fig. 2. Effect of pentagastrin on gastric mucosal histidine decarboxylase activity (upper panel) and histamine concentration (lower panel) in unoperated rats (○) and in antrectomized rats (●). The values are the mean of duplicate determinations on 4–30 animals. Vertical bars give SEM. For statistical details see figure 1.

boxylase. In the antrectomized rats, the basal histamine level was lower than in the control rats[4,14]; however, they still responded to pentagastrin with reduction of the histamine level and activation of histidine decarboxylase (figure 2).

The role of the vagi for the mobilization of mucosal histamine and activation of histidine decarboxylase in the rat stomach has been a subject of some controversy. There have been claims that the vagus affects the histamine-storing cells directly, and that e.g. the response to feeding is the conjunctive result of vagal excitation and increased serum gastrin concentration[6,7]. In previous publications, we have argued that the vagi do not contribute directly to the activation of histidine decarboxylase after feeding and that the enzyme activation induced by vagal excitation is indirect, being the result of gastrin release[4,10,11]. In the present study, we show that in antrectomized rats feeding fails to induce lowering of gastric histamine, whereas the histamine-lowering capacity of pentagastrin is retained also in the absence of antral gastrin. We have previously shown that antrectomy does not prevent vagal excitation from stimulating gastric acid secretion[14]. Thus, neither vagal excitation nor any other physiological mechanism induced by the process of feeding is able to mobilize gastric histamine in the absence of antral gastrin.

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Salivary gland enlargement as a test for a new way of permanent isoproterenol application

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Summary. Low doses of isoproterenol, given permanently by a new application-method, show the same effects on salivary-gland enlargement of rats throughout approx. 6 days than 10fold higher concentrations given by previous workers as single daily injections.

Since it is common knowledge, that large doses of isoproterenol (IS) lead to a considerable enlargement of the salivary-glands of rats[2], we used this well known reaction as a test to prove the effectiveness of a new kind of permanent application of IS.

Material and methods. A number of male and female Wistar rats of approx. 250 g were divided into 3 groups. The animals were laparatomized, and plastic capsules (Brand, Cat.-Nr. 780500) with a volume of 2 ml filled with IS-solutions or with solvent (Aq. dest. with a drop of 0.1 N HCl) and sealed with a dialysis membrane (Union Carbide) were put into the abdominal cavity. After the wounds have been closed, the animals were put back into their cages.

The first group received capsules containing an IS-solution of 4 mg/ml, the second capsules with a 1 mg/ml solution and the control group capsules that were filled with solvent only.

The number of animals shown in the figures were killed on 9 consecutive days at 12 h, the right parotid and submaxillary gland and the capsules were removed. The glands were weighed, cut and stained (H.E.) for histological examination and the remaining IS-concentration in the depot-capsules was determined using a slightly modification of the Vulpian-method[3]. Of the collected data, $\bar{x}$ and SEM values were estimated and a capsule-output diagram was plotted by a Univac-494 Computer, by courtesy of the Rechenzentrum Graz.

Results. Concerning the 8-mg-capsules it could be said that, though the output rates of the first 2 days, as can be seen in figure 1, greatly exceed those of the following days, a measurable flow rate, that can still be considered effectful, is maintained up to the 6th day, while the output of the 2-mg-capsules is naturally smaller but also measurable up to 16 h (figure 1).

1 We are very much indebted to Mrs K. Stökl and Miss E. Stangl for their valuable technical assistance.

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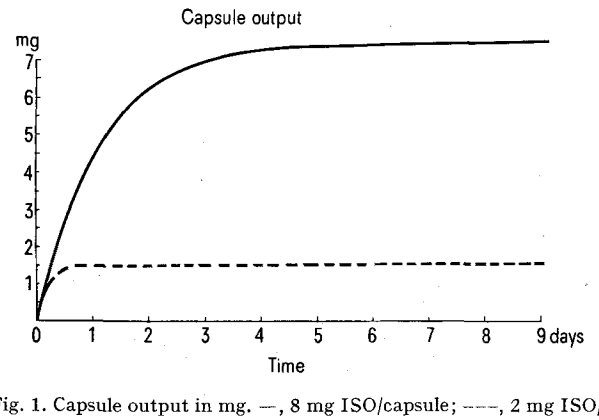

Fig. 1. Capsule output in mg. —, 8 mg ISO/capsule; – – –, 2 mg ISO/capsule. Plotted by an UNIVAC 494 Computer.

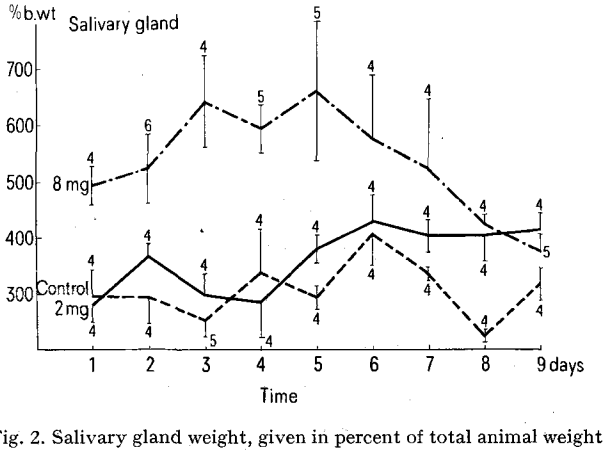

Fig. 2. Salivary gland weight, given in percent of total animal weight at killing time. – – –, Controls; —, 2-mg-capsules; –.–.–, 8-mg-capsules.