du méthimazole ($10^{-3}M$) dans le sang perfusé. Il y avait par contre une augmentation de la sécrétion d'-I* pendant la perfusion de $10^{-3}M$ d'ïodure, indiquant une décharge d'-I* pendant la perfusion de $10^{-3}M$ d'ïodure, indiquant une décharge d'-I* de provenance intra-thyroïdienne. Cet effet est similaire à celui noté auparavant pendant la perfusion de $10^{-3}M$ de ClO_4^-.

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* An asterisk indicates that only the radioiodinated component is considered.

Circadian Rhythm of Progesterone Secretion During Pseudogestation in the Rat

Accounts of studies of the endocrine function of the rat ovary, with particular reference to the secretion of progestins during pseudogestation, are given in the publications of Fajer and Barraclough[1] and Hashimoto et al.[2]. These authors, however, omit to mention the time of day at which the levels of progesterone and 20 α OH-progesterone were determined in their pseudopregnant animals.

In the light of a recently published observation by Freeman and Neill[3] to the effect that a nocturnal release of prolactin takes place in pseudopregnant rats, the present study was undertaken to find out to what extent progestin secretion displays a nychthemeral rhytm.

Material and methods. The experiments were performed on 25 female rats of strain SIV (Ivanovas, Kisslegg, Germany), weighing between 200 and 250 g, which were kept under conventional conditions in respect of temperature, humidity and lighting (light from 06.00 to 20.00 h) throughout the study. These females were mated with sterilized males and subsequently examined for the presence of a vaginal plug, to ensure that copulation had taken place. On the 7th day of pseudogestation they were divided into 4 groups. Every 6 h from 03.00 h onwards, 1 group was cannulated according to the method described by Fajer and Barraclough[1], the cannula being passed via the renal vein into the ovarian vein. To perform this operation on all the animals in 1 group took 2 h.

Blood samples of 0.5 ml were withdrawn slowly, over a period of 3–5 min, from each animal. The blood was extracted and chromatographed (3 × 10 cm migration on aluminium oxide in a cyclohexane: ethyl acetate solvent system) and the content of progesterone determined by a slightly modified version of the protein-binding assay method of Neill et al.[4]. After chromatographic separation, 20 α OH-progesterone was eluted in 5 ml of ethyl acetate and measured fluorometrically in sulphuric medium. Losses of progesterone and its metabolite due to the experimental procedures were estimated by the addition of ^{3}H-progesterone and ^{3}H 20α OH-progesterone in a ratio of 1:1.

The animals were killed after the blood samples had been taken, and the ovaries, corpora lutea and uteri were removed and weighed. The standard error of the mean was calculated according to Lord[5].

Results. The rate of progesterone secretion calculated from the blood levels determined between 03.00 and 05.00 h, 15.00 and 17.00 h and 21.00 and 23.00 h were very similar, ranging from 5.5 to 7.4 μg/h. The differences were not significant. These values may be taken as the basal rate of secretion of progesterone during this phase of pseudogestation[1]. Between 09.00 h and 11.00 h, however, an abrupt increase in the release of progesterone occurred. The mean rate of 19.4 μg/h measured during this period is significantly different from the foregoing values.

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Organ weights and production of progesterone and 20α OH-progesterone in the rat on 7th day of pseudogestation

Clock time	*n*	Body weight (mg)	Duration of cannulization (min)	Weight of ovary (mg)	Weight of corpora lutea (mg)	Weight of uterus (mg)	Blood flow (ml/h)	Progesterone (μg/h)	20 OH-Progesterone (μg/h)	Prog./20 OH-progesterone (ratio)
a) 03.00 to 05.00	5	236.8 ± 5.5	4.0 ± 2.0	39.7 ± 1.9	1.59 ± 0.07	282.3 ± 13.0	18.5 ± 6.3	6.0 ± 1.8	10.7 ± 2.3	0.56
b) 09.00 to 11.00	10	225.4 ± 3.0	3.3 ± 0.5	31.7 ± 1.5 (a–b)[a] $p < 0.01$	1.81 ± 0.08 (a–b) $p < 0.1$ n.s.	292.4 ± 16.3	13.3 ± 2.2	19.4 ± 4.2 (a–b) $p < 0.05$	6.5 ± 1.5	3.00
c) 15.00 to 17.00	5	225.6 ± 3.5	5.1 ± 1.2	31.3 ± 1.9 (a–c) $p < 0.01$	1.58 ± 0.07	260.0 ± 6.3	7.4 ± 1.4	5.5 ± 1.8	4.0 ± 1.2 (a–c) $p < 0.05$	1.37
d) 21.00 to 23.00	6	214.0 ± 14.4	4.0 ± 1.0	32.4 ± 1.0 (a–d) $p < 0.01$	1.59 ± 0.05	277.5 ± 22.9	9.2 ± 1.2	7.4 ± 1.5	3.4 ± 0.7 (a–d) $p < 0.01$	2.18

[a] Difference between values for groups indicated.

The secretion of 20α OH-progesterone diminished progressively from a maximum of 10.7 µg/h between 03.00 h and 05.00 h to a minimum of 3.4 µg/h between 21.00 and 23.00 h. The reductions recorded between 15.00 and 17.00 h (4.0 µg/h) and 21.00 and 23.00 h are significant. Only in the blood samples obtained between 03.00 and 05.00 h was the ratio of progesterone to 20α OH-progesterone less than unity; in all the other samples it was greater.

The weight of the ovary decreased markedly between 05.00 and 09.00 h and remained at the minimum level until 23.00 h (Table).

There appeared to be an increase in the weight of the corpora lutea between 09.00 and 11.00 h, coinciding with the maximum blood levels of progesterone.

Discussion. The basal rates of progesterone and 20α OH-progesterone secretion determined in this study on the 7th day of pseudogestation correspond to those published by FAJER and BARRACLOUGH[1]. Under the same conditions, that is to say in the rat on the same day of pseudogestation (but daily light from 05.00 to 17.00 h), FREEMAN and NEILL[3] demonstrated that the secretion of prolactin is at its greatest between 03.00 and 05.00 h; the release of progesterone by the ovary observed in our experiments (light from 06.00 to 20.00 h) took place 6 h later. It is possible that under our lighting conditions the prolactin peak has been somewhat shifted. It may be assumed that the inverse relation between the rising levels of progesterone and the declining 20α OH-progesterone levels is a result of the liberation of prolactin, since WIEST et al.[6] have shown that prolactin inhibits 20α OH-progesterone dehydrogenase, while according to PUPKIN et al.[7] this enzyme occurs principally in the corpora lutea. Moreover, the variations noted in progesterone: 20α OH-progesterone ratio would seem to be indicative not only of an increase in the de novo synthesis of progesterone but also of a reduction in its rate of conversion to 20α OH-progesterone. These results would then confirm BLENCOE and MOODY's[8] observation that prolactin increases the production of progesterone and causes a reduction in the weight of the ovaries. The variations in the progesterone: 20α OH-progesterone ratio which we noted during the 7th day of pseudogestation could hence be induced by the nocturnal peak of prolactin release. Since this peak has been shown to occur regularly each night throughout pseudogestation[3], it seems reasonable to suppose that peak levels of progesterone also occur daily until pseudogestation ceases.

A circadian rhythm is likewise evident in the weight of the ovaries and the level of 20α OH-progesterone in the ovarian blood. The hormonal conditions governing this phenomenon remain to be elucidated.

Résumé. Par des canulations toutes les 6 heures chez des rattes le 7ème jour de la pseudogestation nous avons pu mettre en évidence un pic de progestérone entre 09.00 et 11.00. Sur la base de nos résultats (progestérone, 20α-OH-progestérone, poids des ovaires et corps jaune) et de la publication de FREEMAN et NEILL[3] nous discutons le rôle de la LTH comme régulation possible de ce phénomène.

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The Role of Nutrition and Endocrine Activity in the Development of Eggs in *Culex fatigans*

A study of the role of nutrition and endocrine activity in the development of eggs of the common Indian mosquito *Culex fatigans* has produced a number of interesting observations. 1. Laboratory-bred female mosquitoes or unmated females obtained from field do not mate in captivity or feed on blood. Mating is essential for the female mosquito to take a blood meal, which is taken only once in the course of a reproductive cycle. Each successive cycle of reproduction must be preceded by ingestion of a blood meal. 2. Both sugar and blood meal cause an increase in the number of the median neurosecretory cells (MNSC). The density of MNSC is at its highest level at 27 h after a blood meal (250% of initial number) when the insect is ready to deposit eggs. The normal MNSC number is restored by 24 h after oviposition. 3. The MNSC of the pars intercerebralis have a major contribution towards the physiological events occurring during the periods of egg maturation and oviposition. However, the MNSC do not show any activity if the mosquito is fed on sugar only, but they show an increase in size in blood-fed mosquitoes with maximum size attained at 72 h after blood meal, at which time fully developed eggs are ready for liberation. 4. The MNSC of the pars intercerebralis are seen to be fully loaded with secretory material up to 48 h after a blood meal. Their maximum size at 72 h after a blood meal and their reduction in size immediately after oviposition clearly indicates that their secretion is released only few hours before oviposition. 5. Starvation and sugar meals fail to induce any activity in the corpora allata (CA), but 24 h after a blood meal, the CA reach their maximal activity, show a sharp decrease at 48 h and thereafter tend to resume the original state of inactivity. 6. Sugar meals do not initiate any development of eggs but ingestion of a blood meal results in an increase in the size of the terminal eggs in a geometrical progression reaching its maximum at 72 h (increase in egg length is 730%). 7. Though the CA activity is very much reduced by 48 h following a blood meal, the eggs continue to grow to their maximum size upto 72 h.

Culex fatigans is an anautogenous species and therefore it does not lay eggs unless allowed to ingest blood, which provides the necessary stimuli for the activation of the endocrine glands and development of oocytes. Several species of mosquitoes of the genera *Aedes*, *Anopheles* and *Culex* have been investigated to establish beyond doubt that egg development is controlled and regulated by hormones. Many earlier workers[1-4] have made very

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