Prothoracic glands: The prothoracic glands of S. litura are semi-transparent white and compact, as reported by Lee[8], with about 51 to 62 bead-like cells in each gland which has a small flattened body made up of a single row of 5 to 16 cells in the middle and double or more rows of cells towards the extremities which bifurcate anteriorly and posteriorly. 2 small-sized branches of anterior extremity bear 2 to 3 tiers of bead-cells which touch the posterior margin of the head. The branches of the posterior extremity are larger in size; the lateral one reaching the spiracle and the inner one terminating near the prothoracic ganglia. The prothoracic glands also increase in length in each larval instar from 0.702 mm in the 3rd instar to 1.503 mm in the 6th instar larva of S. litura, but they do not reveal any changes in their from either before or after a moult.

Cellular and subcellular localization of ^{3}H-diethylstilbestrol in the pituitary, oviduct and uterus of the female rat[1]

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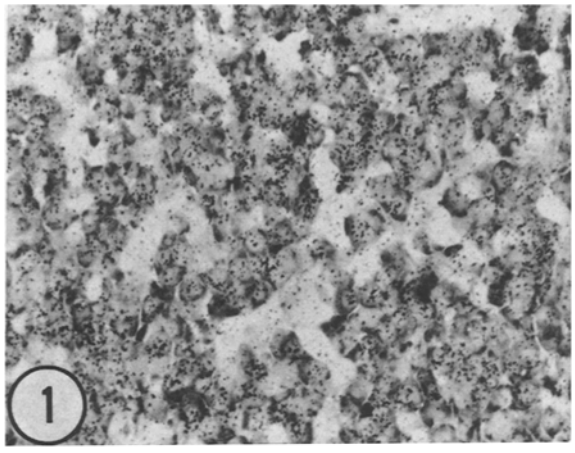
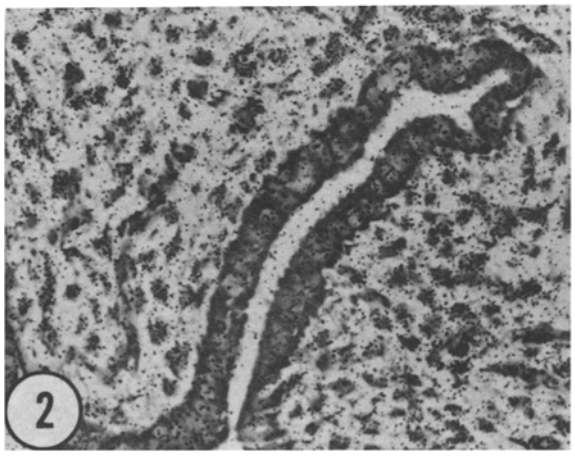
Summary. Female rats were injected with ^{3}H-diethylstilbestrol. The pituitaries, oviducts, and uteri were removed and processed for autoradiography. The nuclear uptake and retention in these tissues appeared similar to that seen after the injection of ^{3}H-estradiol except in the pars nervosa of the pituitary gland.

Numerous studies using biochemical techniques have demonstrated nuclear uptake and retention of ^{3}H-estradiol-17β by the pituitary gland[2,3], the oviduct and the uterus[4]. Autoradiographic techniques have revealed that the nuclear uptake and retention occurs in well over 60% of the cells of the pars distalis, a smaller number in the pars nervosa, of the pituitary gland[5], and in the connective tissue cells, muscle cells and epithelial cells of the oviduct and uterus[6]. Since the commonly used synthetic estrogen, diethylstilbestrol, has been shown to bind to the estrogen receptor in all of these tissues[3,7], it has been generally assumed that the same cells which take up estradiol-17β also take up diethylstilbestrol. The following study was conducted to determine whether the nuclear uptake and retention of this commonly used synthetic estrogen is the same as that of its natural counterpart.





Figs. 1 and 2. Autoradiograms of the pituitary (figure 1), pars distalis, and uterus (figure 2), from 25-day- old female rats, killed 2 h after the injection of ^{3}H-diethylstilbestrol. Exposure time 8 weeks. 4 μm. Stained with methylgreen-pyronin. $\times$400.

Methods. 25-day- old female rats (n = 5/group) were injected i.v. with 1.0 μg of ^{3}H-diethylstilbestrol (62.4 Ci/mM)/100 g b.wt only or in combination with a 15-fold greater dose of unlabeled estradiol-17β or unlabeled testosterone. The animals were killed 2 h after the injection, and the tissues processed for autoradiography. The autoradiographic procedure is described in detail elsewhere[8]. Briefly, the pituitaries, oviducts and uteri were excised, placed on brass tissue holders and simultaneously frozen and mounted by immersion in liquefied propane at about –180 °C. 4 micrometer sections were cut at –30 °C knife temperature in a Wide-Range Cyrostat (Harris Manufacturing Co., Cambridge, Massachusetts, USA). The frozen sections were then freeze-dried with a cyropump for 24 h, and then dry mounted by pressure with a teflon support on emulsion coated slides, previously stored over drierite. The slides were exposed at –15 °C for 8 weeks, then developed for 45 sec at 18 °C in Kodak D-19 developer, rinsed, and fixed for 5 min in Kodak fixer, and finally rinsed and stained with methylgreen-pyronin.

Results. Nuclear concentration of radioactivity after the injection of ^{3}H-diethylstilbestrol was found in well over 70% of the cells in the pars distalis, (figure 1) but not in either the pars intermedia or pars nervosa of the pituitary gland. Nuclear concentration of radioactivity was found in the connective tissue cells, muscle cells, and epithelial

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2 A. J. Eisenfeld, Endocrinology *86*, 1313 (1970).
3 K. S. Korach and T. G. Muldoon, Biochemistry *13*, 1932 (1974).
4 E. V. Jensen and E. R. DeSombre, Science *182*, 126 (1973).
5 W. E. Stumpf, Z. Zellforsch., Abst. Histochem. *92*, 23 (1968).
6 W. E. Stumpf, Endocrinology *85*, 31 (1969).
7 B. W. O'Malley and A. R. Means, Science *183*, 610 (1974).
8 W. E. Stumpf, Acta endocr. Suppl. *153*, 205 (1971).

cells of both the oviduct and uterus (figure 2). Simultaneous administration of a 15-fold greater dose of unlabeled estradiol-17β completely inhibited the nuclear concentration of radioactivity in all cells of all 3 tissues. On the other hand, simultaneous administration of a 15-fold greater dose of unlabeled testosterone had no effect on the nuclear concentration of ^{3}H-diethylstilbestrol in the 3 tissues.

Discussion. The pattern of labeled cells in the pituitary agrees well with that reported after estradiol injection except for the pars nervosa[5]. In contrast to the adult female rat[5], where a number of cells in the pars nervosa were found to be labeled after the injection of ^{3}H-estradiol-17β, no labeled cells were found in the pars nervosa after the injection of ^{3}H-diethylstilbestrol. These data are in agreement with the lack of nuclear concentration of radioactivity in the pars nervosa in the 2-day-old female rat after the injection of ^{3}H-estradiol-17β[9]. The nuclear concentration of radioactivity in the oviduct and uterus after the injection of ^{3}H-diethylstilbestrol is similar to that reported for ^{3}H-estradiol-17β. Thus, with the exception of the pars nervosa of the pituitary gland, the localization seen after the injection of ^{3}H-diethylstilbestrol appears similar to that found after the injection of ^{3}H-estradiol-17β.

The difference found in the pars nervosa is most likely due to 1 of 3 factors. First, it may be due to the structural differences between the 2 estrogens. A second possibility is that there are developmental differences between the adult and immature animal, perhaps in the hormonal milieu. Finally, the differences may be due to procedural differences such as variation in dose or exposure time.

9 P. J. Sheridan, M. Sar and W. E. Stumpf, Experientia *29*, 1415 (1973).

Section des nerfs paracardiaques internes et libération provoquée du facteur gonadotrope médian chez le Criquet migrateur

Section of the nervi corporis cardiaci I and provoked release of median gonadotropic factor in the African locust

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Summary. The in vivo electrostimulation of the pars intercerebralis of Locusta migratoria, after cutting the nervi corporis cardiaci I, does not provoke the release of the gonadotropic hormone from the cell bodies. The provoked release of the gonadotropic hormone recommences soon as the axon terminals are regenerated (reentry of the nerves into the original corpora cardiaca for all the histological controls). The electrostimulation of the pars intercerebralis seems to favour the bilateral reentry of the nerves into the corpora cardiaca.

Les cellules neurosécrétrices protocérébrales médianes de Locusta migratoria produisent un facteur gonadotrope[1]. La section des nerfs paracardiaques internes isole les péricaryones neurosécréteurs médians (groupés dans la pars intercerebralis) des extrémités axoniques (formant la partie neurosécrétrice des corpora cardiaca). Cette section empêche la maturation ovarienne du criquet quand elle n'est pas suivie de la régénération des nerfs[2]. L'effet physiologique de la libération du facteur gonadotrope nécessiterait donc l'intégrité des cellules neurosécrétrices médianes. Ce résultat est conforme au concept actuellement admis de la libération du produit de neurosécrétion aux extrémités axoniques pour les cellules neurosécrétrices du système nerveux central[3].

Cependant, une libération du produit de neurosécrétion à partir des corps cellulaires a été avancée par de nombreux histologistes avant l'apparition de la microscopie électronique (voir Takeda[4]). Une telle libération serait responsable de la rupture de la diapause chez Monema flavescens et viendrait s'ajouter à la libération axonale[4]. Chez Rhodnius, le ganglion mésothoracique renferme les péricaryones de cellules neurosécrétrices diurétiques. Sous l'influence du potassium, ce ganglion libère in vitro d'importantes quantités d'hormones diurétiques[5] même après ablation des nerfs abdominaux qui contiennent les extrémités axoniques des cellules neurosécrétrices.

Il nous a paru alors intéressant d'examiner si les péricaryones neurosécréteurs médians de Locusta, séparés des extrémités axoniques, étaient capables de libérer in vivo leur facteur gonadotrope sous l'influence de chocs électriques. Nous avons donc comparé, après pseudo-électrostimulation (témoins) et électrostimulation (stimulés) de la pars intercerebralis[6], les indices ovariens (longueur moyenne de 10 ovocytes basaux) de 99 criquets femelles adultes répartis en 8 séries: 4 séries de témoins et 4 séries de stimulés. Tous les criquets ont subi au jour 1 de leur vie imaginale une ouverture collaire suivie chez 6 séries seulement de la section bilatérale des nerfs paracardiaques internes. La pseudo-électrostimulation et l'électrostimulation ont été effectuées 1 jour (chez les 2 séries à nerfs intacts et chez 2 séries à nerfs sectionnés), 4 jours (chez 2 autres séries à nerfs sectionnés) et 6 jours (chez les 2 dernières séries à nerfs sectionnés) après l'ouverture collaire. Les indices ovariens ont été mesurés 6 jours après la stimulation chez les 4 séries à stimulation précoce et 4 jours après la stimulation chez les 4 autres

1 A. Girardie, Bull. Soc. zool. Fr. *91*, 423 (1966).
2 M. Cazal, A. Girardie et F. Bentz, Arch. Zool. exp. gén. *112*, 293 (1971).
3 T. C. Normann, Int. Rev. Cytol. *46*, 1 (1977).
4 N. Takeda, Appl. Ent. Zool. *11*, 143 (1976).
5 S. H. P. Maddrell et J. D. Gee, J. exp. Biol. *61*, 155 (1974).
6 A. Girardie, M. Moulins et J. Girardie, J. Insect Physiol. *20*, 2261 (1974).