

from <sup>12-15</sup>. The radioautograms were developed after 3 months of exposure, and stained with hematoxylin-eosin or methyl green-pyronine.

For each experimental condition and for each cell type, radioautographic granules over 40 cells (and/or nuclei) were counted. The radioactivity of the 'extracellular space' was estimated by counting 40 areas of a size comparable to an average eosinophil, marked by a circle in the ocular piece of the microscope, in areas chosen at random between cells located in the deep stroma.

**Results.** The uptake of tritiated estradiol by uterine eosinophils in vivo is confirmed in all the hormonal conditions studied in the present investigation (Tables I and II).

The uptake of tritiated estradiol by the nuclei of glandular, stromal and muscular cells is high in all the hormonal conditions investigated (Tables I and II). The uptake of radioactive estradiol by the nuclei of luminal epithelial cells is high in animals in proestrus and in estrus as well as in estrogen pretreated animals, but is very low in animals in the 1st and 2nd days of diestrus, as well as in animals pretreated with progesterone alone or together with estradiol (Tables I and II, Figure).

The nuclear radioactivity in sections of glandular epithelial cells constitutes 65-75% of the total cellular radioactivity (cytoplasmic + nuclear areas) in all the hormonal conditions studied (Tables I and II). Similarly, the nuclear radioactivity in sections of luminal epithelial cells of animals in proestrus and in estrus, as well as in those pretreated with estradiol alone, constitutes 65-75% of the total cellular radioactivity (Tables I and II). In contrast to this, in the animals in the 1st and 2nd days of diestrus, as well as those pretreated with progesterone alone or together with estradiol, the nuclear radioactivity of the luminal epithelial cells constitutes only 50% of the total cellular radioactivity (Tables I and II).

These figures show that progesterone drastically and selectively reduces the uptake of <sup>3</sup>H-estradiol by cells of the luminal epithelium. This reduction in binding is associated with obvious morphological changes in this cell type (a reduction in cell size), as observed in the Figure.

**Discussion.** The progesterone-induced specific inhibition of estradiol uptake by the nuclei of the luminal epithelial cells, but not by other uterine cells, parallels the inhibition by progesterone of some estrogenic responses in the luminal epithelial cells only. The estrogenic responses inhibited in the luminal epithelium are the mitotic activity<sup>16-20</sup>, the uridine uptake<sup>21</sup> and the increase in the size of these cells. Since the direct competition of progesterone and estradiol for the receptor sites is unlikely<sup>22,23</sup>, it is probable that progesterone has a specific inhibitory effect on either the synthesis of estrogen receptors or the transfer of the estrogen-cytosol receptor complex from the cytoplasm to the nucleus in the luminal epithelial cells.

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## Peripheral Sympathetic Innervation of the Deep Pineal Gland of the Golden Hamster<sup>1</sup>

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**Summary.** Both the superficial and deep pineal components of the intact hamster contain a rich network of green to yellow-green fluorescent nerve fibres. After either superior cervical ganglionectomy or after transection of the nervi conarii the majority of the fluorescing fibres disappeared from both the superficial and deep pineal masses. Although the deep pineal remained intact after surgical removal of the superficial pineal, it was devoid of any green or yellow-green fluorescent fibres.

While examining the pineal system of the hamster, SHERIDAN and REITER<sup>3</sup> observed that the pineal is divided into two parts. In addition, to the mass of pineal tissue (superficial pineal) adherent to the under surface of the confluence of sinuses, there is a second pineal mass associated with the habenular commissure (deep pineal). In physiological studies<sup>4,5</sup>, surgical removal of the pineal gland consists of removal of the superficial pineal only. The present study was designed to investigate the peripheral innervation of the deep pineal in intact hamsters and in hamsters that had their superficial pineal removed. For this study the fluorescent histochemical procedure of FALCK-HILLARP<sup>6</sup> for biogenic amine containing neurons was used.

**Materials and methods.** Groups of anesthetized male hamsters were subjected either to a sham operation, to bilateral superior cervical ganglionectomy<sup>7</sup>, to nervi

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conarii transection (cutting the post-ganglionic sympathetic fibres immediately prior to their entrance into the superficial pineal)<sup>8</sup>, or to superficial pinealectomy<sup>9</sup>. 4 to 8 weeks later, hamsters were decapitated and their brains were quickly removed. The epithalamic region was frozen in isopentane cooled by liquid nitrogen. The tissues were prepared for fluorescent histochemistry according to the technique of FALCK-HILLARP<sup>6</sup>. 8 to 10  $\mu$ m thick paraffin sections were examined with a Zeiss dark-field microscope. High Speed Ektachrome and Tri-X films were generally exposed for 1 to 3 min.

**Results.** The terms, superficial and deep pineal, refer to the 2 pineal components as described by SHERIDAN and REITER<sup>3</sup>. In all 12 intact (sham operated) hamsters, the superficial pineal contained an extensive network of green to yellow-green fluorescent fibres which ranged in size from thin, individual fibres to thick fibrous bundles. Many of these fibres appeared to be associated with connective tissue trabeculae and were judged to be pervascular nerves. In some favorable sections large green fluorescent fibre bundles were observed adjacent to the lateral aspect of the superficial pineal and were considered to be fibres of the nervi conarii. The deep pineal of the intact hamsters had an extensive network of intensely green to yellow-green fluorescent fibres. In 2 hamsters, the plexus was seen to be continuous with a large fibre bundle located in the stalk.

After bilateral superior cervical ganglionectomy, the number of green to yellow-green fluorescent fibres in both the superficial and deep pineal was greatly reduced in 75% (9 of 12) of the hamsters, while in 25% of the animals there was little discernible difference in fibre density with respect to sham operated controls. After bilateral transection of the nervi conarii, no intrapineal fluorescent fibres were observed in either pineal component in 44% (4 of 9) of the hamsters. In the remaining 56%, the superficial pineals contained very few fluorescent fibres while the deep pineal was completely devoid. After superficial pinealectomy, no green fluorescent fibres were observed in the deep pineal.

**Discussion.** Under appropriate conditions, the green to yellow-green fluorescent product, as seen in the present study, is due to the presence of norepinephrine (NE). NE has been shown to be the sympathetic neurotransmitter in the pineal<sup>10,11</sup>. In the present study, both the

superficial and deep pineal components of intact hamsters were found to contain numerous green fluorescent fibres. In several animals many of these fibres were also seen in the stalk which connects the superficial and deep pineal.

The primary autonomic innervation of the mammalian pineal is believed to arise from the superior cervical ganglia<sup>12</sup>. In the rat, the green fluorescent fibres within the pineal gland disappear after superior cervical ganglionectomy<sup>13,14</sup>. After bilateral removal of the superior cervical ganglia or after bilateral transection of the nervi conarii in the hamsters, both the superficial and deep pineal lost most of their green to yellow-green fluorescence. Ganglionectomy also incapacitates the hamster pineal in terms of its antigonadotrophic capabilities<sup>5,7</sup>, probably due to the fact that pineal complex cannot function without an intact sympathetic innervation.

The present study shows that the deep pineal mass remains intact after superficial pinealectomy. However, the green fluorescent product was almost completely lost after removal of the superficial pineal. Thus, as with the superficial pineal, the deep pineal also seems to be non-functional with respect to its inhibitory influence on reproduction if it is denervated, since in light-deprived hamsters with only superficial pinealectomy the deep pineal is incapable of suppressing gonadal function<sup>5</sup>. On the other hand, it is possible that the function of the deep pineal is entirely different from that of the superficial gland as suggested by WIKLUND<sup>15</sup>. The results suggest that the postganglionic sympathetic fibres which terminate in the deep pineal either pass in the vicinity or through the superficial pineal and are interrupted at the time of superficial pinealectomy.

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## Effect of Chemical Sympathectomy, Adrenalectomy and Adrenergic $\alpha$ - and $\beta$ -Blocking Agents on the Development of Hyperglycemia Induced by Streptozotocin in the Rat

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**Summary.** The sympathetic nervous system in the rat does not play any significant role in the streptozotocin-induced and tolbutamide-induced hypoglycemia.

Recent investigations have shown that the effect of a number of pharmacological agents on insulin secretion may be mediated by  $\alpha$ - and  $\beta$ -adrenergic pancreatic islet cell receptors;  $\alpha$ -receptor stimulation by adrenaline inhibited glucose induced insulin release in the rat<sup>1,2</sup>, monkey<sup>3</sup> and man<sup>4-6</sup>. However,  $\beta$ -receptor stimulation by isoprenaline stimulated insulin secretion in man<sup>7</sup>, dog<sup>8</sup> and rat<sup>9</sup>.  $\alpha$ -Adrenergic blocking agents, such as phentolamine, increased insulin secretion in man<sup>10</sup>, baboon<sup>11,12</sup> and dog<sup>13</sup>, whereas  $\beta$ -adrenergic blocking

agents, such as propranolol and MJ-1999, inhibited insulin release in mice<sup>14</sup>, dog<sup>8</sup> and man<sup>15</sup>. Both in vitro and in vivo studies showed that glucose-induced insulin release could be blocked in the rat sympathectomized by 6-hydroxydopamine<sup>16</sup>.

The present study was designed to see the role of the sympathetic nervous system in the development of hyperglycemia following i.v. administration of streptozotocin. Chemical sympathectomy was produced by 6-hydroxydopamine (6-OHD). This substance has been