

were seen to leave the cell clusters in bundles together with non-fluorescent fibres. Both types of perikarya were enclosed by fluorescent terminal nerve fibres in a manner suggesting a synaptic arrangement.

Flask-shaped cells resembling enterochromaffin cells were distributed in quite large number among the epithelial cells of the mucosa (Figure 3a). They emitted an intense yellow cytoplasmic fluorescence, probably derived from 5-hydroxytryptamine, which occurred in

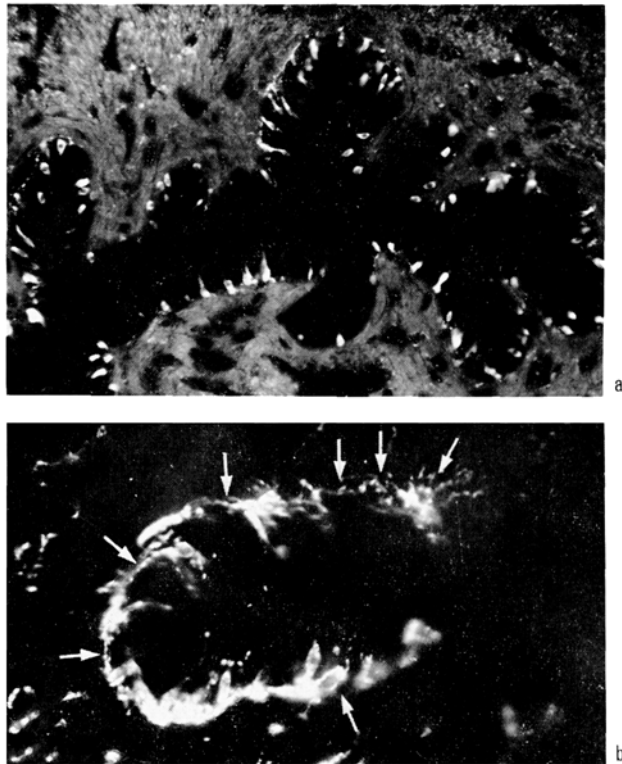


Fig. 3. Mucosa of urethra from cat subjected to hypogastric denervation. a) Large number of yellow-fluorescent, flask-shaped cells in the epithelium. $\times 100$. b) At higher magnification, an abundance of adrenergic nerve terminals (arrows) are seen to run in close contact with the base of the yellow enterochromaffin cells. $\times 240$.

0.37 and 0.48 $\mu\text{g/g}$ (2 determinations) in the urethra. A conspicuous accumulation of delicate adrenergic nerve terminals occurred in the immediate vicinity of, and even contiguous to, the base of the cells (Figure 3b). It is not unlikely that the arrangement represents a direct adrenergic innervation of the cells.

Hypogastric denervation produced a slight but clear decrease in the number of adrenergic nerves running in the smooth muscle wall (Figure 1b). This was consistent with the fluorometric determinations which revealed a significant reduction (Student's *t*-test: $0.02 > P > 0.01$) of urethral noradrenaline to $2.15 \pm 0.44 \mu\text{g/g}$.

No change was found in the number or fluorescence intensity of the urethral adrenergic nerves after removal of the lumbosacral portion of the sympathetic chain.

Conclusions. Fluorescence microscopy has shown the presence of adrenergic ganglion formations in the wall of the proximal urethra of the female cat. These ganglia contribute to the major portion of the urethral adrenergic innervation by way of short neurons. Fluorometric determinations of noradrenaline in combination with denervation experiments indicate that about $1/3$ of the post-ganglionic adrenergic innervation to the urethra derives from the inferior mesenteric ganglia via the hypogastric nerves. The sacral sympathetic ganglia (pelvic nerve) do not seem to contribute significantly to the sympathetic innervation¹³.

Zusammenfassung. Die adrenergische Innervation der weiblichen Urethra bei der Katze wird mit Hilfe fluoreszenzmikroskopischer Technik dargestellt. Adrenergische Ganglien in der proximalen Urethra bestreiten den Hauptanteil der urethralen adrenergischen Innervation (kurze Neurone). Ein Drittel der adrenergischen Bahnen stammen aus dem Nervus hypogastricus.

CH. OWMAN, T. OWMAN
and N.-O. SJÖBERG

*Institute of Anatomy and Histology, and
Department of Obstetrics and Gynecology at the
General Hospital of Malmö, University of Lund,
Lund (Sweden), 31 March 1970.*

¹³ Supported by Ford Foundation (grant No. 68-383).

Proliferation of Spleen Cells from Mice Infected with Friend Virus in the Spleens of Unirradiated and Irradiated Mice

Infection of mice with Friend virus (FV) results in splenomegaly, which is due to proliferation of reticulum cells in the spleen¹. Although injection of a spleen homogenate from FV-infected mice into normal hosts can cause proliferation of host spleen cells, it is not known whether FV-infected cells injected i.v. into normal mice can proliferate in the hosts. Chromosome marker, CBA/HT₆T₆, was used to identify the origin of cells^{2,3}.

CBA/HT₆T₆ mice were infected with a homogenate of FV-infected spleens. 7 days later, the spleens of the infected CBA/HT₆T₆ mice were teased, and samples of spleen-cell suspension, each containing 1×10^7

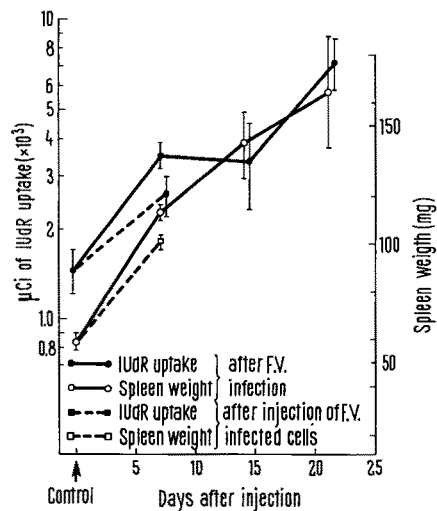
cells, were injected i.v. into normal CBA/H mice, unirradiated or irradiated with 400 R. T₆T₆ cells were scored in the spleens of the CBA/H mice 7 and 14 days after injection of the cell suspension, as described previously⁴. DNA synthesis was measured by injecting ¹²⁵I-UdR into the mice, and counting ¹²⁵I-UdR uptake in the spleen 18 h later⁵.

The Figure shows ¹²⁵I-UdR incorporation and spleen weight in CBA/HT₆T₆ mice before and after infection with FV. For a normal CBA/HT₆T₆ mouse, IUdR uptake was $1.47 \pm 0.24 (\times 10^{-3}) \mu\text{Ci}$ per spleen, and the weight of the spleen was $56.5 \pm 4.2 \text{ mg}$. For an FV-infected

Cells of host and donor types in samples taken from host spleens after injection of 1×10^7 CBA/HT₆T₆ spleen cells infected with Friend virus into unirradiated or irradiated CBA/H mice^a

Radiation dose (r) ^c	Series	Types of cells in host spleens on day of sacrifice ^b					
		Day 7			Day 14		
		Host	Donor	(%)	Host	Donor	(%)
0	1	51	0	(0)	51	0	(0)
	2	50	0	(0)	50	1	(2.0)
	3	50	0	(0)	50	0	(0)
400	1	40	34	(45.9)	18	50	(73.5)
	2	56	62	(52.5)	1	41	(97.6)
	3	15	50	(75.8)	7	50	(87.7)

^a Each CBA/HT₆T₆ mouse was given an i.p. injection of 0.2 ml of a stock homogenate of FV-infected spleens; 7 days later, the spleen of each CBA/HT₆T₆ mouse was removed and shredded, and a suspension of 1×10^7 spleen cells was injected i.v. into each CBA/H mouse. ^b CBA/HT₆T₆ cells were injected on day 0. ^c Irradiation was performed on day 0.



IUDR uptake and spleen weight in mice infected with Friend virus. Each mouse was injected with 0.2 μ Ci of 5-iodo-2'-deoxyuridine-¹²⁵I (¹²⁵I-UdR) per g of body weight 18 h before sacrifice, and was given 10^{-7} mole of 5-fluoro-2'-deoxyuridine (FUDR) 1 h before injection of ¹²⁵I-UdR. Key: solid circles, IUDR uptake per spleen after FV infection; open circles, spleen weight after FV infection; solid squares, IUDR uptake after injection of 1×10^7 FV-infected spleen cells; open squares, spleen weight after injection of 1×10^7 FV-infected spleen cells. Day 0 was the day of infection, and the values for that day in the graph are those for a 7-week \pm 2-day-old control animal. Each point in the graph represents a mean \pm the standard error.

mouse on day 7, IUDR uptake was 3.5 ± 0.35 ($\times 10^{-3}$) μ Ci, and spleen weight was 112.4 ± 7.6 mg. IUDR uptake per unit spleen weight was 0.026 (1.47/56.5) in the normal spleens, and 0.037 (3.5/94.7) on day 7 in the FV-infected spleens.

The ratio of DNA synthesis in the spleens of the infected mice on day 7 to that in the normal spleens was 1.42, indicating that an FV-infected spleen cell had an average of 42% more dividing capacity than did a normal spleen cell. Figure 1 also shows that the spleen

of a CBA/H mouse that received 1×10^7 FV-infected spleen cells also increased in weight and IUDR incorporation.

The chromosome analysis summarized in the Table indicates that T₆T₆ spleen cells from FV-infected mice were not found in the unirradiated spleens of CBA/H mice on days 7 and 14, whereas most of the dividing cells on days 7 and 14 were of donor origin in the spleens of CBA/H mice irradiated with 400 R.

It may be concluded that the increased DNA synthesis and cell division in the spleens of CBA/H mice that received 1×10^7 FV-infected CBA/HT₆T₆ spleen cells was due to proliferation of host spleen cells, and not donor spleen cells, although DNA synthesis was higher in FV-infected spleen cells than in normal spleen cells. The proliferation of host spleen cells may be induced by virus released from the injected spleen cells.

Résumé. Quand des cellules spléniques de souris CBA/HT₆T₆ infectées par le virus de Friend sont injectées à des souris CBA/H normales, les rates des receveurs s'hypertrophient au cours de la semaine suivante. Une analyse chromosomale montra que toutes les cellules en division provenaient de l'hôte, suggérant que la splénomégalie fait suite à la libération du virus des cellules injectées.

A. TAKADA, Y. TAKADA
and J. L. AMBRUS

Springville Laboratories, Roswell Park Memorial Institute,
Springville (N.Y. 14141, USA), 10 August 1970.

1 R. SIEGLER, in *Experimental Leukemia* (Ed. M. A. RICH; Appleton-Century-Crofts, New York 1968).
2 C. E. FORD, J. L. HAMERTON, D. W. H. BARNES and J. F. LOUITT, *Nature, Lond.* 177, 452 (1956).
3 H. A. MICKLEM, C. E. FORD, E. P. EVANS and J. GRAY, *Proc. R. Soc., B* 165, 78 (1966).
4 A. TAKADA, Y. TAKADA, C. C. HUANG and J. L. AMBRUS, *J. exp. Med.* 129, 445 (1969).
5 G. CUDKOWICZ, A. C. UPTON, L. H. SMITH, D. G. GOSSLEE and W. C. HUGHES, *Ann. N.Y. Acad. Sci.* 114, 571 (1964).