

A Histochemical Study on the Innervations of the Uterine Muscles of the Mouse, Rat and Rabbit

Mitsuru OKI,* Hiroaki YOSHIMURA* and
Hitoshi KONDO**

From the Department of Anatomy and
Physiology,***

*Yamaguchi University School of Medicine
(Received August 28, 1969)*

Recent investigations with histochemical techniques for the selective demonstration of adrenergic (fluorescence-containing) and cholinergic (acetylcholinesterase-containing) nerves have demonstrated that the uterine smooth muscle tissues are supplied with numerous adrenergic nerves in a variety of species¹⁻³⁾ However, cholinergic innervation pattern is still obscure in details. Pallie et al⁴⁾ have shown that the large intrinsic nerves of the uterus are made up of plexiform strands, though there exist no ganglion cells in this plexus. On the other hand, Clegg⁵⁾ showed that the uterine ganglia exist in the mesometrium of the guinea-pig uterus. Furthermore, physiological works by Bower⁶⁻⁷⁾ have elucidated that stimulation of the pelvic nerves had no effect, though stimulation of the hypogastric nerve evoked a compound action potential in the uterine nerves. In the present work, therefore, histochemical staining of acetylcholinesterase (AChE)⁸⁻⁹⁾ was employed to investigate the cholinergic innervation of the uterus of mouse, rat and rabbit. (The majority of the experiments was made on mature mouse weighing 20~22 g, which were pretreated with intramuscular injection of ova-hormon depot (Estradiol dipropionate 1, 3, 5 (10)-estratriene-3, 17 β -diol dipropionate) or of oophormin luteum depot (17 α hydroxy-progesterone-17-n-capronate) in 0.1 ml of sesame oil (Teikoku Zōki, Co. Ltd).

Fig. 1 shows the most typical patterns of spontaneous contraction of the isolated uterine muscle strips in estrogen-dominated stage and the administration of eserine (10^{-6} M) to the bath caused the potentiation of the tonic components, which followed an increase of the frequency of the phasic components (Fig. 1-B), although another preparation failed to respond to eserine at the same dosis (Fig. 1-A).

In the eseritized mouse uterine strips, which are in estrogen dominated stage, subsequent addition of adrenaline caused an abolition or reduction of the spontaneous contraction as is shown in Fig. 1-B.

It has been generally accepted that eserine possesses the characteristic pharmacological effects of the anti-AChE agents. Thus, present results may be due to the inhibition or inactivation of AChE at sites of cholinergic transmission, with the consequent accumulation and actions of endogenous Ach liberated both by cholinergic nerve impulse and, in much smaller amounts, by continual leakage during the

resting stage.⁸⁾ In the present study the patterns of staining obtained with acetylthiocholine and butyrylthiocholine substrates were completely abolished by eserine as shown in Fig. 4.

It may thus be assumed that the nerves staining for AchE are cholinergic in nature, supporting the previous histochemical observations⁹⁾¹¹⁾.

Fig. 2 (A-B) shows that the nerve strands or fibers derived from the plexus uterovaginalis, making the individual compartment of muscle tissues, enclose the outer longitudinal smooth muscle layer and spread out in the nerve plexus in the connective tissue layers which exist between two muscle layer of the mice uterus (using modified Bielschowsky silver stain.)

On the other hand, moderate number of AchE-containing fibers (cholinergic fibers) are observed in the both muscle layer, though fewer fibers are present in the circular musculature, even if longitudinal sections were observed (Fig. 3). In the comparison of these observations (as in Fig. 2 and Fig. 3), thus, cholinergic nerve innervation appeared to be very weak on the uterine smooth muscles.

In addition, AchE positive (cholinergic) ganglion cells were observed prominently in rabbit uterine musculature, while no such a ganglion cells were detectable in mouse uterine musculature (Fig. 5). Fig. 6 shows the medullated thick sensory fibers of the intrinsic nerve trunk of the mice uterus. Postganglionic neuron and the well developed nerve plexus was observed in the myometrium of rat (Fig. 7 & 8). The present observations indicate that the manner of cholinergic innervation of the myometrium differs in the animal species and that the excitatory fibres are cholinergic in nature, differing from the previous histological and pharmacological evidence⁶⁻⁷⁾.

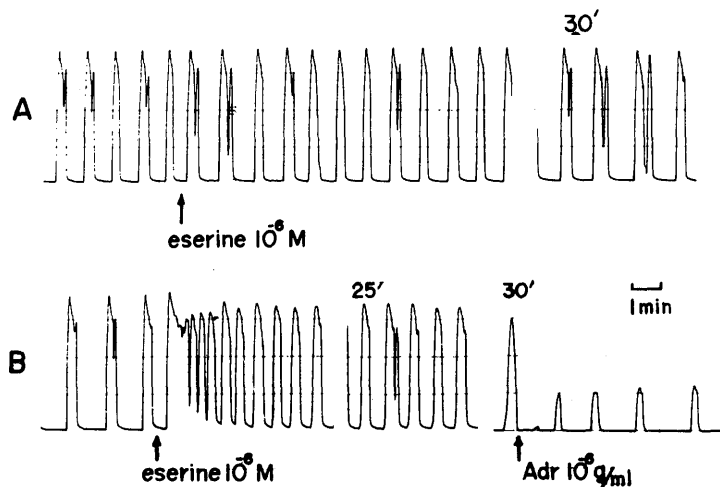
In addition, nonspecific cholinesterase activity (ChE) in a whole uterine musculature becomes fewer after the injection of estrogen. (discussion in the next paper⁹⁾.)

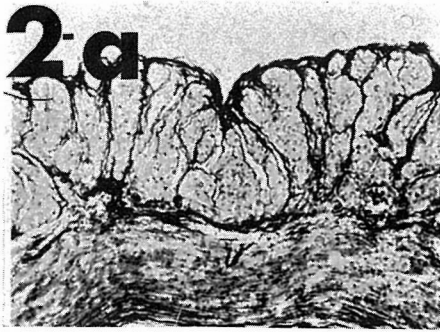
REFERENCES

- 1) Owman, Ch., Rosengren, E. and Sjöberg, N.-O.: Origin of the adrenergic innervation to the female genital tract of the rabbit. *Life Sci.*, **5**: 1389-1396, 1966.
- 2) Owman, Ch. and Sjöberg, N.-O.: Adrenergic nerves in the female genital tract of the rabbit. With remarks on cholinesterase-containing structures. *Z. Zellforsch.*, **74**: 182-197, 1966.
- 3) Rosengren, E. and Sjöberg, N.-O.: The adrenergic nerve supply to the female reproductive tract of the Cat. *Am. J. Anat.*, **121**: 271-284, 1967.
- 4) Pallie, W., Corner, G. W. & Weddell, G.: Nerve terminations in the myometrium of the rabbit. *Anat. Rec.*, **118**: 789-812, 1954.
- 5) Clegg, P. Catherine: The effect of adrenergic blocking agents on the guinea-pig uterus in vitro, and a study of the histology of the intrinsic myometrial nerves. *J. Physiol.*, **169**: 73-90, 1963.
- 6) Bower, E. A.: The characteristics of spontaneous and evoked action potentials recorded from the rabbits uterine nerves. *J. Physiol.*, **183**: 730-747, 1966.

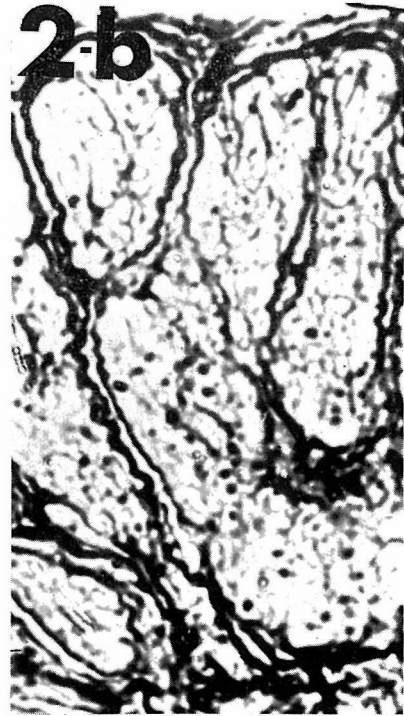
- 7) Bower, E. A.: The activity of post-ganglionic sympathetic nerves to the uterus of the rabbit. *J. Physiol.*, **183**: 748-767, 1966.
- 8) Karnovsky, M. J. & Roots, L.: A "direct-coloring" thiocholine method for cholinesterases. *J. Histochem. Cytochem.*, **12**: 219-221, 1964.
- 9) Oki, M. and Yoshimura, H.: Localization of pseudocholinesterase in uterine muscles of mouse, rat and rabbit. (in press: manuscript available in histochemical observation on the neuromuscular junction of the uterus in the mouse.) *Acta Anatomica Nipponica*. **42**:6, 1967.
- 10) Goodman, L. S. and Gilman, A.: *The Pharmacological Basis of Therapeutics*. 3rd Edition. 1965. The Macmillan Company, New York.
- 11) Bell, C.: A histochemical study of the esterases in the bladder of the toad (*Bufo Marinus*). *Comp. Biochem. Physiol.*, **21**: 91-98, 1967.)

1 Effect of eserine on the isolated uterine muscle

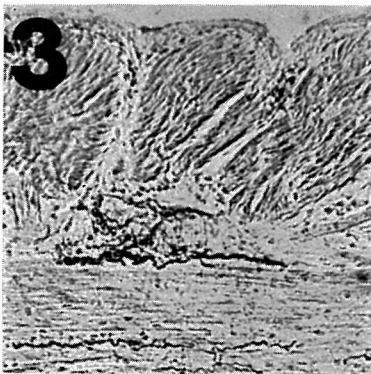




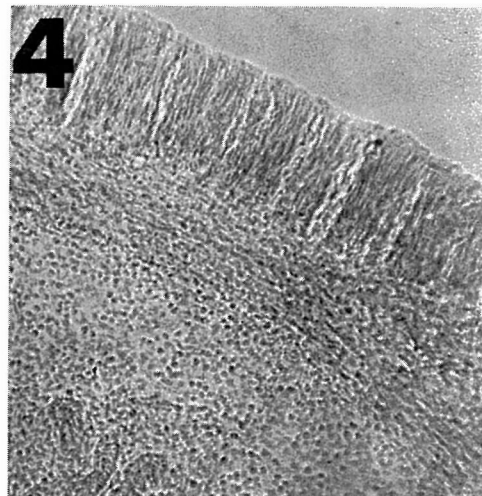
(撮影倍率) × 50



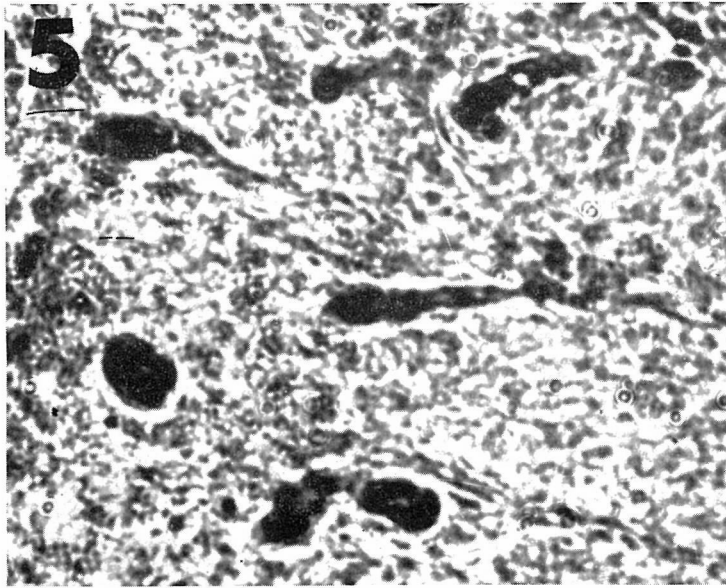
× 400



× 100



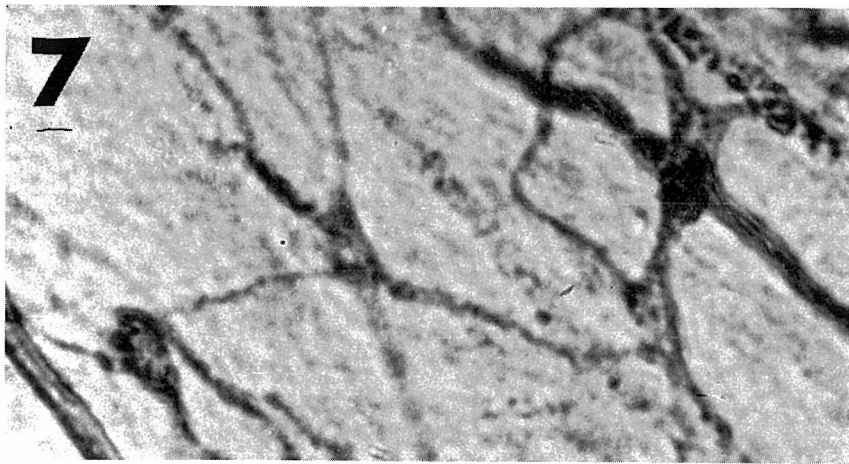
× 100



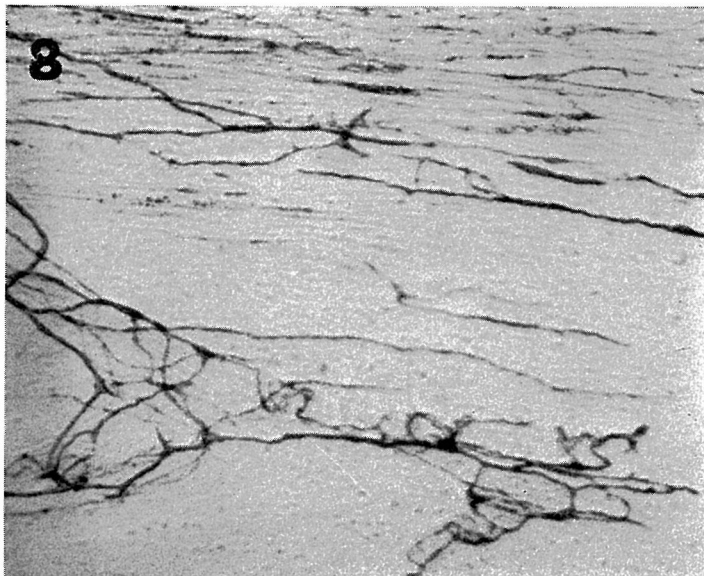
× 400



× 400



× 400



× 150