The distribution of cholinergic and other nerve fibres in the human uterus

R. E. COUPLAND
M.D., Ph.D., F.R.S.E.

Department of Human Morphology, University of Nottingham

NERVE fibres have been identified in the non-pregnant human uterus of subjects aged 24–45 years by means of the cholinesterase histochemical reaction and impregnation with osmium iodide. Both methods give readily reproducible results and both myelinated and non-myelinated fibres may be identified, the latter being the most striking.

Material processed for the cholinesterase reaction was fixed in buffered 4% formaldehyde and processed according to the method described by Coupland & Holmes (1957) and Coupland (1962), using acetyl or butyryl thiocholine as substrates and diisopropyl fluorophosphate and eserine as inhibitors. With this technique cholinergic nerve fibres show a strong reaction for true cholinesterase (AChE) which is localized to the axons and, in the region of the peripheral autonomic ground plexus, any beads associated with these, while supporting elements only react for pseudocholinesterase (ChE).

Impregnation with osmium iodide (Champy technique) according to the method of Sutherland (1963) reveals a fine network of nerve fibres, many of which show fine beaded axons typical of the peripheral autonomic nervous system. The functional type of nerve fibres revealed by this method is currently unknown but from the present work it is clear that the pattern is not the same as that revealed by the cholinesterase technique. In the light of knowledge of the functional types of nerve fibres reaching the uterus from somatic and sympathetic nerve plexuses, it seems likely that some may represent adrenergic fibres and others may be sensory elements.

Cholinergic nerve fibres, as revealed by the histochemical technique, are most numerous in the region of the cervix and very few are seen above the level of the internal os. Myelinated and non-myelinated fibres are seen running along with bundles of smooth muscle in the cervix, traversing the stroma. Fine beaded AChE-positive fibres form rich networks around cervical glands and in the adventitia of cervical arteries; similar fibres form plexuses in the connective tissues adjacent to the

stratified squamous epithelium of the vaginal aspect of the cervix and penetrate into the connective tissue papillae of the mucous membrane. The highest concentration of fibres associated with muscle bundles occurs in the upper part of the cervix and extends up to the junction of body and cervix. Hence this includes the isthmus and possibly the zone which gives rise to the lower uterine segment.

In the body of the uterus AChE-positive fibres are only very occasionally seen although the supporting cells of other nervous elements may react for ChE. When present, isolated AChE-positive fibres lie adjacent to muscle bundles or blood vessels, but never form the dense networks or plexuses seen in the cervix. No nerve fibres have been seen in the functional layer of the endometrium and only very occasional fibres were seen in the basal region running between glands and underlying myometrium.

The osmium iodide technique has revealed a rich plexus composed of fine fibres in association with the smooth muscle of the uterine body, isthmus and cervix (Fig. 1). These nerve fibres usually run parallel to muscle bundles in all these areas, are usually beaded and may be relatively straight or take a sinuous course; some appear to form spirals. Some fine nerve fibres are seen traversing the connective tissue stroma of both body and cervix. In the body of the uterus no evidence of innervation of endometrial glands of the uterus has been obtained although occasional fibres have been seen in the basal layer running in the region between glands and adjacent myometrium. Fine beaded nerve fibres are usually seen lying adjacent to cervical glands but are less numerous than those revealed by the cholinesterase technique. Adventitial plexuses of nerve fibres are seen in association with uterine arteries of both the body and cervix after osmium iodide impregnation. This is in contrast to the results obtained with the cholinesterase technique since the latter shows few or no fibres in the arteries of the body. No nerve fibres have been observed in the vicinity of spiral arteries of the endometrium with either technique.

FIG. 1. Fine nerve fibres revealed by the osmium iodide technique associated with smooth muscle of body of uterus. $\times 350$.

FIG. 2. Distribution of nerve fibres in non-pregnant human uterus as revealed by: (a) cholinesterase technique (cholinergic) and (b) osmium iodide techniques. Density of fibres indicated by degree of cross-hatching.

Comparison of the patterns of nerve fibres revealed by the osmium iodide and cholinesterase techniques suggests that many of the fibres associated with the arteries and glands of the cervix and with myometrium of cervix and isthmus are cholinergic in type. The AChE-non-reactive fibres revealed in other areas by osmium iodide impregnation are likely to be of functionally different types. Since the majority

of the non-cholinergic elements are finely beaded fibres typical of autonomic ground plexuses it is suggested that these are likely to be adrenergic elements. Some may, however, represent sensory nerve fibres and endings since unencapsulated fine spirals are occasionally observed adjacent to muscle fibres. Corpuscles giving a strong reaction for ChE are occasionally observed in the cervical connective tissue and may represent the same elements as the corpuscles described by Keiffer (1936). The pattern of distribution of fibres revealed by the two techniques is illustrated in Fig. 2.

References

- COUPLAND, R.E. (1962) Histochemical observations on the distribution of cholinesterase in the human uterus. *J. Obstet. Gynaec. Brit. Emp.* **69**, 1041.
- COUPLAND, R.E. & HOLMES, R.L. (1957) The use of cholinesterase techniques for the demonstration of peripheral nervous structures. *Quart. J. microsc. Sci.* **98**, 327.
- KEIFFER, H. (1936) De l'existence dans le corps de l'utérus humain, appareil nerveux plus ou moins systématisé, certains d'entre eux semblant jouer le rôle de récepteur sensoriel pour les muscles. *Bull. Acad. roy. Med. belg.* **1**, 508.
- SUTHERLAND, S.D. (1963) The use of zinc iodide in the Champy osmic acid technique. *J. Anat. (Lond.)*, **97**, 624.