

with recent bioassay measurements^{12,13}. The slightly higher values obtained in our experiments could be explained by a more efficient extraction procedure⁹. As seen in Table III, chronic exercise induces an increase in the total acetylcholine content of the heart. Also, the heart tissue acetylcholine concentration as expressed by $\mu\text{g/g}$ is significantly increased in the exercised animals. Very probably, and as shown by HERRLICH et al.³ the increase of the acetylcholine concentration of the auricles is mainly responsible for these results. In addition, EKSTRÖM¹⁴ reported recently that in chronically exercised animals, the choline acetyl transferase activity of the auricles was significantly increased. Although the existence of acetylcholine bound to the myocardium and independent from nerve terminals has been reported¹⁵, it is tempting to suggest that the acetylcholine increase in the heart reflects an increase of the parasympathetic activity on the heart. Whether the activity of the heart in chronically exercised animals is mainly influenced by the sympathetic or parasympathetic system is still a controversial matter³⁻⁶.

Résumé. Le taux de l'acetylcholine du myocarde a été dosé par chromatographie en phase gazeuse chez le rat. Chez l'animal exercé le taux de l'acetylcholine était plus élevé que chez son homologue sédentaire.

C. DE SCHRYVER and J. MERTENS-STRYTHAGEN¹⁶

*Department of Physiology, Faculty of Medicine,
Facultés Universitaires Notre Dame de la Paix,
Rue de Bruxelles 61, B-5000 Namur (Belgium),
11 September 1974.*

¹² J. VLK, S. TUCEK and V. HABERMANN, *Nature, Lond.* 189, 923 (1961).

¹³ V. BHARGAVA, *Nature, Lond.* 215, 202 (1967).

¹⁴ J. EKSTRÖM, *Q. Jl. exp. Physiol.* 59, 73 (1974).

¹⁵ E. CORABOEUF, G. LE DOUARIN and G. OBRECHT-COUTRIS, *J. Physiol., Paris* 206, 383 (1970).

¹⁶ The authors acknowledge the excellent technical assistance of Mr. F. VIGNERON.

Neurotubules: Different Densities in Peripheral Motor and Sensory Nerve Fibres^{1,2}

According to earlier findings in mouse sciatic nerve³, which are substantially supported by other authors^{4,5}, a general rule of inverse relationship between fibre size and density of neurotubules is well established. In a previous paper⁶, evidence was presented that – while the above-mentioned rule is fully valid – the density of neurotubules is significantly higher in ventral (motor) root fibres than in dorsal (sensory) root fibres of comparable diameter. The present investigation is concerned with the question of whether these differences in tubular density are also present in motor and sensory nerve fibres distal to the spinal ganglia and particularly in more distant parts of the peripheral nervous system.

Methods. 3 albino rats (Sprague-Dawley/320–440 g) were fixed by perfusion with cacodylate-buffered 7% glutaraldehyde under nembutal anaesthesia. In 2 animals the dorsal roots of L₅ were excised in connection with the spinal ganglia, each with a short stump of the spinal nerve. In a third animal the saphenous nerve and the nerve to the medial head of the gastrocnemius muscle (MGN) were taken out as examples of a typical cutaneous and a typical muscular nerve. Postfixation in OSO₄; embedding in epon; double staining of ultrathin sections with uranyl acetate and lead citrate; Zeiss EM 9S electron microscope. Micrographs of complete nerve fibre cross sections were made at standard magnification of $\times 5000$; for morphometric measurements prints at $\times 15,000$ were used. Samples of 22–50 large as well as small diameter nerve fibres of each specimen were evaluated. The following parameters have been determined and subjected to statistical analysis: 1. area of cross section (ACS) in μm^2 (planimetry of the micrographs), and 2. total count of neurotubules per ACS and calculation of their density (number per μm^2 of ACS). For statistical analysis the Student *t*-test was applied.

Results and discussion. Nerve fibres of the saphenous nerve, a cutaneous nerve, were compared with those of the MGN, a mixed muscle nerve containing more than 50% of motor nerve fibres. The density of neurotubules was significantly higher in the axons of saphenous nerve fibres as compared with those of the MGN. These differ-

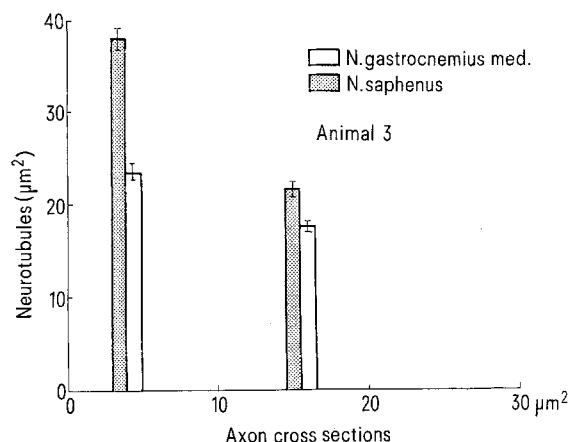


Fig. 1. Comparison of the densities of axonal neurotubules in samples of small and large nerve fibres of a cutaneous nerve (black bars) and a muscle nerve (white bars) of the rat. Ordinate: number of neurotubules per square micron of axon cross section (NT/ μm^2); the bars give the mean and the standard deviation of the mean. Abscissa: position of the bars according to the mean cross sectional area of the axons of each sample; axon cross sections in square microns (ACS/ μm^2) were determined by planimetry.

¹ This investigation was supported by the 'Fonds zur Förderung der wissenschaftlichen Forschung in Österreich'.

² The authors gratefully acknowledge the excellent technical assistance of Miss E. HOHBERG and Mrs. B. STÖGERMAYER.

³ R. L. FRIEDE and T. SAMORAJSKY, *Anat. Rec.* 167, 379 (1970).

⁴ R. MAYR, *Anat. Anz.* 130, 391 (1972).

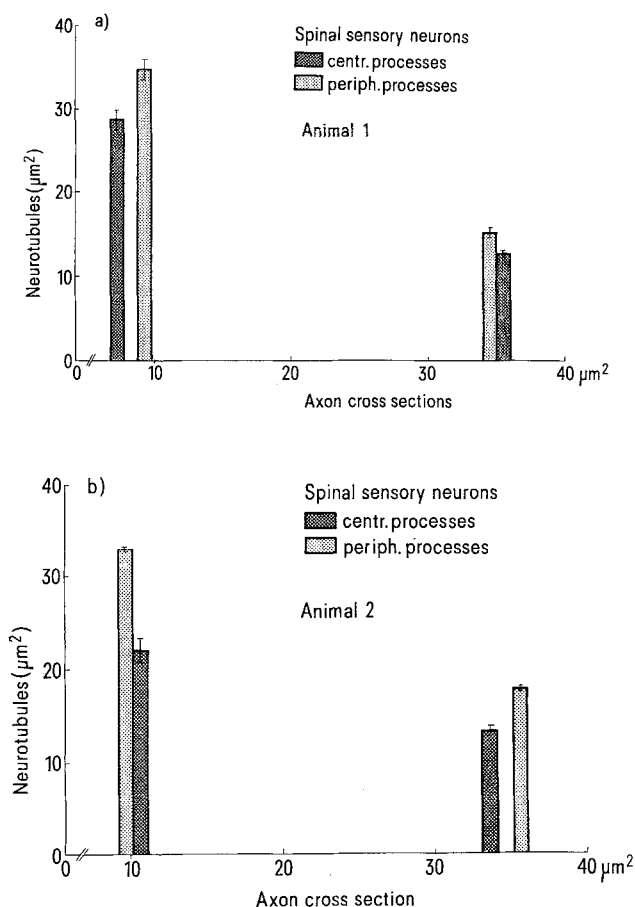
⁵ K. H. ANDRES and M. VON DÜRING, in *Handbook of Sensory Physiology*, (Springer-Verlag, Berlin-Heidelberg-New York 1973), vol. 2.

⁶ W. ZENKER, R. MAYR and H. GRUBER, *Experientia* 29, 77 (1973).

Table I. Density of neurotubules in a cutaneous (N. saphenus) and a muscular (N. gastrocnemius med.) nerve branch (animal 3)

	Small nerve fibers (2.1–3.3 μm diameter)			Large nerve fibers (5.4–6.7 μm diameter)		
	N. saph.	N. gastrocn. med.	Significance	N. saph.	N. gastrocn. med.	Significance
<i>n</i>	50	50		35	35	
Cross sectional area of axon in μm^2 (ACS)	3.66 ± 0.17	4.16 ± 0.19	ns	15.07 ± 0.41	15.54 ± 0.55	ns
Neurotubules (number/ μm^2 of ACS)	38.22 ± 1.37	23.57 ± 0.96	+++	21.84 ± 0.87	17.73 ± 0.63	+++

The values give the mean and the standard deviation of the mean. The values of comparable caliber classes of central and peripheral processes of spinal sensory neurons were statistically examined by the Student *t*-test. ns, not significant; +, $p < 0.05$; ++, $p < 0.01$; +++, $p < 0.001$.



ences were observed in large diameter nerve fibres as well as in small ones (Table I, Figure 1). Within both nerves, according to the general rule, the small fibres exhibited higher tubular densities than the large ones.

How can these results of higher tubular density in a cutaneous nerve than in a muscle nerve be reconciled with our previous findings of higher tubular density in the motor ventral root fibres as compared to the sensory dorsal ones⁶? To answer this question we examined whether there is a difference in tubular density between sensory fibres proximal and immediately distal to the spinal ganglion. In fact, the tubular densities in nerve fibres on both sides of the spinal ganglion proved to be unequal. We found the density of neurotubules to be significantly higher in the peripheral processes of spinal ganglion cells than in the central ones of comparable diameter at the level of the dorsal roots (Table II, Figure 2). This fact probably accounts for the inverse relationship of the tubular densities in sensory and motor nerve fibres between spinal roots and peripheral nerves.

In a recent study on amphibian spinal nerve fibres⁷, a similar difference of tubular density between sensory nerve fibres proximal and distal to the dorsal root ganglion was found. In agreement with our findings in the rat⁶,

⁷ R. S. SMITH, Can. J. Physiol. Pharmac. 51, 798 (1973).

←

Fig. 2. Comparison of the densities of axonal neurotubules in samples of small and large sensory nerve fibres immediately distal (black bars) and proximal (dotted bars) to the spinal ganglion L₅ of the rat. a) Animal 1; b) Animal 2. Ordinate and abscissa as in Figure 1.

Table II. Density of neurotubules in central and peripheral processes of spinal sensory neurons

	Small spinal sensory fibers (3.0–4.5 μm diameter)			Large spinal sensory fibers (8.5–10.3 μm diameter)		
	Central processes	Peripheral processes	Significance	Central processes	Peripheral processes	Significance
Animal 1 (<i>n</i>)	30	30		22	22	
Cross sectional area of axon in μm^2 (ACS)	7.24 ± 0.34	9.22 ± 0.46	++	35.39 ± 1.07	34.35 ± 1.06	ns
Neurotubules (number/ μm^2 of ACS)	28.71 ± 1.23	34.83 ± 1.26	++	12.71 ± 0.43	15.16 ± 0.60	++
Animal 2 (<i>n</i>)	29	30		31	29	
Cross sectional area of axon in μm^2 (ACS)	10.62 ± 0.45	9.57 ± 0.60	ns	33.63 ± 1.10	35.57 ± 1.13	ns
Neurotubules (number/ μm^2 of ACS)	21.91 ± 1.12	33.13 ± 1.04	+++	13.33 ± 0.64	17.80 ± 0.27	+++

Cf. Legend to Table I.

also in the frog the density of the neurotubules at the level of the spinal roots proved to be lower in dorsal root fibres than in ventral root fibres. However, in the frog equal numbers of neurotubules were found in motor and sensory fibres immediately distal to the spinal ganglia. We have now found considerably higher tubular densities in rat saphenous nerve fibres than in MGN fibres of comparable sizes. This fact suggests an even more pronounced difference in the content of neurotubules between motor and sensory fibres of rat peripheral nerves, with lower values for motor nerve fibres than those presented for MGN in Table I; for it has to be taken into account that muscle nerves like the MGN contain a considerable percentage of sensory nerve fibres which could not be excluded by our sampling technique.

Zusammenfassung. Es ergab sich, dass die Neurotubulsdichte in den Axonen eines Hautnerven (Ratte, N. saphenus) klar höher ist als in gleich dicken Fasern eines Muskelnerven (N. gastrocnemius med.). Ein weiterer Befund leistet einen Beitrag zur Erklärung dieses Gegensatzes: Die Neurotubulsdichte in den peripheren Neuriten primärer sensibler Neurone unmittelbar distal des Spinalganglions (L_5) ist signifikant höher als in den zentralen Neuriten in Höhe der Hinterwurzel.

W. ZENKER, R. MAYR and H. GRUBER

2. Anatomisches Institut der Universität Wien,
Währingerstrasse 13, A-1090 Wien (Austria),
5 July 1974.

Visual Cell Coding: Factoring Dioptric Responses from Maintained Discharge

Maintained discharge, common throughout the visual system, has long fascinated and frustrated investigators¹⁻³. Although such activity is now generally accepted as physiological, and indeed vital, to the normal function of that sensory system (see the current review of LEVICK⁴), its very presence complicates the deciphering of transient event signals moving in those pathways.

While complex models have been explored for the analysis of maintained activity in the visual system⁵, certain forms of transient information can at times be extracted through very fundamental models, even in the presence of very high density 'noise' (e.g. 30 spikes/sec or more) generated by the conducting neurone. The relative success of such reduced factoring models depends, of course, on the regularity of the maintained discharge, and is illustrated here in relation to one of the most fundamental of transient signals in the visual pathways: the ongoing assessment of image focus on the retina⁶.

The responses cited are from amongst a cumulative sample of more than 300 neurones within the rabbit mesencephalon and visual cortex now studied. The animals were maintained under light urethane anesthesia (6.0 ml/kg body wt. of a 20% solution in saline, a dosage level found from earlier work to be no more detrimental to cell responsiveness than 'encéphale isolé' techniques). This was supplemented with 3.3 mg/kg body wt./h of gallamine triethiodide to prevent eye and body movements,

¹ H. K. HARTLINE, J. cell. comp. Physiol. 5, 229 (1934).

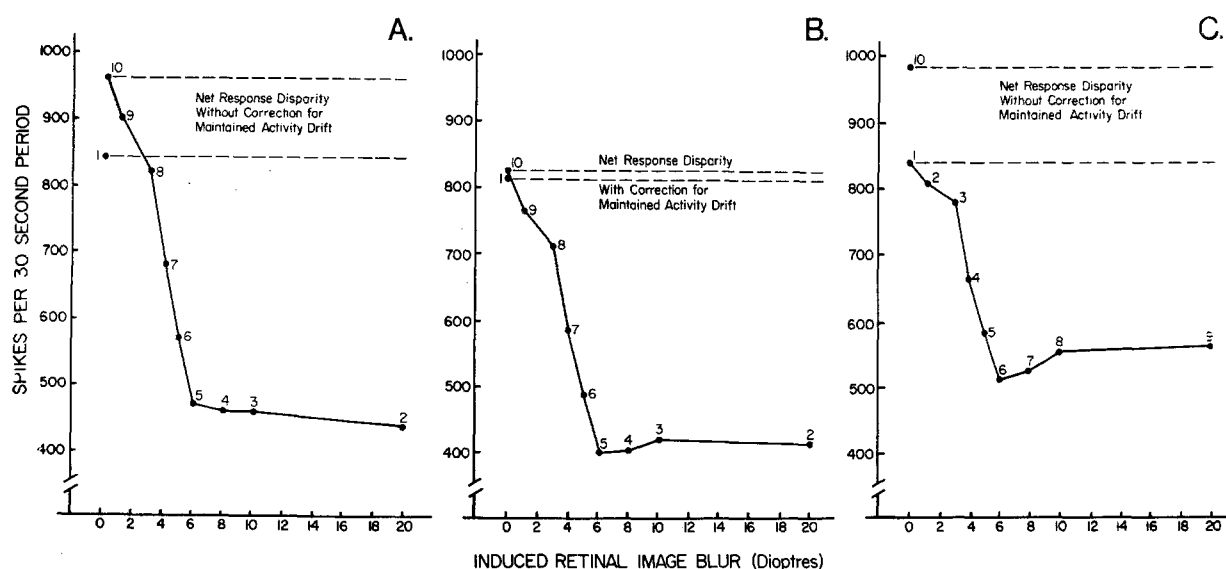
² E. D. ADRIAN, J. Physiol., Lond. 91, 66 (1937).

³ R. GRANIT, Acta physiol. scand. 1, 370 (1941).

⁴ W. R. LEVICK, *Handbook of Sensory Physiology* (Springer-Verlag, New York 1973), vol. 7, part A, p. 575.

⁵ H. B. BARLOW and W. R. LEVICK, J. Physiol., Lond. 202, 699 (1969).

⁶ R. M. HILL and H. IKEDA, Arch. Ophthalm. 85, 592 (1971).



Responses of a midbrain cell to a black edge moving across its receptive field once per sec, integrated over 30 sec periods. The test sets for each focal condition were spaced 90 sec apart, their order of presentation being indicated by the numbers next to the points. A) shows the actual spike totals recorded, i.e. without correction for maintained activity drift; B) shows the same data, corrected for maintained activity drift (see text); and C) shows the maximum case of transient response distortion expected, calculated on the assumption that the data could have been collected in the particular order indicated.