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Force development in smooth muscle strips of the hypertrophied urinary bladder of the rat after autonomic decentralization[1]

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Summary. Active length-tension relations for muscle strips of decentralized bladders differed from those of controls when comparisons were based on length in situ, but not when comparisons were based on lengths relative to optimum length for force development. The decentralized bladder behaved similarly to the denervated bladder, thus indicating that the presence of nerves was of no importance for the force production of directly stimulated muscle cells in the hypertrophied bladder.

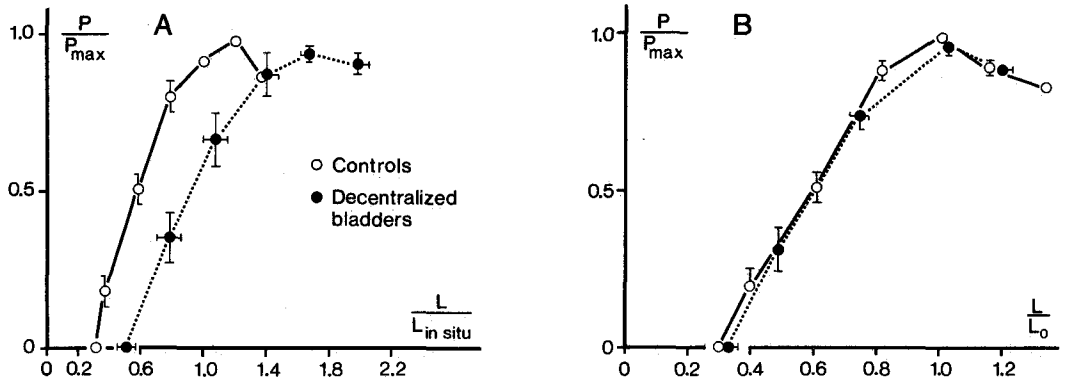
A rat in which the urinary bladder is either denervated or decentralized is unable to pass its urine. During the first few postoperative days, when the bladder is manually emptied, the bladder gains markedly in weight[2,3]. Recently, it was shown that length-tension relations for muscle strips of denervated bladders, studied in an organ bath, differed from those of strips of control bladders[4].

In contrast to the denervated bladder, the decentralized bladder is still supplied with postganglionic nerves. In the present study we investigated whether the presence of these nerves is of importance for the ability of muscle strips from hypertrophied bladders to develop force, using AC current as the mode of stimulation.

Materials and methods. Ten male adult rats of a Sprague-Dawley strain were used; the operation was carried out on five of them. Under ether anesthesia and with the aid of a dissecting microscope, the preganglionic nerves of both the pelvic and the hypogastric ganglion were bilaterally cut[3]. The decentralized bladders were emptied manually once a day.

The animals were killed by cervical fracture, the operated ones 10–14 days postoperatively. The bladders were removed and transferred to oxygenated Krebs solution, with which they were also filled to a volume of 0.75 ml. A longitudinal section was marked out, measured and dissected out. The strips were mounted in the organ bath of an apparatus in which muscle force and shortening could be measured[5]. Force and length output were recorded on a Devices MX 4 linear direct-writing oscillograph. The bathing medium was a Krebs solution of the following composition in mM: NaCl 115, KCl 4.73, $CaCl_2$ 2.5, $NaHCO_3$ 15.5, KH_2PO_4 1.19, and glucose 11.5. It was kept at 37 °C and bubbled with 4% CO_2 + 96% O_2, giving a pH of 7.4. The muscle strips were supramaximally stimulated by 5-sec periods of alternative current via 2 platinum electrodes placed in the bath. The stimulation period was long enough to produce a tension plateau. Time between stimulation was 60 sec. L_{min}, i.e. the length of the unloaded muscle strip, when stimulated, was determined. The length was then increased in steps to and beyond the optimum length (L_0) for active force development. After the experiments the strips were weighed, and cross sectional area at optimum length was calculated assuming a tissue density of 1.05 g per ml. All lengths were expressed in relation to the in situ length of the strip ($L_{in\ situ}$) and to L_0. The whole bladder weight was also recorded. Results are mean ± SE. Student's t-test for unpaired data was used.

Results and discussion. The mean wet weight of the 5 control bladders was 69 ± 5 mg, while that of the 5 decentralized bladders was 334 ± 34 mg; a difference which is significant at a p-level of < 0.005. The length to which the unloaded strip could shorten when stimulated showed a significant difference ($p < 0.01$) between strips of control bladders and those of decentralized bladders; the figures, when related to the length in situ ($L_{min}/L_{in\ situ}$), were 0.30 ± 0.02(5) for control bladders and 0.52 ± 0.06(5) for decentralized bladders. The active length-tension relations for strips of control and decentralized bladders differed markedly when comparisons were based on lengths relative to $L_{in\ situ}$ as can be seen in figure A. The maximal force (P_{max}) developed at optimum length (L_0), expressed in N/cm^2 cross sectional area was, however, about the same for strips of control bladders and for those of decentralized bladders, i.e. 5.9 ± 1.3(5) and 6.8 ± 1.6(5), respectively. When lengths were expressed relative to L_0, the active length-tension curves for both control and decentralized



Active length-tension relations of muscle strips of decentralized bladders and those of control bladders. Abscissa: Length relative to in situ length (A); length relative to optimum length (B). Ordinates indicate force P relative to maximum force value P_{max}. The results are grouped according to $L/L_{in\,situ}$ (A) and L/L_0 (B) into classes with a width of 0.2. Bars indicate ± SE. Number of observations in each class varies between 4 and 28.

bladders were superimposable (fig. B), and there was no difference in L_{min}; i.e. L_{min}/L_0 was $0.31 \pm 0.02(5)$ for strips of control bladders and $0.33 \pm 0.03(5)$ for strips of decentralized bladders.

Tetrodotoxin added to the bath (10^{-5} g/ml) had no effect on the contractions evoked by AC stimulation, thus indicating that the muscle cells were directly stimulated.

The present study therefore shows that the decentralized bladder behaves in a similar way to the denervated bladder previously examined[4]: strips of decentralized and denervated bladders shorten less in relation to the length in situ than those of control bladders; in relation to the in situ length the strips of decentralized and denervated bladders have to be stretched to a greater extent than those of control bladders to attain the optimum length for the development of active force. Maximal active force is the same both for strips of decentralized and denervated bladders and for those of control bladders; and further, when length-tension relations are expressed in relations to optimum length there is no difference between the curves for decentralized and denervated bladders and the curve for control bladders, neither is there any difference in the ability of unloaded strips to shorten when related to optimum length.

As judged by acetylcholinesterase staining[6] and choline acetyltransferase activity[3] most cholinergic nerves persist in the urinary bladders following decentralization.

According to the parameters studied in this investigation, the presence of these nerves does not seem to influence force production of muscle cells in the hypertrophied bladder.

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Butanol extracts from myelin fragments: Morphological and biochemical aspects of the re-formed membranes prepared from myelin butanol extracts

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Summary. The re-formed membranes prepared from butanol extracts of myelin were examined by morphological and biochemical methods. Using freeze-fracturing, re-formed membranes showed 2 types of assembly of membrane particles, i.e., myelin-like and cluster arrangements. Moreover, SDS-urea disc gel electrophoresis indicated that the protein composition of these membranes reflected that of the myelin fragments.

For the isolation of receptor components from biological tissues, an organic solvent extraction technique has been employed to isolate the proteolipid-like receptors[2-5], based on the assumption that the receptor components present in the biological membranes are closely associated with lipids. However, the question has been raised whether this method preserves the inherent biophysical nature of receptor components. Using negative staining for electron microscopy, Vásquez et al.[6] investigated the morphological features of the chloroform-methanol extracts from bovine and cat cerebral cortex, but their biophysical properties were not examined. The objective of the present work was to examine to what extent butanol extracts of myelin (proteolipids), which have a specific binding capacity for 5-hydroxytryptamine (5-HT)[7-9], retain their original biophysical properties. Freeze-fracturing reveals the hydrophobic central plane of biological membranes upon which the lipid-protein interactions are reflected in the occurrence of distinct particles, i.e., membrane particles[10]. Since such interaction may be a criterion for examining the biophysi-