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EFFECT OF SCN AND K IONS ON THE LENGTH-TENSION RELATION AND THE RESTING AND ACTION POTENTIALS IN RAT PAPILLARY MUSCLE.

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It has been shown that, in rat papillary muscle, the contractile force is potentiated by more than 25% by 25%-SCN and 5.7mM-K solutions, and the potentiation is associated with membrane depolarization in both cases (Iwamoto *et al.* 1989). We examined the effect of SCN and K ions on the contractile force and on the resting and action potentials over the length range from 0.7 to $1.0L_{max}$ where the Starling's law of the heart is obvious.

The resting and action potentials did not change significantly when the muscle length was varied from 0.7 to $1.0L_{max}$ in the standard solution. In 25%-SCN and 5.7mM-K solutions, the resting potential decreased by 5-20mV with the same amount of decrease in the action potential amplitude. These changes remained, however, unchanged by the change in muscle length from 0.7 to $1.0L_{max}$. On the other hand, the contractile force in response to a single electrical stimulus decreased from the maximum value to nearly zero with decreasing muscle length from 1.0 to $0.7L_{max}$ in both the standard and the SCN and K solutions, so that the degree of potentiation of contractile force remained the same irrespective of muscle length. These results indicate that (1) the Starling's law of the heart may not be mediated by the length-dependent depolarization, and (2) the SCN- and K-induced force potentiation is not associated with the shift of the active force-length relation.

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ACTIN-BOUND NUCLEOTIDE AND SLIDING MOVEMENT.

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It has been shown that the substitution of the bound nucleotide may cause some conformational change in actin. In an attempt to study the possible role of actin-bound nucleotide in the sliding movement between actin and myosin, we prepared several kinds of nucleotide-substituted F-actin from rabbit muscle. For effective displacement of bound nucleotides, we prepared G-actin containing ADP first by the quick dispersion of actin subunit from F-actin under sonic vibration, and then polymerization was induced by salt and phalloidin in the presence of the ATP analogs : adenylyl imidodiphosphate(AMPPNP), 8-bromoadenosine triphosphate(BrATP), or inosine triphosphate(ITP). HPLC analysis of the nucleotide content in the F-actin thus prepared showed that over 93% of the nucleotides were replaced for AMPPNP, over 99% for BrADP or IDP.

In order to measure the sliding mevement of the nucleotide-substituted F-actins on heavy meromyosin(HMM) or on myosin subfragment-1(S1), rhodamine-phalloidin was introduced as fluorescent probe in place of phalloidin, and assayed by an *in vitro* system on a video-enhanced fluorescent microscopy. No appreciable difference of the sliding velocities were observed among the nucleotide-substituted F-actins and the normal F-actin. They slided continuously on HMM or S1 at the velocitiy of about 10 μm/sec at 30°C. The results suggest that the actin-bound nucleotide may not play a significant role in the sliding movement between actin and myosin.

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FORCE-VELOCITY RELATION OF THE ATP-DEPENDENT SLIDING BETWEEN ACTIN AND MYOSIN AS DETERMINED BY AN *IN VITRO* FORCE MOVEMENT ASSAY SYSTEM.

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To eliminate the gap between muscle physiology and muscle biochemistry, we constructed a new *in vitro* force-movement assay system, in which myosin-coated polystyrene beads (diameter, 2.8μm) were made to slide on the actin filament arrays (actin cables) in giant algal cells and subjected to constant centrifugal forces by means of a centrifuge microscope. The bead movement was recorded with a video system (30frames/s) combined with a stroboscopic light source.

The bead continued moving on actin cables with a constant velocity under a centrifugal force directed opposite to the bead movement. The steady-state force-velocity (P-V) relation thus obtained was hyperbolic in shape for small loads, but deviated from hyperbola for large loads. Similar results were obtained for the beads with Po values ranging from 2 to 39 pN, indicating that the P-V relation reflects the properties of individual myosin heads rather than the change in number of myosin heads involved. Unexpectedly if the bead was subjected to centrifugal forces in the same direction as the bead movement, the velocity of bead movement decreased with increasing centrifugal forces, though the bead eventually detached from actin cables. This work was performed in collaboration with Drs Kamitsubo (Hitotsubashi Univ.) and Shimmen (Himeji Univ. of Tech.).

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MECHANISM OF PROPAGATION OF ACTION POTENTIAL AND THE ELECTRICAL CONSTANTS IN THE SWIMBLADDER MUSCLE OF A TELEOST *SEBASTICUS MARMORATUS*.

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The swimbladder muscle of a teleost fish *Sebasticus marmoratus* exhibits extremely rapid twitches to produce sounds for communication. To study the mode of nervous control in this muscle, we examined its electrical properties as well as its mode of motor nerve innervation with histochemical techniques.

The action potential showed little of no overshoot with a tendency to attenuate with distance when initiated by intracellular stimulation at one point on the muscle fiber. The above decremental propagation of action potential was consistent with the very large membrane capacitance (23-32μF/cm^2) and the very low membrane resistance (180-250Ω•cm^2). On the other hand, if the motor nerve innervating the muscle was stimulated, the action potential was set up at the area of nerve insertion and propagated along the fiber length with a velocity of 7m/s. In accordance with the decrementless propagation in response to motor nerve stimulation, histochemical studies showed the presence of many motor nerve terminals distributed along the entire fiber length. This, together with the very high velocity of action potentials propagation, indicates that the action potential propagates along the motor nerve. Histochemical studies also showed that this muscle consists of only one type of muscle fibers.