However, the soleus muscles did not gain any appreciable weight when their synergists were tenotomized, if their antagonists were excluded from function by denervation (Figure 1, II). In this type of hypertrophy muscle afferents are apparently not involved, since in deafferented soleus muscles the weight gain was comparable to that in muscles with intact sensory innervation (Figure 1, III).

When 'hypertrophying' muscles were subjected to static training, there was no increase in weight 7 days after the operation (Figure 1, IV), even though the residual hypertrophy at longer time intervals was somewhat enhanced (Figure 1, upper I, broken line). Combined excercise had a similar small effect.

It thus appears that the rapid hypertrophy which the soleus undergoes within a week after tenotomy of the gastrocnemius is transient in character and regresses almost completely by the 3rd postoperative week. This hypertrophy is not predominantly due to excessive use of the muscles, but rather to mechanical passive stretching by the antagonistic group, since denervation of the latter prevents the development of muscle hypertrophy. Additional exercise did not further increase the muscle weight increment at its peak as compared with the non-exercised animals. A reflex mechanism is apparently not involved, since even complete limb deafferentation did not prevent the development of muscle hypertrophy.

However, it cannot be excluded that part of this weight gain is actually true working hypertrophy, since a certain weight increment persists for longer periods of time (Figure 1, I) and has also been found in the plantaris and extensor digitorum longus muscles under analogous conditions[5].

Zusammenfassung. Nachweis, dass die schnelle Hypertrophie des M. soleus nach Tenotomie des synergistischen M. gastrocnemius durch passive Spannung des antagonistischen Muskels verursacht wird. Denervierung dieser Antagonisten verzögert eine Hypertrophie des M. soleus, während die Deafferenzierung des ganzen Gliedes ohne Effekt bleibt; auch zusätzliche Arbeitsleistung führt auf dem Höhepunkt der Hypertrophie zu keinerlei Gewichtszunahme.

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Electrophysiology Studies on the Cardiac Non-Function Mutation in the Mexican Axolotl *Ambystoma mexicanum*[1]

HUMPHREY[2] reported the discovery of a recessive and lethal gene *c* (for cardiac nonfunction) in *Ambystoma mexicanum*. Myogenic activity of cardiac muscle was absent in the homozygote of gene *c*, although early morphological developments were similar to those observed in normal embryos[3]. Mutant embryos later became edematous and their developing hearts became thin-walled, elongated and tortuous. Mutant hearts at Harrison stage 40 were only one cell layer thick, had excessive accumulation of yolk and lipid material, and lacked trabeculae which were present normally at stage 38. Mutant animals expired within 21 days after the time when hearts would normally demonstrate myogenic activity.

Transmembrane potential responses of in vivo embryonic hearts of normal *Ambystoma mexicanum* recorded prior to and immediately after onset of myogenic activity have been reported[4]. Typical resting potentials and action potentials, only present after onset of myogenic activity during Harrison stage 34 were recorded (Table). Acetylcholine, norepinephrine and electrical stimulation were without effect on quiescent hearts, in contrast to modification of cardiac activity after onset of myogenic activity[4].

The present study was undertaken to compare the electropharmacological induced responses of in vivo embryonic hearts of normal and homozygous cardiac mutants of *Ambystoma mexicanum*.

Materials and methods. Fertilized eggs from *Ambystoma mexicanum* were incubated until development approximated Harrison stage 34. All embryonic hearts of normal *Ambystoma mexicanum* begin to beat during and after stage 34, whereas cardiac mutants do not show myogenic activity during any stage of development. Individual embryos were transferred to a Syracuse watch dish layered with paraffin and 10 ml Steinberg's solution[4] maintained at 25 ± 0.1 °C. Wire restraints immobilized anesthetized embryos on the dorsal surface, and hearts were then exposed. Animals required 10 min to completely recover from anesthesia and experimental manipulation.

Transmembrane potentials[4] were recorded from 29 mutant homozygous for gene *c* and 29 heterozygous developed to stages 34–38. Hearts were directly stimulated 60–120/min and stimulation parameters were continually monitored. The influence of acetylcholine, norepinephrine and serotonin (0.1 µg/ml to 0.1 mg/ml) on myogenicity was determined. Only one drug was used with each embryo.

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Results and discussion. The mean resting potential of hearts of 29 cardiac mutants was not statistically different from that found for 29 normal hearts (Table). The resting potential magnitude was similar to that found for other inexcitable tissues[5]. The 3 drugs in all concentrations tested did not alter resting potential magnitudes or induce myogenic activity in any mutant embryos, whereas the heart rates of stage 34 + normal embryos were modified by drugs[4]. Electrical stimulation strength, sufficient at times to stimulate somatic muscles, in the presence or absence of these 3 drugs did not alter resting potential magnitudes or induce myogenic activity in mutant embryo hearts. These results suggest that myogenic factors essential to produce action potentials are absent in homozygous gene *c* mutants, and their heart cell membranes may remain in a stage of development similar to that found prior to stage 34. Mutant embryonic hearts never showed myogenic activity during stages 34–38, in contrast to myogenic activity always present during stages 34–38 in normal embryonic hearts.

HUMPHREY[6] transplanted primordial hearts of mutants into homozygous (+/+) or heterozygous (+/*c*) normals and produced myogenic activity. Conversely, transplanting normal primordial hearts into mutant recipients inhibited myogenic activity. The factors or substances controlling initiation of myogenic activity do not appear to be present in body fluids and it seems to be restricted to the region of the heart since parabiosis of normals with mutant siblings failed to induce myogenic activity in mutants or block myogenic activity in normal animals. The experiments reported here also suggest that the 3 neurohumors used were not the natural substances essential to initiate myogenic activity.

Summary of the measured characteristics of in vivo hearts of normal and mutant *Ambystoma mexicanum*

Characteristic	Tissue	Genotype	Stages 34–38
Resting potential (mV)	Random cell sample	Normal	24 ± 2[a]
		Mutant	24 ± 2
Action potential (mV)	Ventricle	Normal	32.9 ± 2
	Ventricle	Mutant	[b]
Heart rate (beats/min)		Normal	75 ± 18
		Mutant	[b]

[a] Mean ± S.E.M. [b] None observed.

JACOBSON and DUNCAN[7] reported that developing hearts may be under the influence of 1. an inducer that increases heart differentiation, 2. an inhibitor that delays heart differentiation, and 3. a stimulator that increases beating heart formation. Embryos of mutant gene *c* may influence synthesis of only 1. and/or 2. which results in modulation of membrane properties essential for excitation, conduction and action potentials. The results reported here suggest that homozygous gene *c* mutants have cardiac cell membranes that are inexcitable. This may be due to differences in membrane permeability to ions essential for excitation. Low resting potential magnitudes may be due primarily to a higher sodium and potassium permeability ratio in nonexcitable cells[5]. The effects of homozygosity for gene *c* seem restricted to cardiac tissues since electrical stimulation evokes somatic movements without concomitant responses in cardiac cells. This lack of cardiac contractility may be attributed to insufficient myofibrillar material[3]. Large numbers of mitochondria and other intracellular structures associated with pathological states have been reported[3]. It is not yet clear whether these cellular pathological-like conditions reflect only intracellular changes and activities or also an alteration in membrane response properties.

Résumé. Le mutant homozygote d'*Ambystoma mexicanum* qui n'a pas d'activité cardiaque myogénique, est aussi réfractaire aux modifications chimiques et électriques des potentiels membranaires et de la fréquence cardiaque. Il diffère en cela de l'animal normal au même stage de developpement embryonnaire.

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The Effect of pH on Quinine-Induced Contractures and Excitation-Contraction Coupling in Crustacean Skeletal Muscle

Although the action of quinine in both potentiating and depressing contractility of skeletal muscle has been known for some time[1,2], the ability of quinine to induce contractures has been only recently reported in frog[3] and crustacean[4] skeletal muscle. In one of these reports[3], it was noticed that changes in pH affected the contracture-induction ability of quinine, but the mode of action of pH changes during quinine activation, and the actual site of action of quinine on skeletal muscle are still not clear. This study of the effect of pH changes both on membrane potentials, quinine contractures and excitation-contraction coupling of crustacean skeletal muscle was carried out in an attempt to clarify the way in which quinine activates the contractile mechanism of skeletal muscle, and the modifying effect of pH on quinine activation.

Methods. The dactylopodite flexor muscle of the walking legs of the common shore crab, *Carcinus maenas* was used throughout this study. Details of procedure and preparations have already been published[4]. In some experiments, whole muscle preparations were used. Flexion of the dactylopodite was measured isometrically on a 50*g* strain gauge, the output of which was fed into a Tektronix 502A oscilloscope. Occasional experiments were carried out on isolated bundles of fibres dissected from the flexor muscle. These were mounted in a 5 ml perfusion chamber and tension was recorded isometrically on a 5 or 10*g* strain gauge. Membrane potentials of muscle fibres were measured with conventional 3*M* KCl electrodes, details of recording procedures have already been published[5]. The crab saline used was that of ATWOOD[6], containing (in m*M*), NaCl 520; KCl 10; $CaCl_2$ 11; $MgCl_2$ 8; $NaHCO_3$ 3, modified