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# The Dependence of Cardiac Membrane Excitation and Contractile Ability on Active Muscle Shortening (Cat Papillary Muscle)*

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**Abstract.** 1. A quick release of an isometrically contracting cat papillary muscle results in a depression of the ability to redevelop tension (deactivation) and an increase in the duration of the accompanying action potential (prolonged depolarization). The nature of the mechanical perturbation influencing both phenomena was investigated.

2. The prolongation of the action potential depends on the amplitude of the release and the time it is applied and, provided quick release-quick restretch cycles of less than 50 ms are used, on the duration of the cycle.

3. No change in action potential duration is observed, if initial muscle length, or the velocity of shortening is altered, or if the muscle is stretched at any time during contraction.

4. Although stretches and releases both have a "deactivating" effect on the muscle the effect is more pronounced with releases. This difference in "deactivation" is related to the prolongation of the action potential in so far as it is also controlled by the time and extent of release and release-restretch cycle duration, and is independent of shortening velocity.

5. Caffeine (8 mmol/l) in the bathing solution prolongs isometric tension development whilst the duration of the action potential is relatively unchanged. Under these conditions release-restretch cycles applied at times when the membrane has apparently repolarized, produce a deactivation and an afterdepolarization which can reach threshold to elicite an action potential.

6. If the membrane is partially depolarized by increasing extracellular potassium to 20 mmol/l, release-restretch cycles still induce deactivation but no change in the action potential.

7. The results are in keeping with the hypothesis that shortening during contraction partly contributes to the deactivating effect by reducing the concentration of internal free ionic calcium. This change in $[Ca]_i$ decreases the outward potassium current to produce a prolongation of depolarization which can take the form of an increase in action potential duration or an afterdepolarization wave.

**Key words:** Cardiac muscle – Electro-mechanic coupling – Mechano-electric recoupling – Contractile deactivation – Active state

## Introduction

The notion that excitation contraction coupling in ventricular myocardium is not strictly unidirectional, has been demonstrated fairly recently (Kaufmann et al. 1971; Hennekes et al. 1977; Lab 1979). Thus releasing an isometrically contracting muscle results in a delayed repolarization. This prolongation of the action potential is then thought to partly determine the tension development over the next few beats.

Apart from some preliminary speculation, the precise mechanism by which the mechanical change alters the action potential has received little attention. While investigating this problem further it became necessary to extend aspects of some previous purely mechanical studies (Brady 1966; Edman and Nilsson 1971; Bozler 1972; Kaufmann et al. 1972; Reggiani et al. 1980). Accordingly we have set out first, to determine the mechanical responses of the preparation which follow various mechanical interventions, secondly, investigate the nature of the relationship between mechanical and electrical changes, and finally perform some experiments to allow some reasonable speculation about the mechanism of the feedback loop between contraction and excitation.

## Materials and Methods

The method used for this investigation (isotonic lever system, force transducer, load control unit, sucrose gap chamber) has been described in detail (Hennekes et al. 1977, 1978).

For the systematic study of the electrical changes, recordings of action potentials were obtained with the sucrose gap technique. For the experiments in which mechanical changes only were measured, a simple test chamber was used because the sucrose gap introduced a compliance which interfered with the mechanical recordings. In the latter chamber, confirmatory microelectrode recordings were obtained for virtually all the sucrose gap studies.

In all experiments the initial muscle length ($L_i$) was adjusted to a standard preload of $0.007\,Nmm^{-2}$ and the average muscle length attained corresponded to $l_{max}$. Rectangular stimuli of twice threshold intensity and 0.3 ms duration were applied at 3 s intervals throughout the experiments.

The study was carried out on 49 preparations and each of the experiments in the figures is one example of more than ten similar observations in not less than five different preparations. Each result demonstrated is a consistent finding in all the experiments and there were no exceptions.

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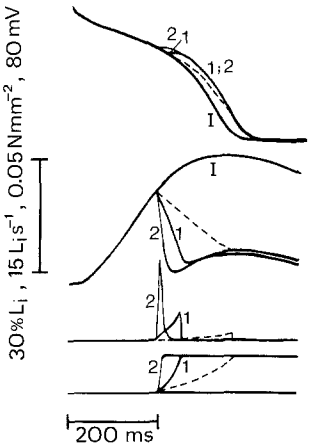


**Fig. 1.** Effect of different shortening velocities on action potential duration and tension redevelopment. Traces from above down: action potential, tension, shortening velocity, displacement. Although the maximum velocity of a release (200 ms after stimulus) is changed from 4 (trace 1) to 12 (trace 2) $L_i s^{-1}$, the effects on action potential prolongation and on tension redevelopment are almost identical in the two releases. For comparison isometric contractions are also superimposed (I). If, however, the release velocity is slowed down further (dotted trace), the action potential prolongation starts later

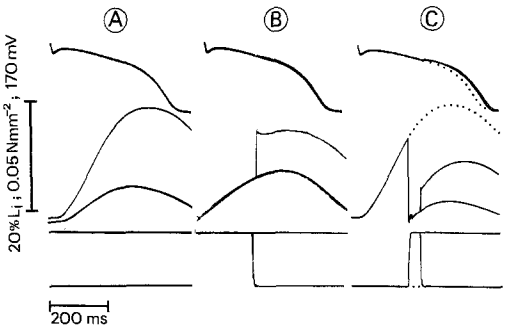


**Fig. 2.** Three examples in which changing the mechanical conditions of contraction of cat papillary muscle produce no detectable influence on the excitation process. Traces from above down: action potential, tension, length. *Part A* demonstrates two isometric contractions at two different muscle lengths ($L_i$ and 90% $L_i$ resp.). *Part B* shows an isometric contraction at 90% $L_i$ and another contraction which starts at 90% $L_i$ and which is stretched to 100% $L_i$ 200 ms after stimulus. *Part C* compares two isometric contractions one following a release from 100% to 90% $L_i$ 200 ms after stimulus, and another at 100% $L_i$ but which has been interrupted 200 ms after stimulus by a release-restretch (QR-QS) cycle 50 ms in duration. The dotted lines indicate uninterrupted isometric conditions with accompanying action potential and tension. In each of the three situations compared the excitation process remains unaltered

## Results

### *A. Effects of Changing Mechanical Conditions on the Action Potential*

*1. Velocity of Shortening.* Previous experiments have shown that shortening of cat papillary muscle altered the configuration of the action potential (Kaufmann et al. 1971; Hennekes et al. 1977). To test under more defined conditions the meaning of the velocity of shortening in particular we changed the maximum shortening velocity of a quick release from 4 – 12 $L_i s^{-1}$ [Fig. 1, trace (1) and (2)] keeping shortening amplitude constant (at 10% $L_i$) and applying both release interventions at about the same time after stimulus (200 ms).

Both release interventions induced the same action potential prolongation. If the difference between the shortening velocities compared exceeded the range of velocities demonstrated here (4 – 12 $L_i s^{-1}$, i.e. using values less than 4 $L_i s^{-1}$), there is a difference in the action potential configuration (dotted trace in Fig. 1). The slower the release velocity the later the action potential prolongation starts. In each case the action potentials, measured at 90% repolarization, are prolonged to the same duration as soon as the final length is attained.

*2. Initial Length ($L_i$) Changes and Stretches.* In contrast to the effect of a release it is of interest to see if stretching the muscle results in an equivalent abbreviation. First, changing the initial muscle length of an isometrically beating cat papillary muscle significantly varies the amount of tension development but the action potential configuration remains unaltered (Fig. 2A). Secondly, stretching the muscle at any time during contraction also does not alter the action potential although it is accompanied by a significant length and tension change (Fig. 2B and Hennekes et al. 1977; Fig. 2). Even (re)stretching the muscle after a series of preceding releases [i.e. switching from quick release (QR) to quick release-restretch (QR-QS) cycles] has no additional effect on the action potential configuration (Fig. 2C). The action potentials are similarly prolonged by the release as well by the release-restretch as compared with an action potential under purely isometric conditions (indicated by dotted lines).

*3. Effect of the Duration of Quick Release-Quick Stretch (QR-QS) Cycles on Action Potential and Tension.* In order first to test the degree to which the duration of a QR-QS cycle influences the prolongation of the action potential, a series of experiments was performed in which the duration of a QR-QS cycle, 10% $L_i$ in amplitude, was varied between 10 and 500 ms (Fig. 3A). The action potential and tension development are compared with those obtained under isometric conditions. A graph of these experiments is shown in Fig. 3B, in which the change in action potential duration measured at 50% repolarization level and expressed as a percentage of the "isometric" action potential duration, is plotted against the duration of the QR-QS intervention. Brief QR-QS cycles are less effective in prolonging the action potential duration than longer ones. The maximum effect is reached with transient releases of more than 50 ms duration. (It is of interest to note that a similar behaviour applies for QS-QR cycles, i.e. the longer the QS-QR duration the more pronounced is the accompanying release-related AP prolongation until a maximum effect is reached with interventions of more than 70 ms in duration.) The finding implies that the action potential is similarly prolonged whether it is induced by a sole quick release or by a quick release-restretch cycle, provided the latter is more than 50 ms in duration (see also Fig. 2C). A 50 ms QR-QS is the intervention used in the following experiments.

*4. Effect of Time, the Stimulus-Intervention Interval, on the Action Potential.* In Fig. 4 it is shown that the degree of action potential prolongation depends on when the release occurs: QR-QS cycles of 50 ms duration were applied during isometric contractions at various times after the stimulus. The

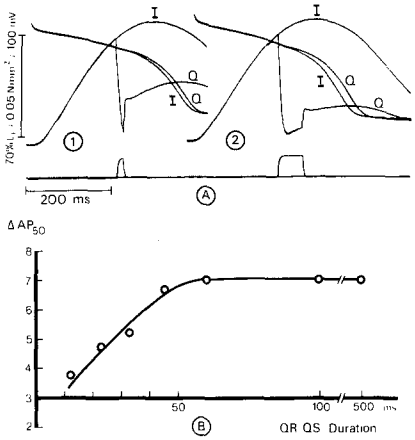


**Fig. 3.** The effect of varying the QR-QS cycle duration on tension and action potential. *Part A:* two QR-QS cycles 200 ms after stimulus are applied during isometric contraction [10 ms duration in (1) and 70 ms duration in (2)]. The duration of the intervention determines how much the action potential is prolonged and tension redevelopment is diminished after restretch. Traces from above: action potential, tension, displacement. The records labelled with subscript Q and I are related to the QR-QS cycle and pure isometric contraction respectively. *Part B:* Plot of QR-QS duration (intervention at 200 ms after stimulus) against the accompanying action potential prolongation (expressed as a percentage of the undisturbed isometric action potential measured at 50% repolarization level). Note the graph flattens out at a QR-QS duration of just under 50 ms

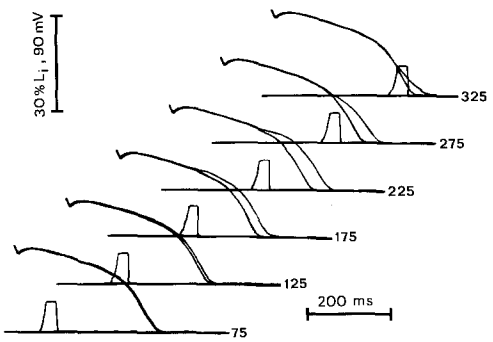


**Fig. 4.** Effect of quick release – quick stretch (QR-QS) cycles at different times after stimulus on action potential duration (stimulus intervention interval). During isometric contraction (shortest action potential in each pair of superimposed action potentials) QR-QS cycles of 50 ms in duration are applied at stimulus intervention intervals of 75, 125, 175, 225, 275 and 325 ms. These interventions induce different degrees of action potential prolongation. The optimum is reached with releases applied at about 225 ms after stimulus

concomitant action potentials were recorded and superimposed on the reference "isometric" action potential. Clearly, releases occurring prior to or early during contraction (until about 75 ms after stimulus) do not change the action potential. However, with releases applied 100 ms after the stimulus or later, the action potential is progressively prolonged. A maximum is reached at about mid-plateau (225 ms after stimulus) after which the prolongation declines.

*5. Effect of Changing the Amplitude of Shortening on the Action Potential.* The experiment in Fig. 5A demonstrates the effects on the action potential of varying the amplitude of QR-QS cycles, 50 ms in duration and applied 225 ms after

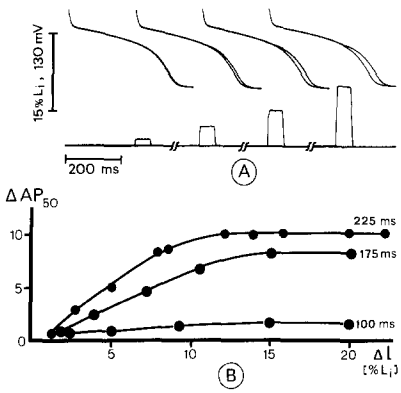


**Fig. 5.** The effect of varying the amplitude of QR-QS cycles of constant duration on action potential prolongation. *Part A:* QR-QS cycles (lower trace) 225 ms after stimulus are varied in amplitude between 2 and 11% $L_i$. The accompanying action potentials which are prolonged according to the extent of the release, are superimposed on the undisturbed "isometric" action potential. *Part B:* graph of variation of the extent of shortening ($\Delta l$) plotted against the prolongation of action potential derived as in Fig. 3. Three curves are presented using QR-QS cycles 100, 175 and 225 ms after stimulus. The curves form functions which flatten out at shortening amplitudes between 10 and 15% $L_i$

stimulus. The action potential was progressively prolonged with increases in amplitude of the release until a maximum was reached with releases of more than 15% $L_i$ (Fig. 5B). In the graph of this figure the QR-QS amplitude is varied at three different times during contraction. The resulting prolongation of the action potential duration are plotted against the amplitude of the intervention. Each curve represents a function which reaches its plateau at about the same shortening amplitude (i.e. between 10–15% $L_i$).

### *B. Differences in Mechanical Deactivation Produced by Stretches and Releases*

It appears so far that the mechanism underlying mechanically induced changes in action potential is unidirectional in that lengthening (stretch) at any time during contraction does not change the action potential configuration but releases do. After a release during contraction, the muscle does not reach the active tension it should develop according to its new position on the length active tension curve (Brady 1965, 1966, 1968; Kaufmann et al. 1972): an "uncoupling" effect (Brady 1968) or deactivation. A consistent observation during these experiments was that whenever a mechanical intervention induced a prolongation of the action potential this deactivation of contraction accompanied the intervention. Some mechanical changes, however, do not produce an action potential change yet they can cause deactivation: a stretch for example (Brady 1965, 1966, 1968; Kaufmann et al. 1972). So one could suggest that perhaps there are differences between the deactivation produced by the mechanical interventions which change the action potential duration and those that do not. The following experiments try to test this possibility by the example of a muscle as a whole.

*1. Stimulus Intervention Interval.* In Fig. 6 experiments are shown in which the muscle first contracted at 90% $L_i$ under steady state conditions (upper part of panels A and B). It was

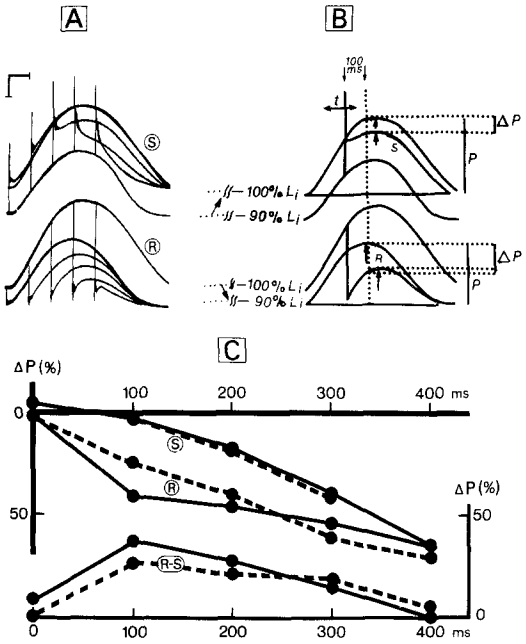


**Fig. 6.** The effect of stretches and releases on tension redevelopment as depending on the stimulus intervention interval. *Panel A and B* demonstrate recordings and a schematic drawing, labelled to clarify the procedure (for detailed information see text). *Panel C:* the drop in tension ($\Delta P$) induced by either intervention (R and S) is expressed as a percentage of the tension (P) developed by the muscle in the undisturbed contraction [$\Delta P/P \times 100 \triangleq \Delta P$ (%)] and is plotted against the time after stimulus, the intervention took place. The curve (R-S) represents the difference between R and S thus representing the augmented deactivation brought about by releases as opposed to stretches. The maximal difference in deactivation induced by the 10% $L_i$ displacements ranges between 25 and 35% (no sucrose gap in this preparation). *Solid lines:* $\Delta P$ measured 100 ms after the intervention (see B). *Dotted lines:* $\Delta P$ measured at peak isometric and peak post-intervention tension (as indicated by arrows in the scheme B)

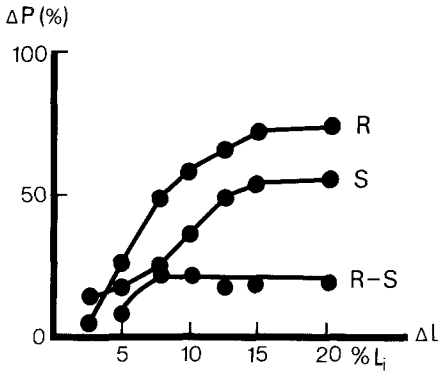


**Fig. 7.** The effect of stretches and releases on deactivation as depending on the amplitude of displacement. Releases and stretches of different amplitudes are applied 250 ms after stimulus and are plotted against the reduction in tension redevelopment (= deactivation) they induce 100 ms later. Note that the difference between the curves becomes constant with displacements greater than about 8% $L_i$. A similar relationship is obtained by relating post-intervention peak force to control peak force like in Fig. 6 (no sucrose gap in this preparation)

then stretched at different times (t), during different beats, to 100% $L_i$. After each stretch the muscle was allowed to regain a steady state at 90% $L_i$. This avoids tension transients (Hennekes et al. 1977) which make mechanical measurements unreliable. Each stretched contraction was compared with a control contraction at 100% $L_i$, the length to which the muscle is stretched 300 ms before stimulation. Stretches applied at the time of stimulation or at various intervals until about 100 ms after stimulation, result in tension curves that superimpose or even slightly exceed the "control curve" (upper part Fig. 6A). From about 200 ms after stimulation stretches become progressively effective in deactivating the muscle.

The reverse operation is shown in the lower parts of panels 6A and B. A muscle contracting under steady state conditions at 100% $L_i$ is released to 90% $L_i$ at the time of stimulation and at different times thereafter. Control reference in this case is an undisturbed isometric contraction when initial muscle length is changed from 100% to 90% $L_i$ 300 ms before stimulus. A contraction released at the time of stimulation superimposes on the control contraction. But with releases applied only a few milliseconds later the deactivating effect of the release appears.

In the graph of Fig. 6C an attempt is made to quantify the efficacy of both, stretches and releases as related to the stimulus-intervention interval, and to calculate their difference. To obtain a simple quantitative measure of the deactivating efficacy of either intervention we measured i) the peak force redeveloped after either intervention and compared it with the peak force developed in the undisturbed contraction at the same length (dashed lines in the graph) and ii) the redevelopment in tension at an arbitrarily chosen point of time of 100 ms after the intervention and compared it with the corresponding control contraction as defined above (solid lines in the graph). $\Delta P$ is expressed as a percentage of the tension developed in the control contraction at peak force or at the corresponding time resp. Clearly at any given time after stimulus the release operation is more effective in producing deactivation than is the same amount of stretch. Graph (S) relates to stretch interventions and (R) to release interventions. The difference in the deactivating effect of stretches and releases may be calculated and is indicated by (R-S). The difference is most prominent with interventions applied between 100 and 200 ms after stimulus. Although the deactivating efficacy of the release may be overestimated as compared to that of a stretch by the two rather arbitrary methods of evaluation and although a stretch may be a rather complex molecular mechanism as compared to a release (Housmans and Brutsaert 1976; Edman 1980a, b), it is worth to note, that the "difference-curve" (R-S) starts at zero, reaches its maximum with interventions applied at already 100 ms after stimulus and declines to zero again at times of peak isometric tension. A similar relationship applies to the action potential prolongation as depending on the stimulus-intervention interval (Fig. 4).

*2. Amplitude of Intervention.* In the graph of Fig. 7, data are collected on experiments in which the amplitude of releases and stretches applied at a fixed time of 250 ms after stimulus is varied between 2.5% and 20% $L_i$. This stimulus intervention interval is chosen because at 250 ms after stimulus not only the deactivation efficacy of the stretch as well as release intervention is clearly seen, but also their difference is most prominent (cf. Fig. 6C). The experimental procedure and data evaluation is similar to that described in Fig. 6. For the "release curve" the muscle is released, in each case, after a series of isometric steady state contractions at 100% $L_i$ to

various new lengths. As in Fig. 6, the tension trace of the released contraction is compared with a control curve which is an undisturbed isometric contraction released 300 ms before stimulus to the same new lengths. Conversely, for the "stretch curve" in the figure the muscle is stretched from a steady state at various initial lengths to the final length, 100 % $L_i$. Post-stretch tension is compared with the control curve which is an isometric contraction at 100 % $L_i$. The muscle is stretched to this length, 300 ms before stimulus during steady state conditions, from the various initial muscle lengths. Thus stretches and releases may be regarded as reverse operations covering the same range of absolute muscle lengths (e.g. release from 100 % to 90 % $L_i$ and stretch from 90 % to 100 % $L_i$: $\Delta 1 = 10\,\% L_i$). From the graph in Fig. 7 it is clear that the efficacy of both types of intervention (releases, curve R; stretches, curve S) on the deactivation depends on the extent of the displacement. However, a release always produces a greater deactivation than does a stretch, provided the amplitudes of interventions compared exceed 4 % $Li_i$. Also the calculated difference between both curves [curve (R-S) in this figure] demonstrates the greater deactivation induced by releases. This curve also bears some resemblance to the curves in Fig. 5, relating the action potential prolongation to the extent of shortening.

It is a general finding that at least in the length range between 115 and 80 % $L_i$, a release is always more effective in deactivating the remainder of the contraction than a stretch of comparable amplitude. It turns out consistently that if applying a stretch or release of the same deactivating efficacy (i.e. of different amplitude), only the release is able to influence the AP.

### *C. Changing the "Active State": The Effect of Caffeine on the Contraction-Excitation Feedback Phenomenon*

If the time course of activation might be a determinant of the contraction excitation recoupling loop a possible approach in testing this hypothesis is to dissociate – for instance by caffeine – the time course of activation and that of the action potential the two of which roughly coincide in normal ventricular myocardium. Caffeine is known to increase and prolong the activation of cardiac muscle (Blinks et al. 1972) while leaving the action potential duration relatively unaffected.

*1. The Effect of Releases 200 ms after Stimulus.* As shown in the preceding sections, a QR-QS cycle applied 200 ms after stimulus under normal conditions is highly effective in both prolonging the action potential and deactivating the muscle (Fig. 8A). After the addition of caffeine, however, the same intervention cannot produce an action potential alteration (Fig. 8B) and, at the same times, is much less effective in causing mechanical deactivation.

With caffeine the onset of contraction is slow but tension development is augmented in amplitude and duration (Blinks et al. 1972). Therefore, a QR-QS cycle occurring 200 ms after stimulus, if related to the time course of this particular active state, may be considered as a comparatively early intervention, and, it is not unexpected that there is less deactivation in the caffeine contraction as compared with the same intervention in a normal contraction.

*2. Stimulus Intervention Interval.* In Fig. 9 recordings are shown from a caffeine treated muscle in which QR-QS cycles are applied at progressively later times after stimulus.

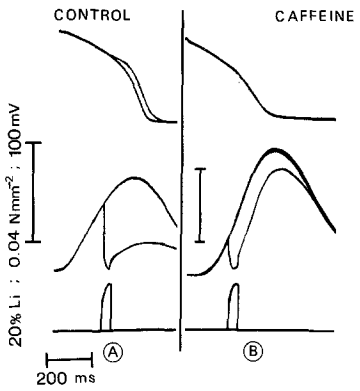


**Fig. 8.** The effect of 8 mmol/l caffeine on the release induced prolongation of action potential. *Part A:* control conditions. Changing from a purely isometric contraction to QR-QS conditions (10 % $L_i$, 200 ms after stimulus) results in an immediate prolongation of the action potential duration. *Part B:* The same mechanical procedure when applied with caffeine in the solution, does not alter the action potential duration. Note that under caffeine the isometric contraction is prolonged in relation to the action potential duration. The latter is nearly the same as the control action potential. The QR-QS intervention produces little deactivation. (The late onset of the "Caffeine-contraction" is overestimated in the figure due to the series elasticity of the sucrose gap region and the comparably high force developed by the muscle thus giving rise to some unregistrable shortening within the active part of the muscle)

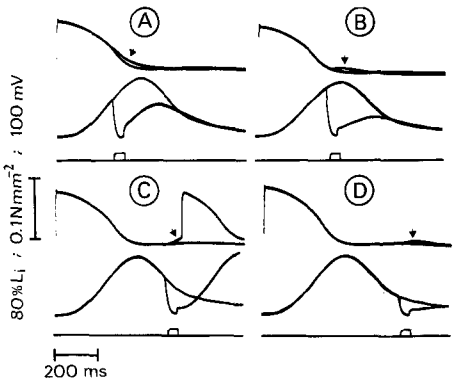


**Fig. 9.** The effect of 8 mmol/l caffeine on the changes in action potential and tension produced by varying the stimulus intervention interval. Traces from above down in each panel: action potential, tension, displacement. QR-QS cycles (10 % $L_i$) applied 200 ms after stimulus and later, prolong the repolarization process (e.g. *panel A*, 280 ms after stimulus) or produce a depolarization wave (*panel B and D*, 400 and 700 ms after stimulus). In *panel C*, 525 ms after stimulus, a depolarization is seen which reaches threshold to elicit an extra action potential

If we take into account the prolonged time course of the "caffeine contraction", the deactivation potency of the QR-QS cycles increases with time (the stimulus-intervention interval) in a similar fashion as compared to control conditions. But the release induced alterations of the membrane potential now reappear and have a similarly extended time course as compared to the deactivation mentioned above. However, we have the interesting situation that mechanical and electrical "recoupling efficacy" reach a maximum when repolarization is complete: the action potential has terminated and the membrane potential has attained almost its resting value. In this situation the QR-QS cycles induce late afterdepolarizations lasting about 200 ms, whereas under

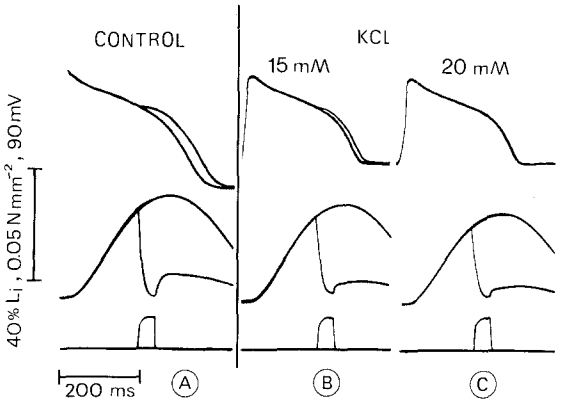


**Fig. 10.** The effect of increasing the potassium concentration in the perfusing solution on the contraction-excitation recoupling loop. Traces in each panel from above down: action potential, tension, displacement. *Part A:* shows the control experiment, in which a QR-QS intervention (10% $L_i$, 200 ms after stimulus) induces a prolongation of action potential duration. *Part B:* after increasing extracellular [KCl] to 15 mmol/l, the action potential prolongation produced by the same QR-QS cycle as in Part A is reduced. *Part C:* the prolongation is abolished when $[KCl]_o$ is increased to 20 mmol/l. The tension redevelopment in all three cases is similarly reduced

control conditions 500 ms after stimulus there is no detectable effect of releases on the membrane. The magnitude of these depolarizations vary with the time after stimulus when the QR-QS cycle is applied. Under the particular conditions described in Fig. 9 (8 mmol/l caffeine), the optimal amplitude of the depolarization induced by 10% $L_i$ QR-QS cycles, is attained with releases applied 500 – 600 ms after stimulus. In this case, the afterdepolarization reaches threshold to elicit an extra action potential (Fig. 9C).

*3. Amplitude of Displacement.* Increasing the extent of the release increases the amplitude and the rate of the induced depolarizations. The threshold for the new action potential is reached sooner. The latency period i.e. the time between the beginning of the release and the first detectable change in membrane potential is less than 10 ms in all the experiments.

### *D. Changing the Action Potential: Increasing Extracellular Potassium Concentration*

The two ions mainly contributing to the transmembrane currents at the time the mechanical interventions are most effective are $Ca^{2+}$ (Beeler and Reuter 1970, 1977) and $K^+$ (cf Noble 1975). The mechanically induced prolongation of the action potential could thus either be due to an increase in inward calcium current or to a decrease in outward potassium current. Tritthart et al. (1976) showed that partial depolarization of the membrane with potassium produced an action potential with characteristics suggesting it was predominantly calcium mediated. If the action potential prolongation induced by a QR-QS cycle is due to an augmented inward calcium current then the prolongation should appear when applying a QR-QS cycle during this type of action potential. We investigated the effect of increasing extracellular potassium on release-induced changes in action potential (Fig. 10A). Under normal Tyrode solution ($[KCl]_o$ = 2.68 mmol/l) a QR-QS cycle 200 ms after stimulus produces a prolongation of action potential of about 11% of the duration under isometric conditions. Increasing extracellular potassium concentration to 15 mmol/l reduces resting potential, action potential upstroke velocity and overshoot (Tritthart et al. 1976).

In the present experiments the change in [KCl] also reduced the ability of the QR-QS cycle to prolong the action potential to about a third of that under control conditions (Fig. 10B). At a [KCl] of 20 mmol/l, no change in action potential configuration is elicited by the QR-QS cycle (Fig. 10C), regardless of the extent of the release or the time after stimulus it occurs. Yet under these conditions, tension development and the mechanical response to the intervention (= deactivating effect) is virtually unaffected compared with control.

## Discussion

The results of this study show that in cat papillary muscle shortening during the later phases of contraction influences both the membrane excitation process and the redevelopment of tension. They also show that there is a quantitative but most probably also a qualitative difference in the mechanical deactivation produced by either a stretch or a release. It appears that mechanical deactivation contains a unidirectional component such that at least in the muscle as a whole releases are more effective in producing deactivation than stretches of equal amplitude. The "surplus in deactivation" with releases and the effects on the membrane excitation, manifest as a prolongation of the action potential (a depolarizing effect in essence) are strikingly parallel. They both appear to depend on the same mechanical parameter (shortening during a certain phase of the contractile cycle). The two phenomena may either condition each other or may be two independent expressions of a common basic event. For reasons discussed below, we think the second possibility is the more likely one.

Many of the changes in contraction presented here are in keeping with those of Edman 1971, 1975, 1976 observed in skeletal muscle. He suggested that "the depressant effect is based on a structural change in the myofilament system". However, with cardiac muscle we cannot exclude the possibility, as Edman does for skeletal muscle, that changes in activator calcium concentration might be involved. A change in the internal free calcium concentration may be a reasonable way of explaining why the mechanical events are so closely related to the electrical events.

It has to be considered whether the effects on action potential configuration observed might simply be the result of alterations in cellular architecture, accompanied by changes in membrane properties. However, besides the objections raised already earlier (Kaufmann et al. 1971; Lab 1978) this hypothesis appears unlikely because the mechanism underlying the mechanically induced action potential alteration is unidirectional[1] (Fig. 2). This argument also applies if the mechanisms were related to changes in architecture leading to localized potassium accumulation (cf Kline and Morad 1976; Attwell et al. 1977). In this case a stretch should induce a repolarizing effect: an abbreviation of the action potential. It appears further, that a prerequisite for this mechanism to operate is the muscle to be in the active state (see Caffeine experiments).

The experiment shown in Fig. 10 suggests that a potassium outward current rather than an inward $Ca^{2+}$ current

1 That means it cannot be elicited or counteracted by stretches

during repolarization may somehow be involved. There is, in fact, evidence that an outward current, probably carried by potassium, is influenced by internal free calcium (Isenberg 1975; Bassingthwaighte et al. 1976; Siegelbaum et al. 1977). Augmenting internal free calcium results in an accelerated repolarization of the action potential, or even in a hyperpolarization. Reducing internal calcium reduces this outward $K^+$ current and will prolong the action potential. Since a delayed repolarization is observed after a release, one may ask whether shortening might somehow reduce the amount of internal free calcium. Indeed, a preliminary observation in keeping with this possibility has been made by Allen (1978).

Also part of an augmented deactivating effect of releases, as compared with stretches (Fig. 7), could be interpreted as a consequence of such a diminution of activator calcium.

From the above, especially from the exclusively shortening dependent effects on the action potential, one may deduce the hypothesis that active shortening in heart muscle induces transient $[Ca^{2+}]_i$ changes at least during a certain period of the contractile cycle. These changes in turn may cause a reduction in outward potassium current and thus by sustaining the depolarization will prolong the action potential. This concept, of course, is highly tentative but could not only explain the parallel effects of releases on both, the action potential duration and the deactivation, but also the delayed afterdepolarizations which occur with late releases in Caffeine treated muscles.

The phenomena appear not to be related to length dependent changes in calcium release or affinity (Allen et al. 1974; Fabiato and Fabiato 1975; Gordon and Ridgway 1976; Jewell 1977) because the process feeding back from contraction to excitation is not dependent on the initial and final muscle length (see e.g. QR-QS cycle and stretches, Fig. 2). Therefore we suggest that shortening itself might be a mechanism which sequestrates free internal $Ca^{2+}$. This might also explain the differences in deactivation when comparing the effects of stretches and releases on tension redevelopment.

Under our experimental circumstances the mechanism by which stretches and releases produce deactivation could perhaps be separated into two components: a structural change in the myofilament interaction as depending on the displacement in both directions including non-uniform movement and a reduction of activator calcium brought about by releases (shortening) only. Releases amongst other would be more effective than stretches in inhibiting reactivation, or causing deactivation, because both mechanisms are involved in the former manoeuvre.

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