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THE EFFECT OF ADRENALINE ON THE SMOOTH MUSCLE OF GUINEA-PIG TAENIA COLI IN RELATION TO THE DEGREE OF STRETCH

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The relaxation of intestinal smooth muscle by adrenaline is due to the cessation of the spontaneous discharge of action potentials which is associated with a rise in membrane potential (Bülbring, 1955; Burnstock, 1958). The magnitude of the hyperpolarization depends on the membrane potential prevailing at the moment when adrenaline is applied (Bülbring & Kuriyama, 1963). Though it is known that the membrane potential is related to the degree of stretch applied to the muscle it has not been possible in the past to set it with regularity at a certain level in different preparations, because the definition of the initial muscle length has been inadequate. Resting length cannot be determined because spontaneous activity prevents complete relaxation. *In situ* length depends on the spontaneous tone at the moment of excision. Thus preparations of equal *in situ* length, mounted at a certain length or at a certain tension, behave quite differently.

The relation between muscle length and muscle tension is complicated by the active production of tension in response to stretch, which prevents the distinction between resting and active tension, the latter being an unknown fraction of the total tension recorded. On the other hand, tension is related to the cross-sectional area of the muscle. When the muscle is stretched, the deformation of the muscle fibres can be measured not only by the increase in length but also by the reduction in cross-sectional area. An estimate of this is given by the ratio of muscle weight (mg) to its length (mm). The $W:L$ ratio takes into account the variable weight of different muscles, and thus it would be expected to provide the standard conditions required for the comparison of different preparations.

In the present work reproducible conditions have been obtained by stretching the muscle to a given length in relation to its weight. Within certain limits the initial membrane potential as well as the changes produced by adrenaline were found to be proportional to the $W:L$ ratio.

METHODS

Guinea-pigs of similar size, weighing about 400 g, were used. Pieces of taenia coli weighing 6–12 mg were dissected and weighed on a torsion balance. Their *in situ* length varied between 5 and 10 mm. One end of the taenia was attached to a mechano-electronic transducer valve (RCA-5734) which could be moved to extend the muscle strip, the other end was attached to a device by which it could also be extended. We did not use a Perspex disk to keep the muscle flat as described previously (Bülbring, 1955), otherwise the experimental method was similar.

The constant-temperature organ bath was heated by a Nicron wire heater controlled by a thermistor. A metal screen was interposed between the heating wire and the organ bath.

The electrical activity was recorded by the micro-electrode technique. The resistance of the micro-electrodes varied from 30 to 60 MΩ, and only electrodes with tip potentials of less than 5 mV were used for determination of the membrane potential. A Nihon Koden Ltd pre-amplifier was used.

The normal solution in all experiments was a modified Krebs's solution containing (mM): Na^+ 137·4, K^+ 5·9, Ca^{2+} 2·5, Mg^{2+} 1·2, Cl^- 134, HCO_3^- 15·5, $H_2PO_4^-$ 1·2 and glucose 11·5, equilibrated with a gas mixture of 3 % CO_2 and 97 % O_2 to give a pH of 7·3 at 35–36° C. It was prepared from isotonic stock solutions.

Adrenaline concentrations of 10^{-8} and 10^{-7} were produced in the isolated organ bath (capacity 2 ml.) by injecting 0·05 ml. of a 20 times stronger solution. This concentration was therefore present only at the moment of injection and was thereafter progressively diluted by the solution flowing continuously at 2 ml./min.

RESULTS

Effect of stretch on membrane activity

Figure 1 illustrates the effect of stretch on membrane potential, amplitude and rates of rise and fall of the action potential, spike frequency and tension. Abscissae express the degree of stretch as the $W:L$ ratio. This figure was obtained from the mean values of three experiments in which preparations of 5·9, 6·1 and 6·5 mg were used. When no stretch was applied the muscle curled up. The folding of the muscle, however, disappeared gradually and it contracted to a short lump. The $W:L$ ratio then usually exceeded 1·1 and as the tissue was quite loose the insertion of the micropipette was difficult. When the ratio was between 1·0 and 0·9 the muscle fixation was adequate for the insertion of the electrodes and at this length the passive tension was still zero. This will be described later (cf. Fig. 7). For each point on the graph the muscle was stretched from the condition in which $W:L$ was 1·0, and the measurements were made 10–15 min later. The procedure was repeated 3 times in each experiment, the order being changed.

The maximum membrane potential occurred at a $W:L$ ratio between 0·9 and 0·8. Increasing or reducing the stretch from this value reduced the membrane potential. Overshoot potentials were observed in a $W:L$ ratio range from 1·0 to 0·6. The highest rate of rise was observed in the range 0·9–0·7. At $W:L$ ratios greater than 0·7 the rate of fall was reduced more

markedly than the rate of rise. The tension development in Fig. 1 represents the total tension measured, i.e. passive as well as active tension. It changed in parallel to the spike frequency. However, overstretched tissue ($W:L$ ratio less than 0·5) had a low spike frequency while the tension remained high.

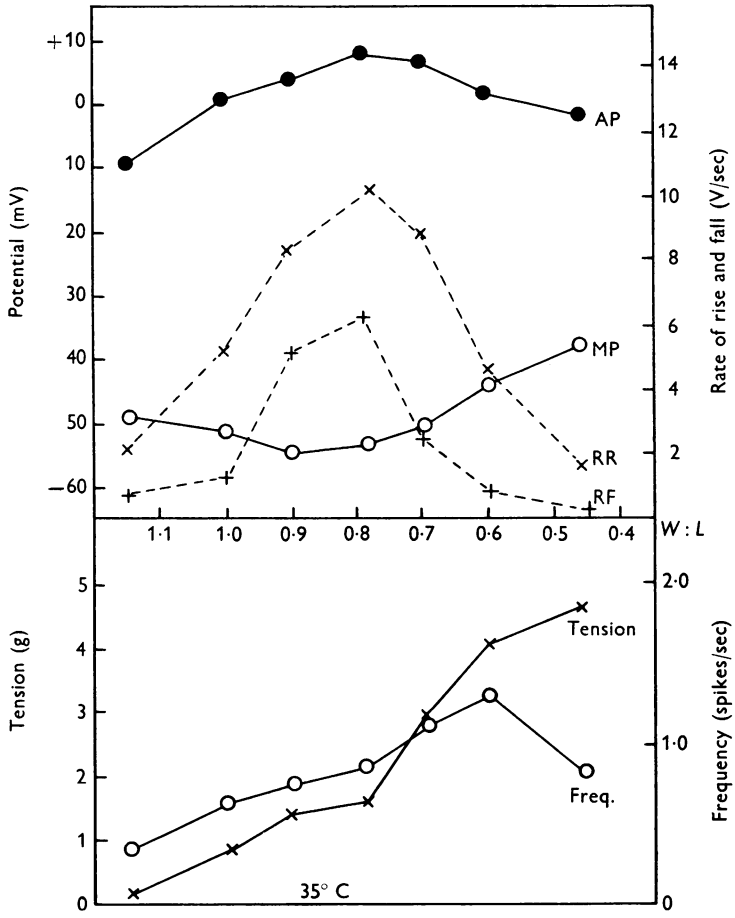

Fig. 1. The effect of stretch (defined as $W:L$ on abscissa) on membrane potential (MP), action potential (AP), rate of rise (RR) and fall (RF), spike frequency (Freq.) and tension. For description see text.

Figure 2 shows the simultaneous recording of the membrane activity and the tension at increasing stretch. Often in unstretched tissue a slow depolarization of the membrane preceding each spike was observed, similar to that seen in pace-maker cells of cardiac muscle (see also Fig. 5*a*). This depolarization did not always trigger a spike. When the muscle

was stretched the spike frequency increased and spike height became more regular. When it was stretched to a $W:L$ ratio of 0·7 the membrane became depolarized, and when the $W:L$ ratio was less than 0·5 the amplitude of the spike became irregular and an overshoot potential was no longer

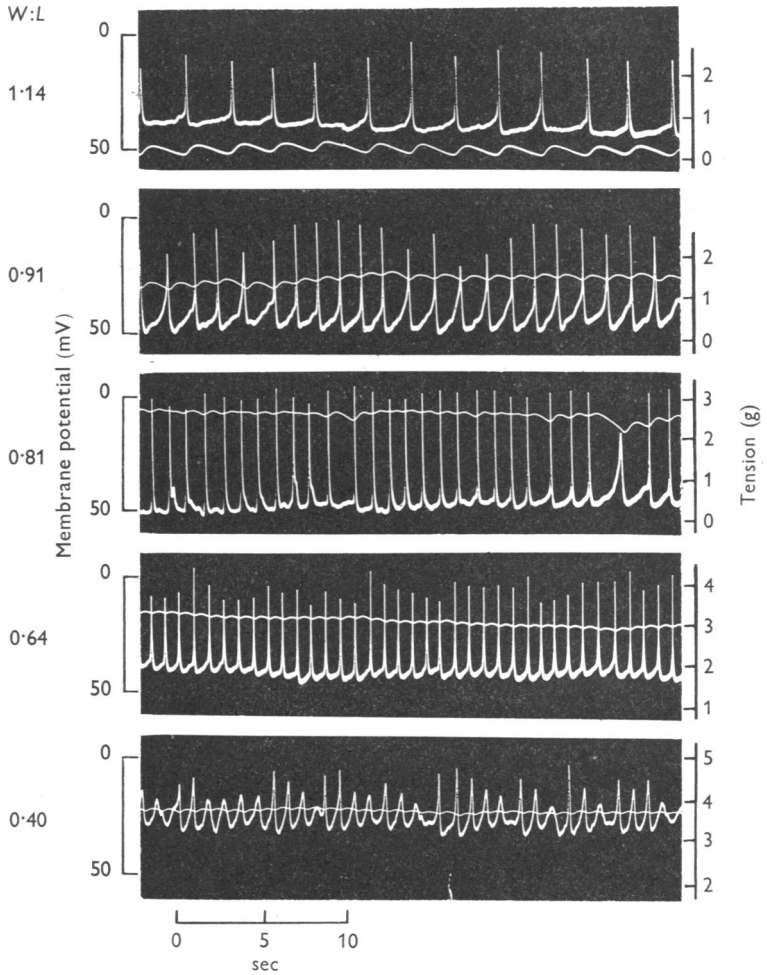

Fig. 2. Records from one experiment showing the effect of stretch on membrane potential, membrane activity, and tension. The degree of stretch is indicated by the values of $W:L$.

observed. Further stretching depolarized the membrane, sometimes to less than 30 mV, and blocked spike generation. The effects of stretch described so far were observations made after a steady state had been reached. The mean values observed in sixteen different preparations are shown in Table 1.

Accommodation of the membrane activity and tension after sudden stretch

In the present experiments a sudden stretch from $W:L$ ratio 1·2 to 0·5 could not be obtained in less than 1·5 sec. When this was applied, the tension rapidly increased and then fell to a steady level within 10–20 sec. This is shown in Fig. 3*a*. The curves are constructed from the mechanical

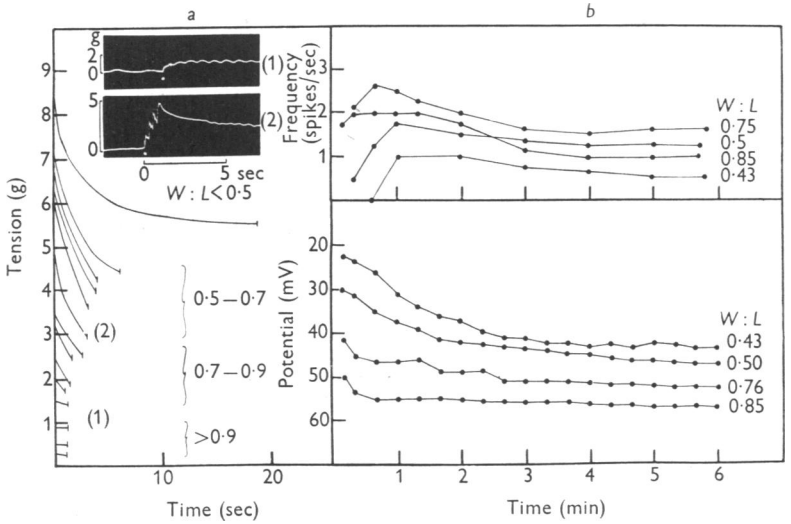

Fig. 3. Accommodation to sudden stretch. *a* shows the accommodation of tension up to the point when activity begins. The inset shows that at low stretch (1) tension is maintained by spontaneous activity which begins almost at once, but at high stretch (2) tension drops at first during the block of activity. The duration of this period increases with the degree of stretch. *b* and *c* show the time course of accommodation of spike frequency and membrane potential.

records, of which two examples are shown in the insert. The curves represent decline of tension up to the moment at which the first phasic contraction appeared, i.e. up to the moment when the muscle produced active tension. From that time onwards the tension was more or less maintained at the level at which spontaneous activity started.

The membrane potential took longer than the tension to become steady following a sudden stretch, and the time taken increased with the extent of the stretch (Fig. 3*c*). The spike frequency was increased. However, the peak was not reached at once but only in the course of the first minute (Fig. 3*b*). In contrast to the tension, the membrane potential and the spike frequency took from 40 sec to 5 min to become steady after sudden stretch.

Sudden release of the muscle

Figure 4 shows the changes of the membrane activity and tension following sudden release of the muscle after stretch. The muscle stretched to a $W:L$ ratio of 0·61 showed a high membrane activity (*a*). When the tension was suddenly lowered membrane activity was at first abolished, then

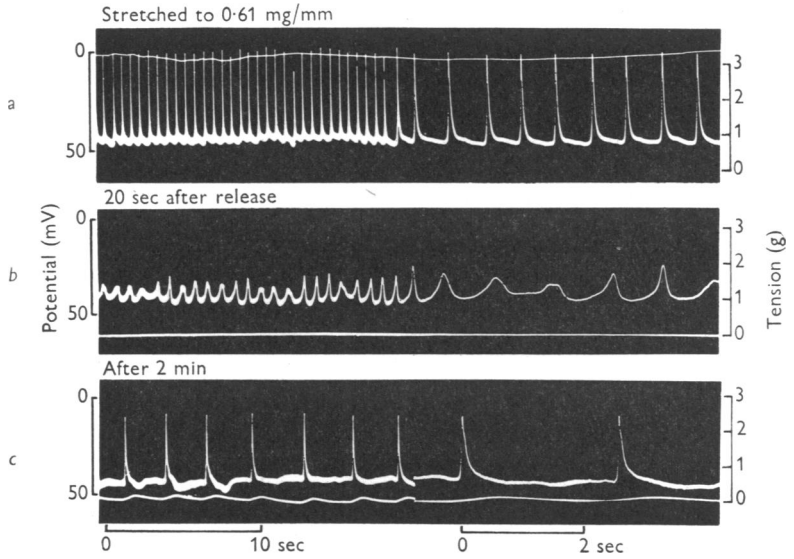

Fig. 4. Accommodation to sudden release. Records, taken at two different film speeds; *a* while the muscle was stretched to a $W:L$ of 0·61; *b*, 20 sec after sudden release; *c*, 2 min later. For description see text.

oscillatory local potentials appeared. Figure 4*b* shows the condition 20 sec after the sudden release. The local potentials gradually decreased in frequency and increased in height until they reached threshold and spikes were triggered. The membrane activity and tension 2 min after the sudden release are shown in Fig. 4*c*. Occasionally, during the first 30–60 sec after discharge began, the spikes were not accompanied by tension development, and sometimes tension was seen to develop without spikes in the impaled cell. These observations indicated a block of conduction.

When the muscle was released from $W:L$ 0·5 to $W:L$ 1·0, it failed at first to take up the slack and tension fell to zero. However, after 3–5 min spikes and tension reappeared. This is shown in Fig. 4. On the other hand, when the muscle was only released from 0·8 to 1·0, tension fell transiently and slightly, but spike discharge continued.

Effects of adrenaline on the membrane activity and tension at various degrees of stretch

In a previous paper (Bülbring & Kuriyama, 1963) it was shown that the effect of adrenaline depended on the membrane potential prevailing at the time of application. It was found that adrenaline caused hyperpolarization only if the initial membrane potential was lower than 65–70 mV, beyond which adrenaline only stopped spike discharge without producing a rise in membrane potential.

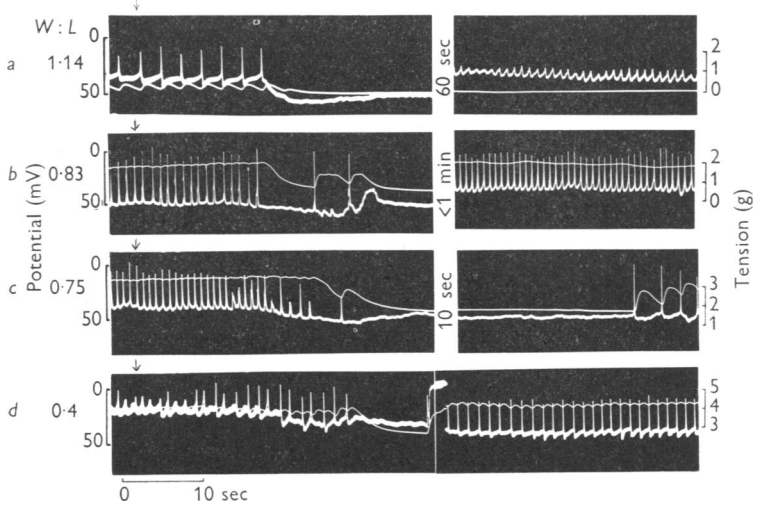

Fig. 5. The action of adrenaline 10^{-7}, applied at the arrow, at four different degrees of stretch. The magnitude and duration of the hyperpolarization increases with increasing stretch, while the duration of the block of spike discharge decreases and lasts only 9 sec in the severely stretched muscle.

In the present experiments the range of the initial membrane potential could be predetermined by a given $W:L$ ratio, and Fig. 5 illustrates the effect of adrenaline 10^{-7} in four different conditions. The activity in the unstretched preparation ($W:L$ = 1·14) was of typical pace-maker type. The membrane potential was relatively low and each spike was followed by a pronounced positive after-potential, the maximum polarization between spikes reaching 42 mV. Adrenaline increased the membrane potential to 55 mV. However, the hyperpolarization lasted only 30 sec.

In Fig. 5*b* and *c* ($W:L$ = 0·83 and 0·75) the hyperpolarization was 14 and 16 mV, respectively, but the micro-electrode was dislodged and the duration of the hyperpolarization could not be accurately measured, though in Fig. 5*c* it lasted at least 50 sec. In Fig. 5*d*, when the muscle was severely stretched, the hyperpolarization was 28 mV and it lasted for

70 sec (50 sec is shown in the record, and it lasted 20 sec longer.) The period, however, for which the spikes were stopped was only 9 sec, while in the unstretched preparation (Fig. 5*a*) oscillatory potentials appeared first and spikes which produced phasic tension responses returned only after 3 min. Sometimes phasic tension responses were also seen without spikes being observed in the impaled cell, indicating that for a considerable time after the application of adrenaline, the conduction in the muscle was impaired. In general the duration of the quiescent period caused by adrenaline was roughly proportional to the $W:L$ ratio of the stretch. In the unstretched tissue the silent period was longer than in highly stretched tissue. Though the micro-electrode was, of course, frequently dislodged, the silent period could be measured by observing the time for which phasic tension responses were absent. The mean duration of the block in three experiments was 110 sec for $W:L$ = 1·2–1·0, 65 sec for 1·0–0·8; 45 sec for 0·8–0·6; and 28 sec for 0·6–0·4.

TABLE 1. The effect of adrenaline at different degrees of stretch

	Adrenaline 10^{-8}			Adrenaline 10^{-7}		
$W:L$	Before (mV)	After (mV)	Hyper-polarization (mV)	Before (mV)	After (mV)	Hyper-polarization (mV)
1·3 –1·05	—	—	—	47·2 ± 0·84	55·8 ± 0·76	8·6 (n = 13)
1·15–0·95	47·2 ± 1·10	51·6 ± 1·21	4·4 (n = 12)	—	—	—
1·05–0·9	—	—	—	51·3 ± 0·94	60·7 ± 0·92	9·4 (n = 13)
0·9 –0·8	55·5 ± 0·99	62·6 ± 0·95	7·1 (n = 8)	55·7 ± 0·78	65·1 ± 0·71	9·4 (n = 12)
0·8 –0·65	52·6 ± 0·96	60·9 ± 0·95	8·3 (n = 8)	48·1 ± 1·43	60·9 ± 1·19	12·8 (n = 14)
0·65–0·4	43·8 ± 0·90	54·0 ± 1·00	10·2 (n = 8)	38·6 ± 2·30	53·7 ± 1·10	15·1 (n = 11)

Table 1 illustrates the effect of adrenaline on the membrane potential at different degrees of stretch. The results were obtained in sixteen different preparations. Though the membrane potential was highest in the range of $W:L$ 0·9–0·8, adrenaline caused the smallest hyperpolarization in the unstretched preparation ($W:L > 0·9$). When the muscle was stretched and the $W:L$ ratio decreased to less than 0·9, the hyperpolarization became progressively greater.

The effect of temperature on the action of adrenaline at varying degrees of stretch

Figure 6 shows the effect of different temperatures on the membrane activity. The muscle was stretched to a $W:L$ ratio of 0·75, and the temperature was varied from 22 to 39° C. The highest membrane potential and spike amplitude were recorded between 29 and 32° C. When the temperature was lowered to 29° C the membrane potential became stable and the spike amplitude was most regular. With further cooling to 25 and 22° C the membrane potential became unstable again and started to

fluctuate. Spike discharges were often blocked, leaving only the slow waves. When spikes occurred, they were preceded by a slow depolarization of the membrane, similar to that observed in the pacemaker of cardiac muscle. The spike frequency was linearly related to the temperature, the highest frequency occurring at 39° C. The critical temperature for cessation of spontaneous discharge was 20° C. The mean Q_{10} of the membrane potential between 22 and 32° C was 1·3. The mean Q_{10} of the spike parameters between 22 and 36° C was as follows: spike height 1·2; rate of rise 2·8; rate of fall 2·4; spike frequency 6·4. These values were measured in five different preparations.

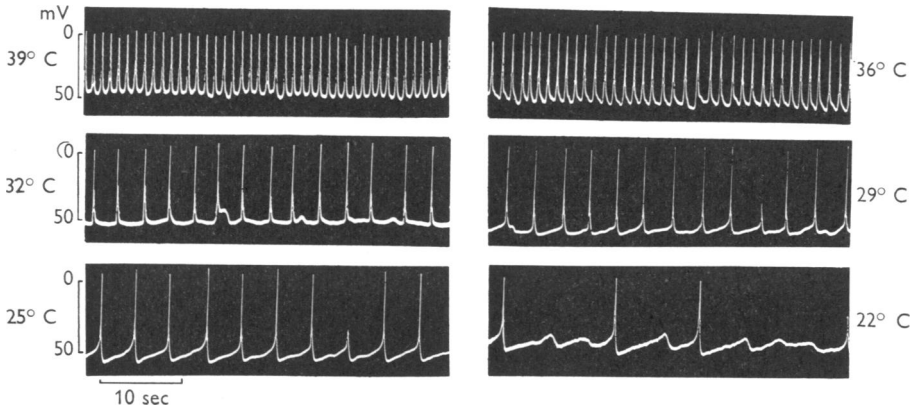

Fig. 6. Effect of temperature, recorded at $W:L = 0{\cdot}75$, on membrane potential and membrane activity.

From the above observations we selected three different temperatures for the study of adrenaline at various degrees of stretch. These were 36° C for the normal activity, 32° C for the condition of the maximum membrane potential and most regular spike activity, and 20° C for the critical temperature below which spontaneous spike generation ceases. Figure 7*a* illustrates the effect of adrenaline 10^{-7} on the membrane potential at two different temperatures (32° C is not shown because the adrenaline effect was closely similar to that at 36° C.) The hyperpolarization produced by adrenaline was less at 20° C than at 36° C at every degree of stretch. It always blocked spike generation but more time was required to produce this block at the lower temperature even in highly stretched preparations.

The tension which was observed during the period when activity was completely suppressed by adrenaline might be regarded as 'resting tension'. It was then possible to obtain some estimate of the 'twitch tension' in relation to the resting tension at different degrees of stretch. Figure 8 illustrates how the measurements were made with adrenaline 10^{-7} at 32° C, and in Fig. 7*b* similar observations at 20 and 36° C are plotted against

$W:L$ ratio. The graph shows the tension recorded during the silent period (R = resting tension), the tension produced by the first phasic 'twitch' after the silent period (A = active tension), and the sum of both (T = total tension).

It may be seen that the preparation developed active tension at lengths much less than that at which resting tension appeared. No resting tension

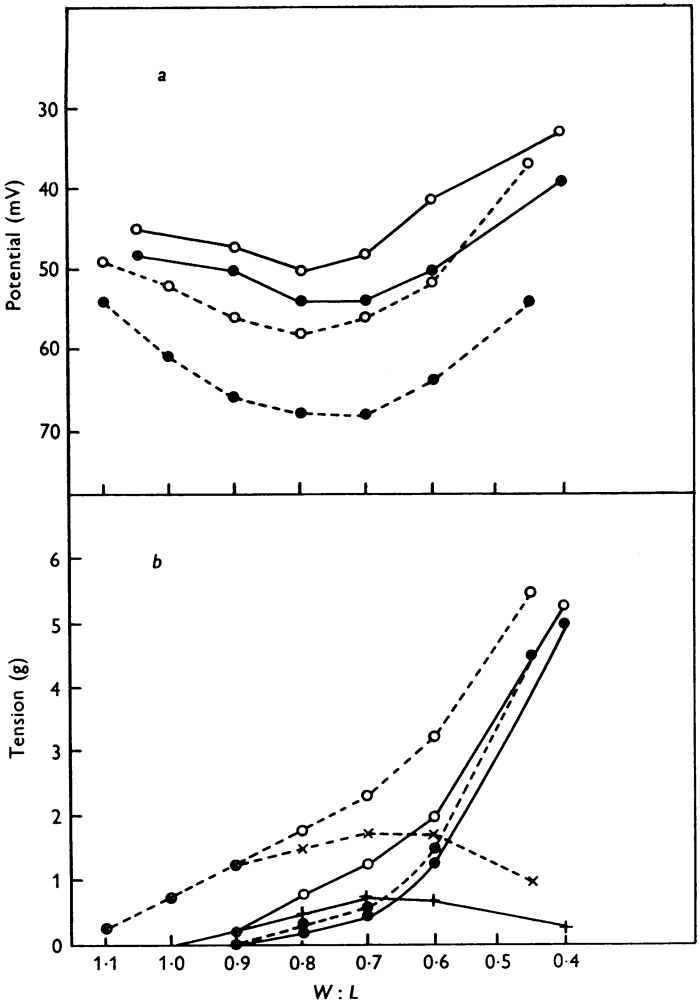

Fig. 7. *a* Effect of adrenaline 10^{-7} on the membrane potential at different temperatures (20 and 36° C) in relation to the degree of stretch ($W:L$). ○, membrane potentials before, ● in the presence of adrenaline. Continuous line at 20° C, interrupted line at 36° C. In *b* the resting tension is recorded during the silent period (●), the active tension is the height of the first phasic contraction following the silent period (×), and the total tension the sum of both (○).

was recorded until the preparation was stretched to a $W:L$ ratio = 0·8. The peak of the active tension was, however, not observed at this point but in the range of $W:L$ 0·7–0·6, and it was not sharply defined. The active twitch tension declined when the preparation was severely stretched.

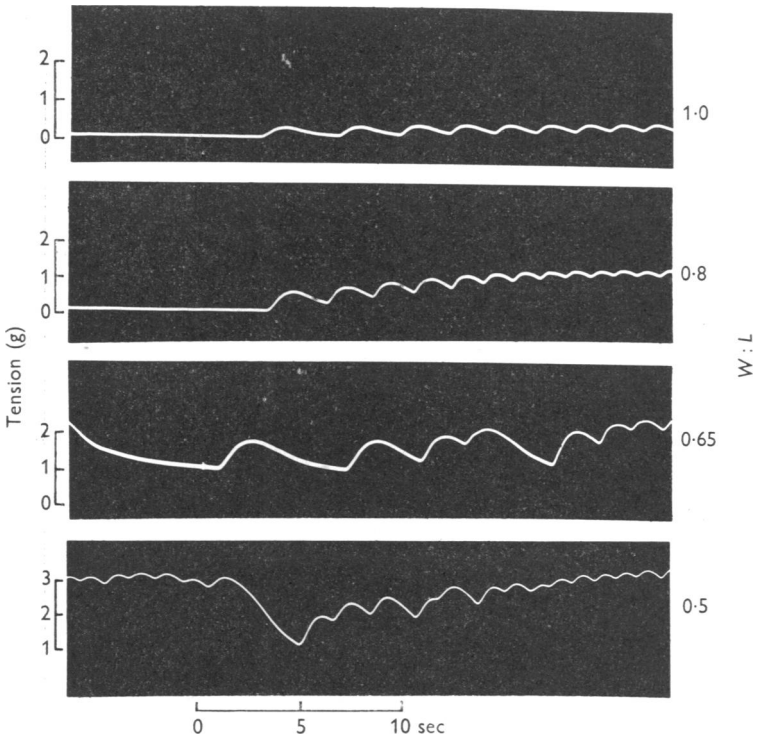

Fig. 8. Records of the first phasic tension responses following the relaxation produced by adrenaline 10^{-7} at different degrees of stretch and 32° C.

The resting tension–length curve was not greatly affected by temperature changes, indicating that the passive compliance of the tissue is not temperature-sensitive. However, the active tension was temperature-dependent. This may be due to an effect of temperature on the number of cells active (only spontaneous activity was measured) or on the tension developed by each active cell.

DISCUSSION

A study of the action of adrenaline on the taenia coli has been made in various conditions of stretch. These were defined by the $W:L$ ratio expressing the reduction in the muscle's cross-sectional area. It was found that the membrane potential varied with the degree of stretch. The

maximum was recorded at a $W:L$ ratio of about 0·9, and further stretch caused progressive depolarization.

In the completely loose, unstretched preparation the membrane potential was also lower. This may be partly due to difficulties in inserting the micropipette. But since most of the successfully impaled cells at $W:L > 1·0$ were found to be of pace-maker type this might explain the lower membrane potential.

When the muscle was stretched to $W:L$ less than 0·9 the membrane potential decreased in direct proportion to the $W:L$ ratio, i.e. the membrane potential was directly proportional to the cross-sectional area of the muscle, and it was within the same range for a given $W:L$ ratio in different preparations.

Burnstock & Prosser (1960) observed the effect of quick stretch with the sucrose gap and found that the membrane potential was increased in spontaneously active cells, but that it was decreased in quiescent cells. It may be that they worked in the range of $W:L$ ratio between $> 1·0$ and 0·9. It is not possible to repeat their experiments exactly with microelectrodes, since the quick stretch would dislodge them.

When the time course of adaptation to a fairly sudden stretch (taking 1–2 sec) was investigated it was found that the tension reached a steady state much more quickly than the membrane potential. The observations of the effects produced by adrenaline were therefore made when both were in a steady state, i.e. 10–15 min after the stretch.

With adrenaline it was possible to make some observations on the relation between resting and active tension. The behaviour of the taenia was different from that of skeletal muscle (Hill, 1951–2) and uterine muscle (Csapo & Goodall, 1954; Kuriyama, 1961), in that the peak of active tension did not occur at zero resting tension and was ill-defined. In this respect the taenia resembled the papillary muscle of the heart (Abbott & Mommaerts, 1959).

Stretch causes considerable deformation of smooth muscle cells, as shown by histological examination (Bülbring, 1957, Pl. 1; Freeman-Narrod & Goodford, 1962, Pl. 1). The cell deformation may be the primary reason for the depolarization, e.g. by increasing the sodium permeability of the cell membrane. According to the data of Harris (1953), there is an increase of the sodium efflux but not of the potassium efflux during stretch of skeletal muscle. Ishiko & Sato (1960) found that the membrane resistance of the frog sartorious is decreased by stretch. This has not been measured in smooth muscle. But it has been shown that stretch increases the rate of loss of ^{42}K (Born & Bülbring, 1956) as well as the rate of uptake, while the rate of loss of ^{24}Na is reduced in the unstretched preparation (Freeman-Narrod & Goodford, 1962). These authors found, however, that in the

steady state (30 min after stretch) the sodium and potassium content was unchanged. An increased sodium content in the cell due to increased sodium influx after stretch is bound to accelerate metabolic processes involved in sodium extrusion (Whittam, 1962), and thus the original ionic distribution is re-established. These mechanisms require a greater metabolic energy supply.

Bülbring (1953) observed that the tension increase produced by stretch was accompanied by a proportional activity increase in the rate of oxygen consumption. Also in skeletal muscle, Feng (1932) observed that stretch increased the metabolism two- or fourfold, and Eddy & Downs (1921) found an increased production of carbon dioxide. Recently Bueding, Bülbring, Gercken & Kuriyama (1963) observed that in taenia coli the content of the creatin phosphate and ATP was lower in muscle stretched for 1 hr to a $W:L$ ratio $< 0{\cdot}9$, than in unstretched muscle at a $W:L$ ratio $> 0{\cdot}9$. The energy-rich phosphate compounds may be partly utilized for active processes at the membrane, including sodium extrusion, but the stretched preparation appears to be unable to replenish the stores.

Adrenaline blocked the spike generation and increased the membrane potential in all conditions of stretch. The highest membrane potential reached in the presence of adrenaline was recorded at a $W:L$ ratio corresponding to the optimum condition of stretch ($W:L = 0{\cdot}9$–$0{\cdot}8$). In this range the maximum membrane potential, the maximum spike amplitude, the shortest spike duration, and a spike frequency of 0·6–1·0/sec are observed. However, the absolute magnitude of the hyperpolarization was the greater the more the muscle was stretched and smallest in the completely unstretched preparation. This relation was observed both with adrenaline 10^{-8} and 10^{-7}. The stronger concentration produced a greater effect at every $W:L$ ratio. The duration of the quiescent period caused by adrenaline was longer in unstretched preparations than in stretched ones. Furthermore, in the latter the hyperpolarization persisted for a long time after spike activity had restarted. Thus, when the muscle was depolarized and the spike amplitude had declined as a result of severe stretch, adrenaline raised the membrane potential to a level at which the initiation and the overshoot of the spike was restored. On the other hand, in the unstretched muscle, in which the threshold was already high, a small rise in membrane potential suppressed spontaneous discharge for a long period.

At low temperatures (20° C) the hyperpolarization of the membrane was smaller than at 36° C, even when the membrane was depolarized by stretch (cf. Axelsson, Bueding & Bülbring, 1961). This observation indicates a relation between the effect of adrenaline and tissue metabolism, and it has recently been shown (Bueding *et al.* 1963) that adrenaline produces a greater increase in the creatin phosphate and ATP content of

stretched preparations ($W:L < 0{\cdot}9$) than of unstretched preparations ($W:L > 0{\cdot}9$). It may well be that adrenaline replenishes the stores by stimulating the synthesis of energy-rich phosphate compounds and also stimulates their utilization for active processes involved in the hyperpolarization and stabilization of the membrane. It is interesting to note, in this connexion, that at low initial tension the inhibitory action of adrenaline may be associated with an increase in oxygen consumption (Bülbring, 1953).

SUMMARY

1. The effect of stretch on membrane potential, membrane activity and tension was observed in the smooth muscle of guinea-pig taenia coli. The degree of stretch was defined by the ratio (wet weight (mg)/length (mm)), the $W:L$ ratio, which corresponds to the cross-sectional area.

2. In the steady state the membrane potential, the amplitude and rate of rise of the spike were maximum at $W:L = 0{\cdot}9$. The membrane potential was lower in completely loose, unstretched tissue ($W:L > 0{\cdot}9$). It was much lower in stretched muscle ($W:L < 0{\cdot}9$), in which the membrane potential decreased in direct proportion to the decrease of the $W:L$ ratio.

3. After sudden stretch the adaptation of tension took 10–20 sec, but the membrane potential reached a steady state only in 5 min. Similarly, after sudden release, membrane activity and tension were completely abolished, and adaptation took up to 5 min.

4. Adrenaline 10^{-8} and 10^{-7} was applied during the steady state, 10–15 min after stretch. It stopped spike discharge and caused hyperpolarization at every degree of stretch.

5. The highest membrane potential in the presence of adrenaline was observed at $W:L = 0{\cdot}9$. The absolute increase of membrane potential produced by adrenaline was, however, directly proportional to the degree of stretch over the whole range of $W:L$ from 1·3 to 0·4, being greatest and of longest duration at the greatest stretch. On the other hand, the block of spike generation was more prolonged in unstretched preparations than in stretched ones.

6. The effect of temperature was investigated at the $W:L$ ratio of 0·75. The Q_{10} for the membrane potential was 1·3 (22–32° C), for the spike amplitude 1·2, rate of rise 2·8, rate of fall 2·4, and spike frequency 6·4 (22–36° C).

7. The hyperpolarization caused by adrenaline was much smaller at 20° C than at 36° C over the whole range of $W:L$ from 1·1 to 0·4.

8. The block of spontaneous activity by adrenaline was used to measure 'resting tension', and the first phasic tension response observed after the

silent period was taken to be a measure of 'active tension'. Active tension was not maximum at the length at which resting tension first appeared, and the maximum active tension was not sharply confined to a single length. The active tension–length curve was affected by temperature, but the resting curve was not affected.

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REFERENCES

ABBOTT, B. C. & MOMMAERTS, W. F. (1959). A study of inotropic mechanisms in the papillary muscle preparation. *J. gen. Physiol.* **42**, 533–51.

AXELSSON, J., BUEDING, E. & BÜLBRING, E. (1961). The inhibitory action of adrenaline on intestinal smooth muscle in relation to its action on phosphorylase activity. *J. Physiol.* **156**, 357–374.

BORN, G. V. R. & BÜLBRING, E. (1956). The movement of potassium between smooth muscle and the surrounding fluid. *J. Physiol.* **131**, 690–703.

BUEDING, E., BÜLBRING, E., GERCKEN, G. & KURIYAMA, H. (1963). The effect of adrenaline on the adenosine triphosphate (ATP) and creatine phosphate (CP) content of intestinal smooth muscle. *J. Physiol.* **166**, 8–9*P*.

BÜLBRING, E. (1953). Measurements of oxygen consumption in smooth muscle. *J. Physiol.* **122**, 111–134.

BÜLBRING, E. (1955). Correlation between membrane potential, spike discharge and tension in smooth muscle. *J. Physiol.* **128**, 200–221.

BÜLBRING, E. (1957). Physiology and pharmacology of intestinal smooth muscle. *Lectures on the Scientific Basis of Medicine*, **7**, 374–397.

BÜLBRING, E. & KURIYAMA, H. (1963). Effects of changes in ionic environment on the action of acetylcholine and adrenaline on the smooth muscle cells of guinea-pig taenia coli. *J. Physiol.* **166**, 59–74.

BURNSTOCK, G. (1958). The action of adrenaline on membrane potential and excitability in the taenia coli of the guinea-pig and the effect of DNP on this action and on the action of acetylcholine. *J. Physiol.* **143**, 183–194.

BURNSTOCK, G. & PROSSER, C. L. (1960). Responses of smooth muscles to quick stretch; relation of stretch to conduction. *Amer. J. Physiol.* **198**, 921–5.

CSAPO, A. & GOODALL, M. C. (1954). Excitability length–tension relation and kinetics of uterine contraction in relation to hormonal status. *J. Physiol.* **126**, 384–395.

EDDY, N. B. & DOWNS, A. W. (1921). Excitability of muscle. The production of carbon dioxide by a muscle when it is made to support a weight. *Amer. J. Physiol.* **56**, 188–195.

FENG, T. P. (1932). The effect of length on the resting metabolism of muscle. *J. Physiol.* **74**, 441–454.

FREEMAN-NARROD, M. & GOODFORD, P. J. (1962). Sodium and potassium content of the smooth muscle of the guinea-pig taenia coli at different temperatures and tensions. *J. Physiol.* **163**, 399–410.

HARRIS, E. J. (1953). The exchange of frog muscle potassium. *J. Physiol.* **120**, 246–253.

HILL, A. V. (1951–2). A discussion on the thermodynamics of elasticity in biological tissues under the leadership of A. V. Hill. *Proc. Roy. Soc.* B, **139**, 464–526.

ISHIKO, N. & SATO, M. (1960). The effect of stretch on the electrical constants of muscle fibre membrane. *Jap. J. Physiol.* **10**, 194–203.

KURIYAMA, H. (1961). The effect of progesterone and oxytocin on the mouse myometrium. *J. Physiol.* **159**, 26–39.

WHITTAM, R. (1962). The asymmetrical stimulation of a membrane adenosine triphosphatase in relation to active cation transport. *Biochem. J.* **84**, 110–118.