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THE RELATIVE UNIMPORTANCE OF THE TEMPORAL PATTERN OF THE PRIMARY AFFERENT INPUT IN DETERMINING THE MEAN LEVEL OF MOTOR FIRING IN THE TONIC VIBRATION REFLEX

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SUMMARY

1. A study has been made of the effect of varying the temporal arrangement of the mechanical stimuli used to elicit the tonic vibration reflex in the soleus muscle of the decerebrate cat. The reflex was elicited by brief mechanical pulses, applied repetitively, either as a regular series or, at the same mean frequency, in groups of 2, 3 or 4 pulses with a separation between the pulses of 3·5 msec. Mean frequencies of 140/sec and 100/sec were used. The amplitude of the pulses was such that it could be presumed that each pulse excited every Ia fibre from soleus to discharge a spike, irrespective of the patterning employed.

2. Alterations in the stimulus pattern produced only minimal alterations in the size of the resultant reflex recorded myographically. The grouped stimulation regularly tended to produce the larger effect, but even with groups of 4 at 100/sec the modal effect was only 10% of the pre-existing response; expressed another way this was equivalent to an increase of 11 Hz in the mean frequency of stimulation. Thus under these conditions grouping the stimuli cannot have had an appreciable effect either in increasing the firing frequency of those motoneurones which were already active, or in recruiting those which were initially quiescent.

3. Recording from individual motor units with fine electrodes placed on the surface of the muscle showed that they were not significantly changing their frequency of firing on altering the pattern of stimulation.

4. Gross electromyographic recording showed that the motor discharge was locked in time to the mechanical stimuli and of appropriate latency for it to be presumed that the actual discharge of impulses was triggered by Ia monosynaptic action.

5. Similar insensitivity to the temporal pattern of the afferent input was found when the motoneurones were excited via two separate channels, one

being the pulsed mechanical stimulation of soleus, the other being the weak electrical stimulation of the nerve to the medial head of gastrocnemius; altering the relative timing of the two sets of stimuli had little effect on the myographic result.

6. Thus, during tonic firing the timing of the motor output reflects the timing of the afferent input while the mean motor output reflects the mean value of the afferent input, as seems physiologically appropriate. The latter finding is, however, paradoxical; as detailed in an appendix the amount of motor firing produced by synchronous Ia monosynaptic action might be expected to increase on grouping the stimuli and so apparently favouring e.p.s.p. summation.

INTRODUCTION

Longitudinal vibration of the soleus muscle of the decerebrate cat at 100–200 Hz elicits a 'tonic vibration reflex' contraction of the muscle (Matthews, 1966). This appears to be no more than a facet of the well known stretch reflex, albeit elicited by a slightly unusual form of stretching, for vibration is known to be a powerful stimulant of the Ia fibres from the spindle primary endings. However, the Ia afferent input excited by vibration differs in two respects from that elicited by simple stretch. Firstly, the discharges of the various Ia fibres are highly synchronized when they are excited by vibration, whereas they fire largely randomly when excited by stretch. Secondly, vibration converts the normal irregular discharge of the primary ending under fusimotor bias to a completely regular stream of impulses. The present work investigates this latter effect by varying the pattern of the afferent input elicited by the vibration, although maintaining its synchronization. This was done by eliciting the tonic vibration reflex by a train of mechanical pulses, instead of the usual sinusoidal vibration, and then altering the spacing between the pulses. Thus a pulse train of constant mean frequency could be delivered as a regular series, or as pairs, triplets or quadruplets with each group being separated by an appropriately prolonged interval. Such changes in the pattern of afferent input have a surprisingly small effect on the mean tension developed in the reflex (cf. Matthews, 1974); as outlined in an Appendix grouping the stimuli would be expected to favour the summation of monosynaptic e.p.s.p.s and thereby elicit a greater overall motor discharge.

The study of neural patterning is also of interest in a wider sphere. The tension developed by certain crustacean muscle fibres has long been recognized to be dependent upon the pattern of motor discharge as well as upon its mean frequency, being greater for grouped than for regular stimulation (Ripley & Wiersma, 1953; Gillary & Kennedy, 1969), and this has been

suggested to be a principle of importance for integrative operations within the central nervous system. Bullock (1965), for example, has put the case in general terms while Porter & Muir (1971) have argued in the particular that the action of pyramidal tract neurones on the motoneurone is markedly dependent upon the pattern of impulses within a burst. On the other hand, the depressor effect on the arterial pressure of electrically stimulating the baroreceptor nerves is unchanged when the pattern of stimulation is altered while the mean frequency is maintained (Douglas, Ritchie & Schaumann, 1956).

METHODS

Preparation. The experiments were performed on the soleus muscle of the decerebrate cat. Nine were studied. They all gave tonic responses to vibration at 100 Hz, but with one exception they only showed weak or absent tonic responses to stretch applied on its own; additional preliminary observations were made on a further five cats while developing the methods. The animals were decerebrated by section between the colliculi while anaesthetized by Halothane; both common carotid arteries were ligated beforehand. The soleus tendon was connected to an isometric myograph mounted upon an electromagnetic stretcher. Except for the soleus the leg and hip were widely denervated. The hind limb was fixed by pins in both ends of the tibia; the femur was left intact. The soleus was covered by warm liquid paraffin. Further details have been given earlier (Matthews, 1966, 1969; McGrath & Matthews, 1973).

Recording. The output of the myograph was displayed on a cathode ray oscilloscope and photographed on moving paper throughout the experiment. In addition, it was also recorded along with various other signals on a 7-channel FM tape recorder (Philips, Analog-7) for subsequent analysis. Most observations were made with the soleus stretched to within a few mm of its maximum *in situ* length; typically it was stretched to this length while applying vibration (cf. Fig. 4). The electromyogram was usually recorded both with widely separated belly-tendon leads and also with one or two pairs of fine flexible wires (0·2 mm diameter, separated by about 0·5 mm) placed on the exposed surface of the muscle; the latter sometimes allowed the discharge of single motor units to be recognized in the course of a fairly powerful reflex contraction involving many units, while regularly providing a massed e.m.g. wave of much briefer duration than that obtained with the gross electrodes thus facilitating observations on motor timing.

The high-frequency response of the myograph was deliberately restricted by an electrical filter with a cut-off frequency of 37 Hz in order to minimize the small oscillatory changes in its output which occurred in synchrony with the mechanical pulses. This should not have interfered with the recording of the lower frequencies which are relevant for the study of the stretch reflex of such a slow muscle as the soleus. (A cut-off frequency of 37 Hz corresponds to a time constant of 4·3 msec so events lasting for only 10 msec would not be appreciably attenuated, while the rise time of the soleus twitch is 60–100 msec.)

Generation of brief mechanical pulses. The stretcher was induced to give a brief mechanical stretching pulse, of about 3·5 msec total duration (see Fig. 1), by feeding its control circuit with a triphasic pulse (cf. McGrath & Matthews, 1973). The first phase of the pulse caused a rapid acceleration in the stretching direction. The second phase of the pulse was of opposite polarity and caused the direction of movement of the stretcher to be rapidly reversed. The third phase of the pulse once more accelerated the moving element in the stretching direction so as to overcome its momentum

and thereby bring it to rest in the original position. By this means the stretcher could be made to respond far more rapidly than it did when it was given a single pulse in the stretching direction as a command for a brief stretch. The adjustment of the relative sizes and durations of the three phases of the triphasic control pulse was made by trial and error while observing the resulting movement. Such rapid movement unfortunately tended to be followed by a small oscillation, probably as a result of a mechanical resonance in the mounting of the transducers controlling the stretcher. But the amplitude of any oscillation was judged to be small enough for it to be ignored (cf. Fig. 1).

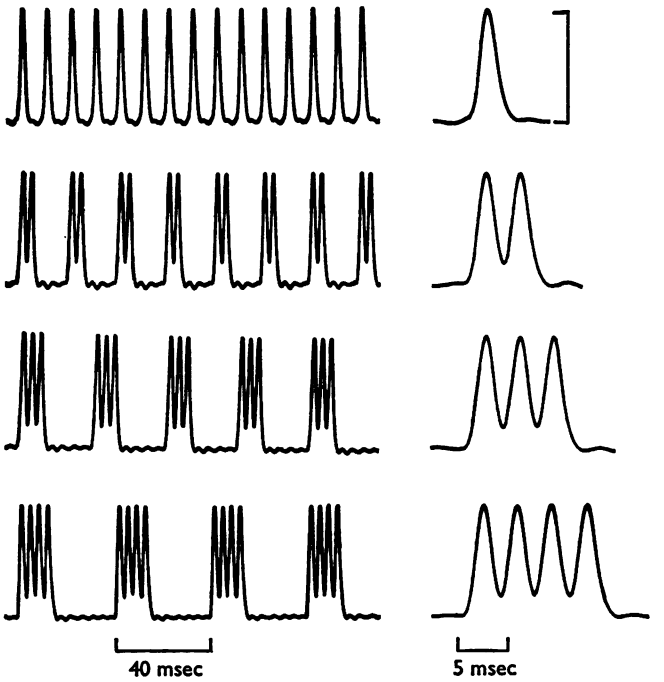

Fig. 1. Examples of the wave forms of the grouped mechanical stimuli. In each case the mean pulse frequency is 100/sec. Records taken with the length transducer incorporated in the electromagnetic muscle stretcher. The wobble on the base line, representing a movement of about 3 μm, is due to a slight resonance in the stretcher. Calibration bar, 70 μm.

Generation of patterns. Pulses at a constant mean frequency but of variable pattern were all initiated by a single master oscillator. When a regular stream of pulses was required the master oscillator itself provided the pulses. For separated pairs of pulses, the master pulses were fed to a divide-by-two circuit the output of which was used to trigger a pair of pulses; one occurred immediately and the other with a chosen delay. This ensured that the mean frequency of the pairs was identical with that of the master oscillator. Likewise, groups of three or four pulses at the required mean frequency were obtained by feeding the master pulses to divide-by-three or divide-by-four circuits which then triggered the appropriate number of pulses.

In seven of the nine experiments the pulse train supplied to the stretcher could be abruptly changed from the regular master series to one of the patterned trains (or vice versa) by means of a reed switch. This operated sufficiently rapidly for the switching to be completed between one pulse and the next so that there was no period of missed stimulation on changing the pattern in mid-stream. The reed switch was controlled by logic circuits so that, after giving the command to switch by applying an external voltage, the switching only occurred at the desired phase of the pattern. This guaranteed that the fine patterning at the time of transition remained constant from trial to trial. It also allowed a certain amount of choice as to what such patterning should be. For example, on switching from a regular series of pulses to pairs of pulses the gap between the last single pulse and the first pair could be made either a short one (corresponding to the normal interval between single pulses) or a long one (corresponding to the normal interval between the beginning of each pair). Such differences in the fine structure of the pattern at the time of switching produced no gross effect on the response and so will not be discussed further.

RESULTS

Patterns of stimuli

The four patterns of stimulation that have been studied are illustrated in Fig. 1; in all four the mean frequency is 100/sec. The total duration of an individual pulse is under 4 msec. Its rising phase lasts about 1·5 msec. On the basis of earlier recordings made from single afferent fibres (McGrath & Matthews, 1973; Brown, Engberg & Matthews, 1967) it may be provisionally concluded that when of appropriate amplitude each pulse excites a single spike in every soleus Ia fibre, irrespective of whether the pulses are delivered individually or in groups. The spacing of pulses within a group was regularly made 3·5 msec, which was the shortest practicable separation that did not produce an unacceptable degree of fusion of successive pulses; even so, the later members of a group are about 20% smaller than the first. A group of pulses corresponds approximately to a brief period of sinusoidal stretching delivered at 300 Hz, which is a frequency that Ia afferents follow without difficulty. The four patterns of stimulation illustrated in Fig. 1 would therefore all be expected to excite Ia firing at a mean frequency of 100/sec, but with very different temporal arrangements.

There are three ways in which the mean Ia firing might fail to be the same as that of the pulses. *Firstly*, a spike might fail to be excited by every pulse so that the firing frequency might fall below the pulse frequency. This is unlikely in view of the ease with which Ia fibres may be excited by sinusoidal movement and the finding that saw-tooth movements of comparable rising phases to the present pulses are appreciably more effective than sinusoidal movements (McGrath & Matthews, 1973). In addition, when the amplitude of the pulse was increased while observing the magnitude of the resulting reflex response a maximal sized reflex was produced for pulses of 20–30 μm upwards and then maintained constant to 100 μm, the largest available. This is most simply interpreted as due to 1:1 excitation of the Ia afferents by the stimulus (Matthews, 1966, 1969; McGrath & Matthews, 1973). *Secondly*, it might be suggested that the individual pulses sometimes excited a pair of spikes

instead of just one. But earlier findings (McGrath & Matthews, 1973) make it very unlikely that pulses with such brief rising phases would do so. *Thirdly*, and most seriously, it is important that the longest gap in the pattern should not be as great as the normal inter-spike interval found in the absence of pulsed stimulation. Otherwise the afferent will have time to fire a 'normal' impulse in between a burst of impulses elicited by a group of pulses. Any such effect would increase the mean frequency of afferent firing during the grouped stimulation in comparison with that

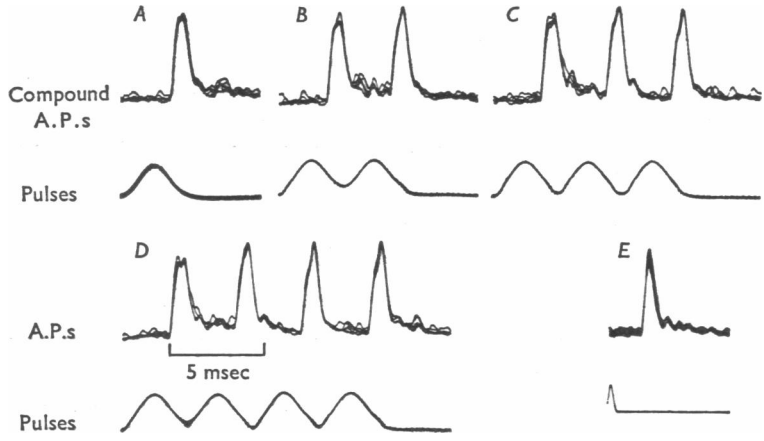

Fig. 2. The compound afferent action potentials (A.P.s) elicited by stimulating soleus with closely spaced mechanical pulses. Potentials above, stimuli below. Several traces superimposed. *A*, *B*, *C*, *D*, the number of pulses within a group is increased from 1 to 4 (mean frequency of stimuli 100/sec, amplitude 40 μm). *E*, the potential elicited by a single 100 μsec duration electrical stimulus to the intact soleus nerve (intensity 1·2 times maximal for group I, frequency 40/sec). Both ventral and dorsal roots cut, thus eliminating fusimotor and reflex activity. Records taken from the peripheral end of L7 dorsal root. The amplification for the electrically evoked action potential is half that for the mechanically evoked ones which are about 100 μV amplitude. Muscle stretched to approximately physiological maximum. (All records have been replayed from an FM tape recorder with a high frequency cut-off at 1·25 kHz for the particular speed employed here; accidental near saturation of one channel of the recorder has led to a slight distortion of the mechanical wave form.)

elicited by regular stimulation. However, this is unlikely to have occurred on any significant scale when the mean pulse frequency was 140/sec for even when the pulses were delivered in groups of 4 the maximum interval was still only 14 msec, corresponding to a frequency of 70/sec which would be a large response for a primary ending to static stretch (cf. Jansen & Matthews, 1962; Matthews & Stein, 1969). For the pairs or triplets of pulses, the risk would be correspondingly less. But the risk of such breakthrough of normal impulses complicates the use of low mean frequencies, and those below 100/sec have not been studied systematically (the interval between quadruplets is then 26 msec corresponding to a frequency of 39/sec).

Fig. 2 supports the view that each pulse did excite a constant Ia afferent volley irrespective of the particular arrangement of the grouping. The size of the major component of the compound action potential set up in the L7 dorsal root is approximately the same both within and between groups. Small differences in the height of different volleys appear to be largely compensated for by changes in the form of the wave so as to hold the area more nearly constant; in addition, the later waves may perhaps be superimposed on a small after potential. The afferent volleys are also seen to be remarkably well synchronized. The initial major component, which may be attributed largely if not entirely to Ia activity, lasts slightly under 1·5 msec. However, the potential elicited by electrical stimulation of the soleus nerve (*E*) was even briefer showing that some dispersion was introduced by the finite duration of the mechanical stimulus and the different locations of the various receptors within the muscle. (N.B. Since the nerve remained in continuity with the muscle, the tail of the potential in *E* will be attributable to Ia 'early discharges' as well as to group II activity.) Similar results were obtained in one other preparation on recording from the dorsal roots and from two further preparations on recording from the severed nerve to the soleus. Thus, changing the patterning of pulsed stimulation at a constant mean frequency may be concluded to produce large changes in the temporal patterning of the Ia input with little if any effect on its overall value averaged over the repeat period of the grouping.

Golgis and spindle secondaries. On the basis of previous work (Brown *et al.* 1967; Stuart, Willis & Reinking, 1971; McGrath & Matthews, 1973) the brief pulses would be expected to have relatively little action on both Golgi tendon organs and spindle secondary endings. The failure of the pulses to excite Ib fibres in the de-efferented state is supported by the observation in Fig. 2 that the size of the mechanically evoked compound action potential is below two thirds that of the electrically evoked potential (which is displayed on half the gain), showing that the pulses were exciting only a proportion of the group I fibres. However, in a reflexly contracting muscle the effect on the tendon organs might well be enhanced (cf. Brown *et al.* 1967). Some secondary endings may well have been weakly excited by the pulses since small delayed wavelets may be seen following the large initial wave in Fig. 2. However, even if such waves are attributable to group II activity they seem likely to be partly due to synchronization of the pre-existing spontaneous firing of secondary endings in time with the pulses and to be accompanied by only a moderate increase in their natural frequency of firing; this can be seen in single fibre records (cf. McGrath & Matthews, 1973). An extensive single unit study would be required to define the variety of patterns of firing of tendon organs and secondaries occurring for the various patterns of grouped stimulation. The two questions at issue are how far changing the grouping changes the mean frequency of firing of a unit, and how far the grouping influences its time of firing. A variety of answers might be expected for different endings. Change in the mean frequency of firing of an ending on changing the grouping would appear to be the most important action in relation to the present experiments. But in the absence of direct evidence it seems impossible to decide whether changing the pattern from, say, a regular series to groups of 4 would on the average

have the effect of increasing or of decreasing the total afferent inflow from the secondaries; arguments can be adduced for either form of behaviour on the part of single units. *Pro tem*, any such effects may be suggested to be of second order since the majority of secondary endings would appear to be rather insensitive to vibratory stimuli of the amplitudes currently employed. In contrast, altering the grouping clearly has the first order action of grossly transforming the temporal pattern of the Ia input without appreciably changing its mean level.

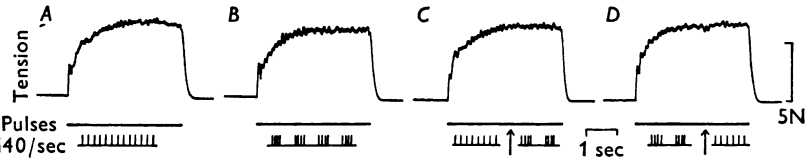

Fig. 3. Myographic records showing an immaterial effect on the magnitude of the tonic vibration reflex on altering the pattern of stimulation. In all records the reflex was elicited by brief pulses of 70 μm amplitude delivered at a mean frequency of 140/sec. In *A*, the pulses were delivered as a regular stream. In *B*, they were delivered in groups of 4 with an appropriately prolonged separation between groups. In *C* and *D*, the pattern of stimulation was reversed, as indicated, in the middle of the period of stimulation. If anything, in this experiment the grouped stimulation was slightly the less efficacious; however, the fact that *B* is just smaller than *A* would appear to be fortuitous since it is also smaller than the first half of *D*. Stimulus markers diagrammatic. Muscle initially well stretched.

Myographic reflex responses

Figs. 3*A* and *B* compare the reflex response elicited by a regular pulse train at 140/sec (*A*) with that elicited when the pulses were delivered in groups of 4 at the same mean frequency (*B*). The responses are virtually equivalent. The pulse amplitude in this as in other experiments was set at 70 μm since this value was found appropriate to elicit reliably a maximal reflex response, which may be taken to indicate the occurrence of 1:1 driving of the Ia afferents by the pulses (Matthews, 1966). The response to the pulses was then identical with the maximal response elicited by sinusoidal oscillation of the same frequency, although the latter requires a slightly higher amplitude of movement; recording from single afferents shows that this arises from the greater stimulating efficacy of the more rapidly rising wave form (McGrath & Matthews, 1973). *C* and *D* show the absence of effect on changing abruptly from one pattern to the other in the middle of a period of stimulation. In these examples, the action of the grouped stimulation is if anything slightly weaker than that of the regular stimulation, but the difference is less than the inevitable spontaneous variation in the size of the reflex and so cannot be taken as significant. In any case, the effect is atypical. In all other experiments, 140/sec stimulation tended to be slightly more efficacious when delivered in groups than when

delivered as a regular train, but often the effect could only be detected with difficulty if at all.

As might be expected, the effect of changing the pattern was more pronounced on stimulating at 100/sec. It could then virtually always be detected on altering the grouping from 1 to 4, quite often on changing from

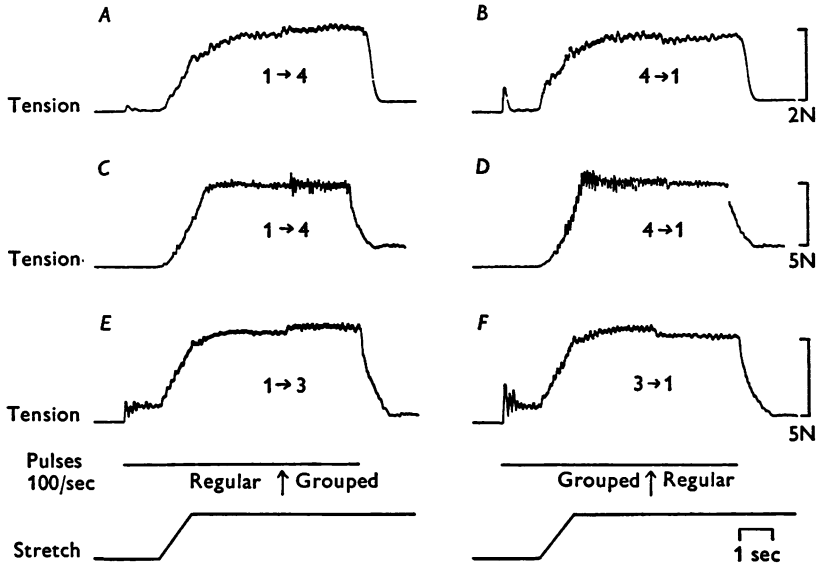

Fig. 4. Myographic records showing definite but slight effects on the tonic vibration reflex on altering the pattern of stimulation. The mean frequency of stimulation is lower than in Fig. 3 (100 Hz as compared with 140 Hz), so that a group of a given size is followed by a longer gap. The records are arranged in pairs (*A*, *B*; *C*, *D*; *E*, *F*) the members of which were obtained together. The pulsed stimulation was started while the muscle was slack. The muscle was then stretched by 5 mm at 5 mm/sec to make it taut; only then was an appreciable reflex developed. The magnitude of the response may best be judged by the fall in tension occurring at the end of each record on the cessation of the pulsed stimulation. Mechanical pulses, 70 μm amplitude. Diagrammatic stretch and stimulation markers apply approximately to all records. (*A*, *B* from experiment of Fig. 3 but taken at a time when the reflex was smaller. *C*, *D* another preparation; *E*, *F* second preparation again but taken 3 hr earlier when the reflex responsiveness was different.)

1 to 3, but only occasionally on changing from 1 to 2. Such effects are illustrated in Fig. 4. *A* and *B* show a definite albeit small effect in the experiment of Fig. 3. In *C* and *D* the effect is almost non-existent in spite of the frequency of stimulation being 100/sec. *E* and *F* show a definitely greater action of the grouped stimulation at a different time in the same experiment as *C* and *D*. But the effect is still a small one, the difference in tension

between the two modes of stimulation being only about 10 % of the overall response.

Thus, all in all, on stimulation at 100–140/sec the mean level of motor output, as assessed by the reflex tension, appears to depend upon the mean level of the afferent input irrespective of its precise fine structure over periods of up to 40 msec (the repeat cycle for groups of 4 at a mean frequency of 100/sec). In other words, the reflex centres treat small temporal variations in the afferent discharge as unwanted noise and successfully avoid responding to them as far as the average response is concerned. Nor can this behaviour be attributed to the sluggishness of muscle which, whatever the pattern of motor firing, would not allow the tension to be actively modulated at frequencies approaching those of the input pattern. All that is required of the muscle is that it should provide an indicator of the over-all level of motor discharge; it is difficult to envisage a more physiological measure of this than the isometric tension. Thus to a first approximation, the constancy of the reflex tension on varying the input pattern suggests that grouping the stimuli neither recruits fresh motoneurones nor increases the firing of those that are already activated by the regular input.

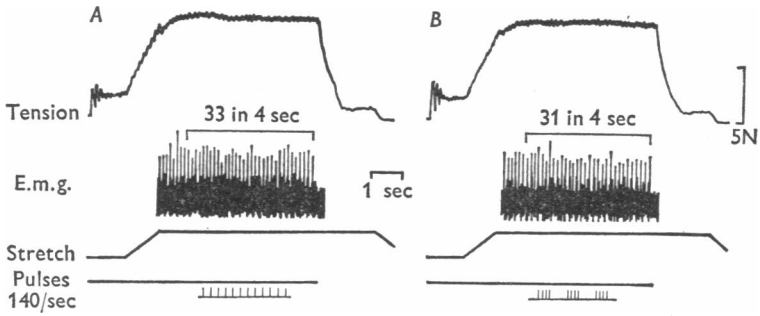

Fig. 5. The discharge of a single motor unit during pulsed stimulation. In this case the mean frequency of the unit was very slightly lower during stimulation with groups of 4 pulses (*B*) than during regularly spaced pulses (*A*) at the same mean frequency (140/sec). The unit was recorded from the surface of the soleus with fine bipolar electrodes. The e.m.g. records are on the same slow time scale as the myographic records and were obtained simultaneously. (The terminal portions of the e.m.g. records have been omitted.)

Because of spontaneous variations in the size of the reflex, any effects were always best demonstrated by changing the pattern of stimulation in mid-stream as in Fig. 4. The new equilibrium level then reached by the new pattern of stimulation did not seem to differ appreciably from that which was attained when the second pattern was applied throughout. There was, however, a suggestion that the change in tension was a little more marked when the change was from a grouped to a regular series rather than vice versa, but any effect was too small to be readily analysed with the

present methods. The time course of the change in tension was usually rapid, the new level being reached in under 0·3 sec. This contrasts with the slow build up of tension seen at the beginning of a period of vibration (cf. Fig. 3) and which is also seen on changing the frequency of vibration in the middle of a response (unpublished observations).

Firing of single motor units

It might be suggested that individual motoneurones were increasing their frequency of firing on using the groups, but failing to produce an increase in muscular tension because the motoneurones were already firing at the tetanic fusion frequency of the muscle fibres. This would not, of course, explain the failure of the grouped stimulation to recruit additional motor units. The possibility was further excluded by recording from single motor units by fine bipolar electrodes placed on the surface of the muscle. Fifteen units were observed in five preparations, usually with 140/sec stimulation. All fifteen units fired at a relatively low frequency in response to the regular stimulation (4–8 impulses/sec). These values are well below the expected fusion frequency of soleus, which varies from about 15–20/sec depending upon the precise length of the muscle (Cooper & Eccles, 1930; Matthews, 1959; Rack & Westbury, 1969) and parallel earlier observations obtained during excitation of soleus motor units by simple stretch of the muscle (Denny-Brown, 1929; Granit, 1958; Grillner & Udo, 1971). With such frequencies of firing small changes in the mean rate should produce relatively large changes in contractile tension, since they lie on the steepest part of the tension–frequency curve; for example, doubling the firing rate from 6 to 12 impulses/sec might be expected to more than double the resultant tension (cf. Rack & Westbury, 1969, Fig. 9).

With one exception, when grouped stimulation was used, none of the units fired at an appreciably higher frequency than with regular stimulation. Nor was any appreciable difference in mean firing frequency found when the pattern of stimulation was changed during the course of a single period of stimulation. The measurements were made for an integral number of spikes occupying a period of over a second, and agreed to within 10%. The exception was a unit which increased its firing from 6 to 7 impulses/sec on changing stimulation at 140/sec from a regular train to groups of 4. An example of the typical lack of effect is illustrated in Fig. 5, where the large spike could be recognized simply on the basis of its size, without the need for high speed display to permit observation of its precise wave form. It fired at just over 8/sec during the regular stimulation and at just under 8/sec during the grouped stimulation; such behaviour would not appear to be statistically significant. It may be concluded that on studying single motor units, as on studying the mean tension, there is little sign of an appreciably greater effect of grouped stimuli. This is not particularly surprising, since the firing frequencies of individual

motoneurones are known to be maintained at an approximately constant value on varying the extent of stretch or frequency of vibration (cf. Brown, Lawrence & Matthews, 1968; Anastasijević, Anojcić, Todorović & Vuco, 1968; Grillner & Udo, 1971).

Comparison between the magnitude of the effects of altering the grouping with that of altering the mean frequency

The size of the tonic vibration reflex increases approximately linearly with the frequency of vibration (cf. Matthews, 1966). For soleus, a typical value for the slope of the tension–frequency relation is 30 mN/Hz. Such increase in the reflex with frequency seems likely to depend largely upon the recruitment of fresh motoneurones rather than upon an increase in the frequency of firing of those that are already firing, though the matter has yet to be studied in detail. Either way, however, the value of this slope provides a reference for assessing any myographically observable effect of altering the grouping of a series of pulses at a constant mean frequency. Thus, for example, the increase in tension of some 10 % of the pre-existing value seen on grouping the stimuli in Fig. 4*A* is equivalent to an increase in the frequency of regular stimulation of 10 Hz, since in this preparation the slope of the tension–frequency relation was 26 mN/Hz and the increase in tension on grouping the pulses was approximately 260 mN. Such a comparison was made in all nine experiments on changing the grouping in the middle of a period of stimulation using mean frequencies both of 100 and of 140/sec. The tension–frequency curves were usually determined using sinusoidal stretching of 150 μm amplitude in the manner done earlier (Matthews, 1966, 1969), but control observations confirmed that this gave the same result as using a regular stream of pulses of 70 μm amplitude. The typical effect of grouping the stimuli in fours, expressed as the median value, was 7 Hz at 140 Hz and 11 Hz at 100/sec. In contrast, the reflex usually continues to increase approximately linearly with frequency for frequencies of up to 200 Hz or more, thus eliminating any suggestion that the small effect of the grouping arose from a 'saturation' of the reflex centres.

In eight of the nine experiments, the vibration reflex was well developed and the effect of changing the grouping was small. In these, with a mean frequency of 140 Hz, the reflex developed a tension of 4 N or over and, when detectable, the effect of increasing the group size to 2, 3 or 4 was 10 % or less of the existing value. In seven cases this corresponded to a frequency of 10 Hz or less; in the eighth the value was 15 Hz. With a mean frequency of 100 Hz the reflex developed a tension of 2·4 N or above and the effect of increasing the group size was 14 % or less of the existing value. This corresponded to a change in the mean frequency of 15 Hz or below. The effects of paired stimuli were always small, while the groups of 4 tended to be slightly more efficacious than the groups of 3.

In the ninth experiment the tonic vibration reflex was poorly developed (1·1 N at 100 Hz, slope 26 mN/Hz) and the effect of changing the grouping was large, both

in relation to the pre-existing tension and when expressed as the equivalent increase in mean frequency (at 140 Hz mean, changing the grouping from 1 to 4 gave a 21 % tension increase, corresponding to 17 Hz; and at 100 Hz gave a 70 % increase, corresponding to 30 Hz). The finding of a marked effect of grouping in the presence of a poorly developed initial reflex lends support to the idea that the effects are normally small because they are kept so by central regulatory mechanisms, which manifest themselves only in the presence of an appreciable level of activity.

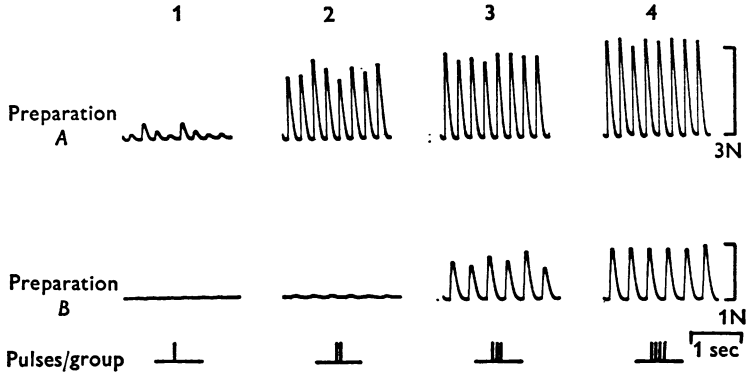

Fig. 6. 'Jerk' type myographic responses elicited by a series of well separated groups of pulses. Groups repeated at 4/sec in *A* and at 2·5/sec in *B*. The response increases with the number of pulses in the group thus indicating the occurrence of temporal summation. Results from two separate preparations. Pulse amplitude, 70 μm. Separation of pulses within a group 3·5 msec. Muscles stretched to close to physiological maximum.

Experimental indications of e.p.s.p. summation

As outlined in the appendix, the several e.p.s.p.s. elicited by a group of pulses might be expected to summate in their action. Evidence for the occurrence of a degree of summation between successive real e.p.s.p.s. in the present experiments was obtained by recording the reflex responses to isolated bursts of pulses of mechanical stimulation. As illustrated in Fig. 6, the size of the myographically recorded reflex response then increases with the number of pulses within a burst. In addition, a given number of pulses was more effective when they were delivered with a spacing of 3·5 msec, corresponding to that used in the grouped stimulation, than when they were separated by one of the longer intervals corresponding to the two frequencies of regular stimulation that have been used.

Fig. 6 illustrates results from two separate preparations because the summation effects appear to manifest themselves more readily by raising a subthreshold response to well above threshold rather than by augmenting an already large response. Possibly any appreciable motor firing evoked by the *n*th pulse of a group leads to a significant Renshaw inhibition of motoneurones that would otherwise have fired

to the summated e.p.s.p. elicited by pulse $n+1$ of a group. Recording of the twitch responses elicited by maximal stimulation of the nerve after sectioning it at the end of the experiment showed that any apparent saturation of the reflex was not attributable to the mobilization of all the soleus motoneurones. The maximal reflex was usually below half and always below two thirds the size of the maximal twitch.

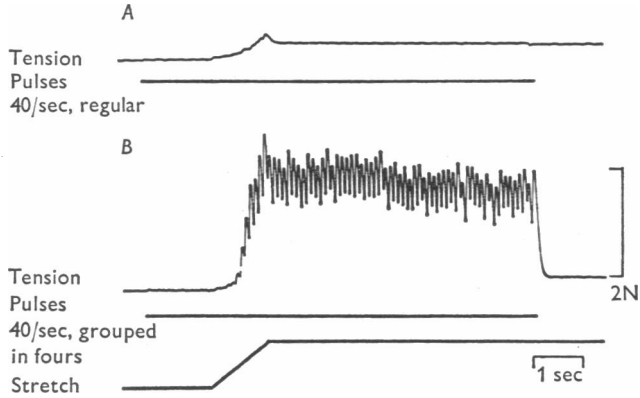

Fig. 7. The marked effect of grouping the stimuli at a constant mean frequency found when the frequency of stimulation is low and when it fails to elicit a response on being delivered as a regular stream of pulses. *A*, virtually absent response to regular stimulation at 40/sec (the just detectable shift in base line is largely due to a slight d.c. stretch applied along with the pulses). *B*, clonic response at 10/sec when the stimuli are delivered in groups of 4. Stimulation started while muscle slack; it was then stretched 5 mm. Pulse amplitude, 70 μm. Same experiment as in Fig. 3.

Fig. 7 illustrates another effect which further demonstrates the efficacy of summation of the activity elicited by the grouped stimulation. With the low mean frequency of 40/sec a regular series of pulses entirely failed to elicit contraction, whereas groups of 4 pulses at the same mean frequency successfully did so. It follows that there must have been summation of the e.p.s.p.s. elicited by the successive pulses within the groups, since single pulses applied at the repeat frequency of the groups were also without overt action. In addition, contrary to all that has been presented hitherto, this experiment shows that under appropriate conditions grouped stimulation may be far more efficacious than ungrouped stimulat ion. Two essential prerequisites for this to occur would seem to be first, that the regular stimulation should fail to elicit a significant response, and second that the frequency of stimulation should be low. However, although typical, the phenomenon has not yet been sufficiently analysed to be properly assessed. It certainly shows that there are indeed limits to the efficacy of action of the the central smoothing mechanisms, but this sort of experiment does not permit the limits to be accurately defined.

Two sorts of difficulties prevent an immediate solution. Firstly, it seems likely that with low pulse frequencies the mean frequency of Ia firing is greater during grouped than during regular stimulation. This is simply because the interval between groups becomes so long that 'spontaneous' impulses, evoked by static stretch, can be expected to break through in between those evoked by the pulses. When, for example, the mean pulse frequency is 40/sec, as above, then there is an interval of 86 msec between groups so that every ending firing spontaneously at above 12/sec would fire additional impulses (cf. an ending with a spontaneous rate of 40/sec would fire at 70/sec during the grouped stimulation but only at 40/sec during regular stimulation). Secondly, the failure of the regular stimulation to evoke a reflex removes from play any integrative mechanisms that are related to motor firing, a notable example of which is the Renshaw feed-back loop. Sporadic observations in the one preparation in which regular low frequency stimulation was seen to evoke a response suggested that the difference between the grouped and ungrouped stimulation was then less marked. Responses to low frequency stimulation would appear to occur in preparations with a significant tonic stretch reflex in response to stretch on its own. This has been uncommon in the present series of experiments so the matter has not been pursued as it deserves. The present study has been concentrated on frequencies of stimulation of 100–150/sec, for which the frequency of Ia afferent firing may be taken to be close to that of the pulses, and in preparations in which regular stimulation at these frequencies evoked an appreciable response (over 2 N). Thus, the thesis of this paper, namely the near equivalence of the effects of grouped and ungrouped stimulation, has only been established within these limits of frequency and of reflex action and should not be extrapolated beyond them.

Electromyograph recording

An intrinsic sensitivity of the motoneurones themselves to the pattern of their input might possibly be submerged in the present type of experiment if some Ia excitation were to reach the motoneurones indirectly via polysynaptic pathways as well as directly via the monosynaptic route. Whether or not this occurs continues to be a matter for debate (cf. Matthews, 1972; Homma & Kanda, 1973). Evidence arguing against a predominant dependence of the response upon polysynaptic pathways was obtained by recording electromyographically. This invariably showed that the motor output occurred in bursts locked in time to the mechanical pulses, rather than more or less continuously as might perhaps be expected if excitation were to be predominantly polysynaptic. This is illustrated in Fig. 8. Moreover, within each large burst of e.m.g. activity there would appear to be separate peaks of activity attributable to the individual mechanical pulses within a group. Limited observations on a few of the identifiable single units confirmed that every spike of a given unit occurred with a constant delay after a pulse, although its mean frequency of firing was far below that of the pulses.

Gaps in pulse train. A polysynaptic pathway might perhaps be expected to be more capable of eliciting motor firing when one or two pulses were simply omitted from an otherwise regular train, since the gap would not

then have been immediately preceded by a particularly large monosynaptically elicited e.p.s.p. as presumably occurs during grouped stimulation. However, as illustrated in Fig. 9, the omission of one or two pulses from trains at 100 and 140/sec simply led to missing periods of electromyographic discharge on all seven occasions on which it was tested. A sufficiently powerful polysynaptic pathway might have been expected to manifest itself by being able to continue to excite the motoneurone to

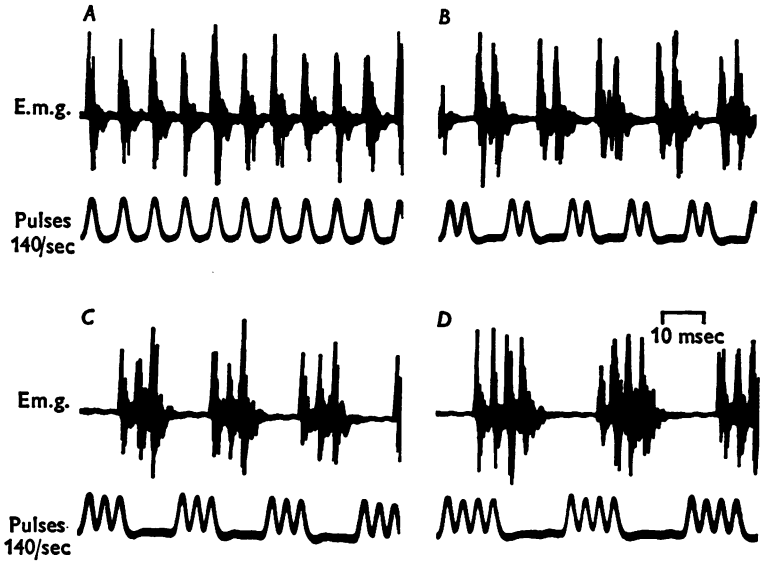

Fig. 8. The occurrence of a considerable degree of synchronization of the motor discharges demonstrated by electromyography. About thirty traces superimposed. Mean pulse frequency, 140/sec. The e.m.g. was recorded with fine closely-spaced bipolar electrodes thus minimizing the duration of the individual spikes and so facilitating the detection of synchronization. Same experiment as Fig. 5. Peak to peak amplitude of potentials about 1 mV.

discharge in the intervals between their monosynaptic excitation. These various e.m.g. findings, however, should not be taken to exclude the existence of a polysynaptic pathway since under related conditions others have found motor discharges which were not closely locked to the stimulus (Alvord & Fuortes, 1953; Tsukahara & Ohye, 1964; Homma & Kanda, 1973). Rather, they simply show the importance of the monosynaptic pathway for actually triggering off the motor firing in the present experiments in spite of whatever presynaptic inhibition the Ia terminals may be enduring, or whatever other influences may be impinging upon the motoneurones.

Latency of responses. The omission of pulses from a regular train allowed the latency of the electromyographic response to be determined during maintained stimulation, since this permits the allocation of a particular e.m.g. wave to a particular pulse. This showed latencies of 7–10 msec. Similar values were obtained on the basis of comparing the EMG responses to groups of various sizes when it was again possible to allocate individual responses to individual stimuli (cf. Fig. 8). In both cases, however, the first pulse of a group or following a gap sometimes failed to elicit firing, as judged by the appropriate prolongation of the latency of the first e.m.g. response. The values of 7–10 msec are very similar to the minimal values found at the beginning of a period of sinusoidal (Matthews, 1966) or of pulsed stimulation. In addition, they agree with those found on eliciting a reflex response with isolated pulses or groups of pulses as in Fig. 6. When a single pulse failed to produce firing it was readily possible by comparison of the various records to allocate the initial e.m.g. response to the individual pulse within a group that was responsible for eliciting it. In view of the appreciable time required for peripheral conduction these findings are all in accordance with the idea that the individual pulses elicit the triggering of the discharge of the motoneurones via the monosynaptic pathway, as has been assumed above, rather than via a polysynaptic pathway. But this conclusion rests primarily on the variety of evidence demonstrating the role of the monosynaptic pathway in the tendon jerk (cf. Matthews, 1972) rather than the exclusion by the present measurements of the existence of a disynaptic pathway (for which there is no evidence) or other very short latency central pathway.

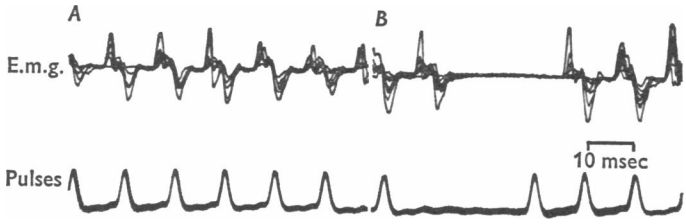

Fig. 9. The gap occurring in the electromyographic response on omitting two pulses from an otherwise regular train. 5–6 superimposed traces of the e.m.g. responses recorded with well-separated electrodes, which would be expected to pick up activity from a large proportion of the soleus. *A*, normal; *B*, with gap. *A* and *B* were taken during separate periods of stimulation because of the particular triggering arrangements employed. Mean frequency of pulses, 90/sec, of gaps, 2/sec. Peak to peak amplitude of potentials about 1 mV.

Combination of electrical and mechanical stimulation at different sites. A further situation in which a relative insensitivity to the temporal pattern of the input may be seen is when the soleus motoneurones are simultaneously excited via two separate channels. This was achieved by electrically stimulating the central end of the severed nerve to the medial head of gastrocnemius at the same time as applying mechanical pulses to the soleus. Weak repetitive electrical stimulation of the gastrocnemius nerve produces a tonic contraction of the soleus which may be presumed to be mediated

monosynaptically via the Ia fibres (Alvord & Fuortes, 1953; Matthews, 1959). The two sets of stimuli were delivered as regular trains at the same frequency, and were derived from a single master oscillator. They were either given 'in phase' with the electrical stimuli coinciding with the peak of the pulsed movements, or given 'out of phase' with the electrical stimuli set mid-way between the mechanical stimuli. The timing could be switched between the two arrangements without missing any stimuli.

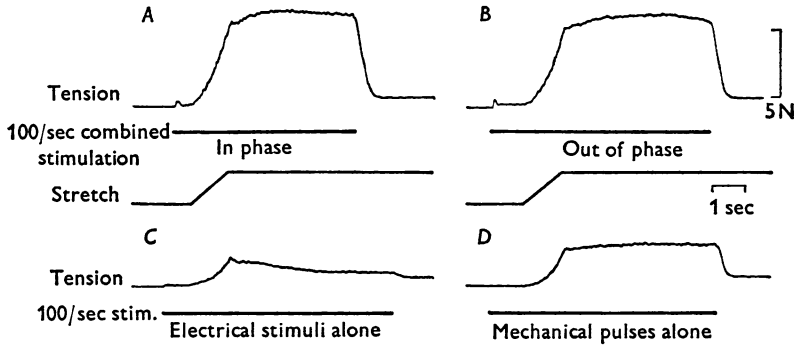

Fig. 10. The insensitivity of the overall reflex response to the relative timing of two separate sets of stimuli of moderately high frequency. Myographic responses of soleus to the combined stimuli of mechanical pulses applied to the soleus itself and of electrical stimuli applied to the nerve to the medial head of gastrocnemius, both at 100/sec. *A*, stimuli delivered in phase with each other. *B*, stimuli delivered out of phase, i.e. with a 5 msec separation between them. *C*, electrical stimulation on its own. *D*, mechanical stimulation alone. The stimulation was started while the muscle was slack. It was then stretched by 5 mm, as indicated, to make it taut. Mechanical pulses, 70 μm amplitude. Electrical stimuli 100 mV amplitude and 100 μsec duration which was just suprathreshold for eliciting a reflex response on its own; on the basis of other experiments this would be expected to be considerably submaximal for group I.

The strength of the electrical stimulus was set to produce the largest reflex possible. Further increases in strength diminished the response, presumably by recruiting inhibitory fibres more rapidly than excitatory ones. In three of the nine preparations the maximal reflex that could be elicited was too small to be worth studying. In two experiments recordings were taken from the nerve during its stimulation (recording electrodes placed peripherally to stimulating electrodes). This showed that the reflex was optimally elicited by stimuli which excited a compound action potential that was only about 20% maximal and so probably only a fraction of the Ia fibres were excited. (The maximal potential will have been derived both from group I afferents and α motor fibres.) Such recording also showed that it was not practicable to alter the patterning of the afferent input by altering the patterning of a train of purely electrical stimuli. When the stimuli were delivered in groups the refractoriness of the nerve prevented the successive stimuli within a group from exciting as many fibres as when they were delivered regularly, judged by the behaviour of the compound action potential.

A priori, synchronization of the electrical and mechanical stimuli might be expected to produce optimal summation of the resulting e.p.s.p.s. within the motoneurone and so elicit appreciably more excitation than when the two types of stimuli were delivered asynchronously. However, on testing the matter experimentally little difference was found between the effects of the two patterns of stimulation, as illustrated in Fig. 15 where the frequency of stimulation was 100/sec. Equivalent findings were obtained in the five further preparations in which such testing was performed.

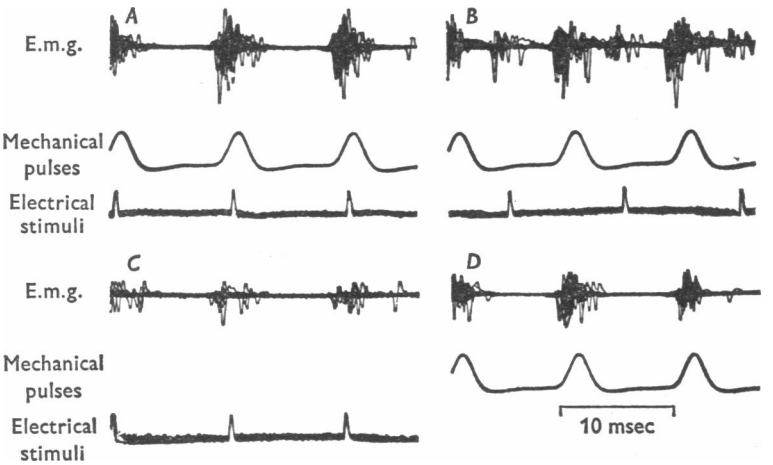

Fig. 11. The electromyographic responses of soleus to the combined stimuli of mechanical pulses applied to the soleus and of electrical stimuli applied to the nerve to the medial head of gastrocnemius. *A*, stimuli delivered in phase with each other. *B*, stimuli delivered out of phase. *C*, electrical stimulation on its own. *D*, mechanical stimulation alone. Frequency of each stimulus, 100/sec. E.m.g. recorded with fine bipolar electrodes; its peak to peak amplitude is approximately 2 mV. About thirty traces superimposed. Same experiment as Fig. 15. E.m.g. records taken during the approximate plateau of reflex tension with the muscle stretched.

It might be suggested that the phase of the nerve stimulation was immaterial because it produced its excitatory action solely by virtue of some non-specific facilitation, possibly via higher centres, rather than monosynaptically. This was largely excluded by electromyographic recording. When the nerve stimuli were delivered on their own then the e.m.g. potentials were phase-locked to the stimulus. When the two types of stimuli were delivered in combination, but out of phase, then the majority of the potentials were timed appropriately to be initiated by the mechanical stimuli (which was the more reflexly potent) but some were

attributable to the electrical stimuli. When the electrical and mechanical stimuli were synchronized then all the e.m.g. potentials occurred at the same time. These points are illustrated in Fig. 11. It should be noted, however, that on occasion the nerve stimulation seemed to produce its effect as much by facilitating the response to the pulses as by producing overt excitation on its own. Such an effect could only be established by quantitative study of the e.m.g., which has not been presently attempted. Fig. 11 also shows that the latency of response to the electrical stimulation is the same as that to the mechanical stimulation. This confirms that the two sets of stimuli are appropriately timed to give optimum summation when they are delivered simultaneously. This is to be expected since both effects may be presumed to be mediated by Ia monosynaptic action and since their afferent pathways are of very similar length.

With low frequencies of activation, however, very different results were obtained on combining the two types of stimuli, as illustrated in Fig. 12. At a frequency of 17/sec the two reflexes only significantly facilitated each other when the two sets of stimuli were delivered in near synchrony, and their synergistic action was virtually abolished by a separation of 6 msec either way. In contrast, at a frequency of 80/sec an equivalent absolute alteration in their relative timing had a negligible effect. Again, as on applying low frequency grouped stimulation, it would appear that the smoothing mechanisms can only operate in the presence of an appreciable level of neural activity. In the absence of significant reflex action under the least favourable set of conditions then the intrinsic summation properties of the motoneurones would appear to be given the opportunity to manifest themselves.

With higher frequencies, the findings on combined electrical and mechanical stimulation entirely parallel those obtained by altering the pattern of a train of purely mechanical stimuli. Both show that the mean motor output reflects the mean afferent input and is largely uninfluenced by small changes in its temporal arrangement, even though this can be shown to alter the time at which the motor spikes are actually discharged. The particular virtue of altering the relative timing of two separate trains of stimuli, delivered to separate structures, is that in this case there is little possibility that the average afferent input can change on altering the timing of the stimuli. Moreover, since the spacing of the volleys down each of the afferent channels remains constant, there should be no change in the quantity of transmitter released by each individual volley as might occur on changing the grouping within a single train of afferent volleys.

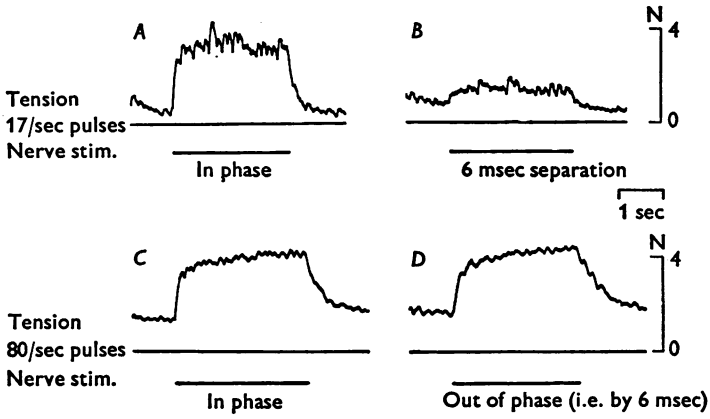

Fig. 12. The reduction of the reflex response found on altering the timing of two sets of stimuli that were being delivered at a low frequency. Myographic responses of the soleus to the combined stimuli of mechanical pulses applied to the soleus and of electrical stimuli applied to the nerve to the medial head of gastrocnemius, both at the same frequency. *A*, *B* frequency of stimulation 17/sec. *A*, stimuli delivered simultaneously. *B*, the nerve stimuli preceded the mechanical pulses by 6 msec – a similarly small response was obtained when the pulses preceded the nerve stimuli by 6 msec. *C*, *D*, frequency of stimulation 80/sec. *C*, stimuli in phase. *D*, stimuli out of phase, which at this frequency corresponds to a separation of 6 msec as in *B*. The records show the responses obtained with the muscle stretched and held at a constant length. The position of the calibration zero corresponds to the passive tension at the final length in the absence of all reflex contraction. The pulse stimulation was commenced before the stretching and the slight initial decay in tension at the beginning of the records follows the completion of the dynamic phase of stretching; this was usual with low frequency pulse stimulation. Other stimulation details as in Fig. 15, although that was from a different preparation.

DISCUSSION

The present findings contain an element of paradox. On the one hand, the electromyographic recordings confirm the expected importance of the monosynaptic reflex in producing overt firing of the motoneurones during their tonic excitation. Yet, on the other hand, altering the timing of the input so as to favour summation of monosynaptically elicited e.p.s.p.s. usually produced trivial effects on the total amount of firing from the soleus motoneurone pool. To a first approximation, during tonic firing the total motor output appears to depend upon the total Ia afferent input irrespective of its precise temporal arrangement. Whatever its mechanism this may be taken as a physiologically meaningful mode of response since it will smooth out moment to moment variations in the afferent input,

much of which will depend upon meaningless 'noise' resulting from fusimotor activity and the unfused contractions of extrafusal muscle fibres. Equally, the findings made it unlikely that as far as the tonic stretch reflex is concerned any biologically important information can be carried by the precise temporal pattern of the spindle afferent discharges evoked by stretch when the period under consideration is below about 40 msec.

There appear to be three broad types of explanation for the present observations. First, and least likely, the smoothing of the average level of motor output might arise from the behaviour of motoneurones acting individually. On current information, the Ia monosynaptic pathway would be expected to evoke a greater momentary depolarization of the motoneurone with the grouped stimulation. This is detailed in the Appendix which outlines the response of a 'model motoneurone' to monosynaptic excitation by the particular afferent patterns that were used to elicit the reflex. With standard values for its various parameters, grouping the stimuli led to appreciably greater summations of its e.p.s.p.s with a greater peak value of depolarization. If the model is correct in this respect it becomes necessary to explain why the grouping fails both to recruit fresh motoneurones and to increase the firing in the active ones, since either would increase the myographically recorded tension; explanations of the latter in terms of refractoriness are not applicable to the former. However, the motoneurone is sufficiently complex an entity to leave it undecided whether the model is inadequate in itself, rather than for avowedly neglecting all synaptic inputs other than the Ia monosynaptic connexions.

The second possibility is that there exists a significant polysynaptic input from the Ia fibres to their own motoneurones which is little affected by the patterning of the Ia input. This could smooth the main excitatory drive to the motoneurones even though Ia monosynaptic action provides, on the evidence of the phased electromyograph activity, the essential additional stimulus which actually triggers off the discharge of a spike. The present type of evidence does not permit such matters to be resolved.

The third, and more interesting possibility is that the response of the system is held constant in the face of the grouped inputs by some kind of negative feed-back acting in opposition to a greater sensitivity of the motoneurones acting individually. The immediate candidate for such an action is the inhibitory pathway from the recurrent motor collaterals to the Renshaw cells and back to the motoneurones. The Renshaw cells are known to fire tonically in the decerebrate, to be excited by vibration, and to grade their firing with changes in the level of afferent excitation (Pompeiano, Wand & Sontag, 1974*a*, *b*). It therefore seems almost inevitable that they must be playing some part in modulating the motor responses in the present experiments. For example, if they were more

sensitive to grouped than to regular inputs then any such increase in firing on their part would counteract that of the motoneurones. Equally, the interneurones mediating presynaptic inhibition, perhaps activated by the Ia fibres themselves, might be affected by the pattern of the input. Alternatively, inhibitory processes acting appropriately slowly may help to provide the 'integrator' which averages the level of neural activity over an appreciable period of time and so prevents momentary excesses of excitatory synaptic action leading to repeated increases in motor output. Again, the Renshaw cells would appear suited to play such a role, although certain other difficulties might then arise. A particular virtue of all explanations based upon inhibitory feed-back is that they provide a means whereby all the relevant motoneurones can be influenced, irrespective of whether or not they are excited to discharge by the regular inputs, thus helping to explain why appreciable numbers of additional motoneurones fail to be recruited by the grouped stimulation.

The Golgi tendon organs might also be suggested to be providing a significant negative feed-back since in the present experiments they will have been excited to discharge by the reflexly elicited contraction. Under such circumstances they may perhaps have been sensitized to the pulsatile stimulation so that they fired more rapidly than they would have done to the level of tension *per se*, thus offering the possibility that their mean level of discharge was different with the grouped and with the regular stimulation. More interestingly, however, it seems likely that their natural firing in response to the contractile tension may well have been partly synchronized in phase with the pulses, thus offering scope for postulating a variety of interactions between Ia and Ib inputs when the pattern was altered. However, these seem unlikely to be important in the present context since the mean frequency of firing of presumed soleus motor fibres, isolated from ventral root filaments after severing the ventral roots, shows no change on altering the input pattern (P. Dale & P. B. C. Matthews, unpublished); in the absence of muscle contraction the Golgi tendon organs may be presumed to have been largely silent and unaffected by the pulsed stimulation (cf. Brown *et al.* 1967).

Likewise, it seems improbable that any change in the firing pattern of the spindle secondary endings on changing the grouping of the stimuli can be held responsible for concealing an otherwise large reflex action of changing the temporal pattern of Ia firing. To achieve this, the secondary action would have to be remarkably finely balanced so as to produce the consistent reduction of a large effect to a trivial one, without ever by accident causing the response to the grouped stimuli to be grossly smaller than that to the regular stimuli. Moreover, any such explanation could not be readily applied to the absence of effect on changing the relative timing of separate trains of stimuli to the gastrocnemius nerve and to the soleus muscle.

Further investigation would appear justified of what might tend to be dismissed as simply an amusing paradox, dependent upon an unphysiological mode of stimulation. The effect seems likely to reflect upon normal integrative properties of the motoneurone pool and reflex centres, yet ones that have not hitherto been highlighted by the study of the responses of anaesthetized motoneurones to isolated volleys. Of course, none of the

present findings have indicated whether or not the central smoothing action is attributable to a single neuronal mechanism. Finally, it should be noted that the present experiments have shown that the magnitude of the tonic vibration reflex is not dependent upon the unwonted regularity of its afferent input, thus lending support to the practice of making quantitative comparisons between it and the normal tonic stretch reflex evoked by simple stretching.

I should like to thank Mr J. D. Mittell for general assistance and for building the various pulse generating circuits.

APPENDIX

The summation of simulated monosynaptic e.p.s.p.s

The motoneurone itself, by virtue of its membrane properties, must tend to smooth the afferent input. Its level of depolarization at any instant is due to the temporally summed effects of several successive volleys and thus reflects the mean level of afferent firing as well as the effects of the most recently arrived impulses. The question thus arises whether simple summation of monosynaptic e.p.s.p.s would suffice to explain the behaviour of the reflex. Various observations in the literature suggest that it would not (Curtis & Eccles, 1960; Westbury, 1972; Homma & Kanda, 1973), but it seemed desirable to test the matter further by observing the degree of summation occurring in a 'model motoneurone' of appropriate time constant activated by the present patterns of input. As the first approximation the model consisted of a single R-C element, corresponding to a simple globular neurone without dendrites. Fig. 13 shows, as expected, that the peak value of its depolarization increases on grouping the input at a constant mean frequency. The duration of the current pulse was made 1·5 msec thus producing an e.p.s.p. with a rising phase slightly longer than that observed on exciting motoneurones by a synchronous Ia volley (Eccles, Eccles & Lundberg, 1957) so as to allow for the slightly greater lack of synchrony of the mechanically elicited volley. The time constant was allocated a value of 5 msec in accordance with recent measurements on both fast and slow motoneurones (Burke, 1968*a*, *b*; Jack, Miller, Porter & Redman, 1971; Iansek & Redman, 1973*a*). The motoneurone is normally held to discharge an impulse whenever the depolarization reaches a critical level, so in life behaviour like that of Fig. 13 on grouping the stimuli would be expected to recruit fresh motoneurones to fire and to increase the firing of those that were already active. The latter effect could well be small, since various mechanisms act to stabilize firing at a low constant value (Granit, 1972).

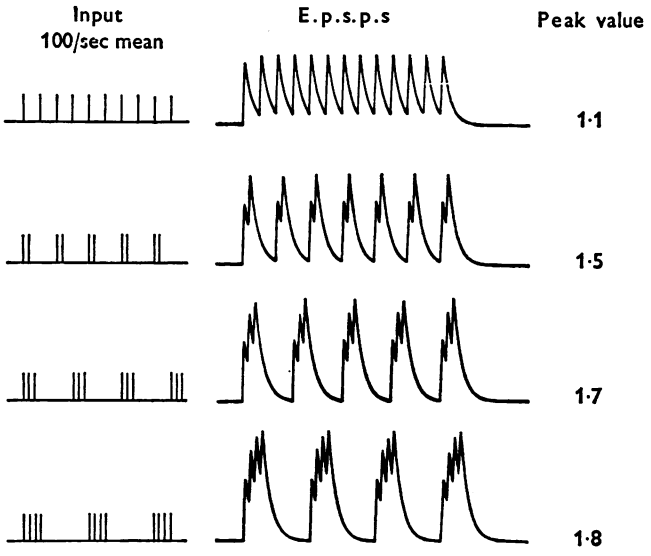

Fig. 13. The effect of grouping the stimuli in increasing the magnitude of the summed e.p.s.p. elicited in a model motoneurone. Membrane time constant, 5 msec. Pulse duration 1·5 msec. Pulse separation 3·5 msec.

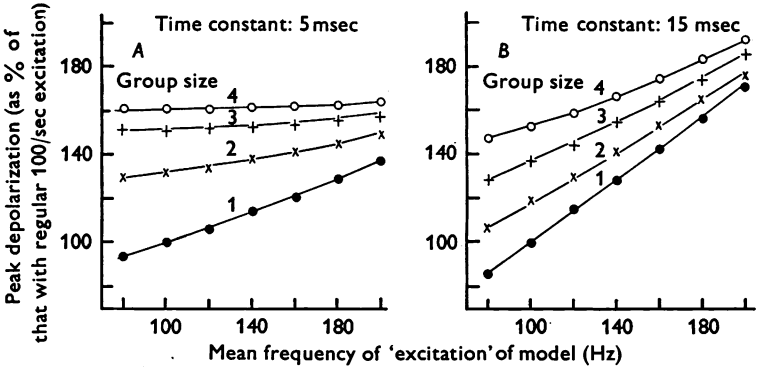

Fig. 14. The peak value of the depolarization elicited in the model motoneurone by various mean frequencies of excitation with different groupings of the stimuli. The time constant of the neurone was made 5 msec for *A* and 15 msec for *B*. The depolarization is expressed as a percentage of that elicited by excitation at 100/sec with regularly spaced pulses. In *A*, 100% represents a depolarization of 1·16 times the size of the unit e.p.s.p.; in *B*, the value is 2·04. The interval between the pulses within a group is 3·5 msec. The plotted values have been computed by summing the relevant exponential terms rather than by direct measurement on the model.

Fig. 14*A* compares the effect in the model of grouping the stimuli with that of increasing the frequency of a regular train. In contrast with the behaviour of the reflex the effect of grouping is very large. For example, at a mean frequency of 100 Hz grouping the stimuli merely in pairs is equivalent to increasing the frequency of the regular train by over 80 Hz. The effect for groups of three and four pulses is even greater. Likewise, the effects at a mean frequency of 140/sec are also markedly greater than those found physiologically. Nor were these findings dependent upon any slight error in the choice of time constant. When the value is increased threefold (Fig. 14*B*) the model still shows much larger effects than the reflex.

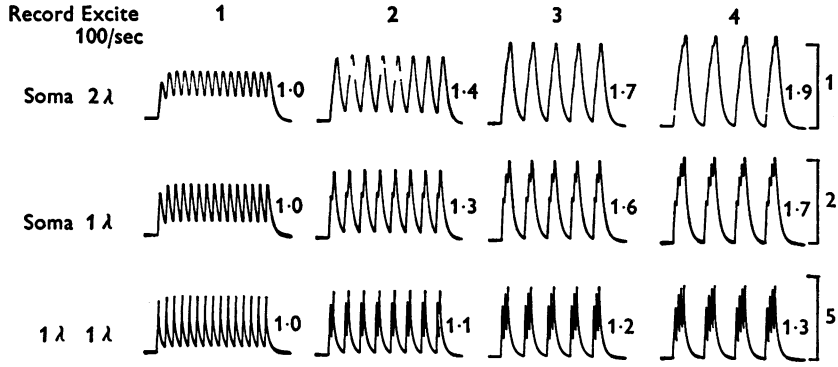

Fig. 15. 'E.p.s.p.s' elicited in a model neurone with dendrites. The model consisted of a cable two length constants (λ) long, composed of a series of R-C elements. Constant current pulses at a mean frequency of 100/sec were injected at one point and the resulting potentials recorded at various points as noted on the left. With each arrangement the stimuli were delivered both regularly and in groups of 2, 3 and 4 pulses. The figures to the right of each record show the peak value of its depolarization relative to that found with the equivalent regular input. The calibration bars to the far right indicate the relative sizes of each set of recordings. Membrane time constant, 6 msec. Pulse duration 1·5 msec. Pulse separation 3·5 msec. Ends of cable open circuit. (The cable contained ten elements each consisting of a 'membrane', with a resistor of 27 kΩ and a capacitor of 0·22 μF in parallel, connected to similar elements by a resistor of 1·2 kΩ. An identical element was used for the soma.)

So far, however, the model lacks dendrites. In the real motoneurone the Ia fibres terminate at up to about one 'space constant' (λ) away from the soma and the e.p.s.p. induced in the soma by the most distant fibres is rather slower than that elicited by nearby fibres (Rall, Burke, Smith, Nelson & Frank, 1967; Burke, 1967; Jack *et al.* 1971). It seems possible that such slowing of the e.p.s.p., by smoothing out and blurring any fine pattern in

the input might cause the depolarization of the soma to be very much the same irrespective of the temporal patterning of the Ia input. This possibility was explored by constructing a cable type network from a series of R-C elements and injecting current pulses at various points along its length. Fig. 15 shows the simulated e.p.s.p.s recorded from the 'soma', and in one case from the 'dendrite', on injecting current at one or two space constants out along the 'dendrite'. This corresponds to the simultaneous activation of equivalent points on every dendrite of a neurone, as seems appropriate in mimicking an input which is approximately synchronous in a large number of different Ia fibres. On comparing the records in vertical columns it can be seen, as expected, that the smoothing of the e.p.s.p.s increases with the separation between the sites of stimulation and recording. However, on comparing the horizontal rows it is apparent that the smoothing was not associated with obliteration of the greater depolarizing action of the grouped stimulation. Indeed, the least effect of grouping occurs at the site of 'synaptic' action; this is because rapid lateral spread of charge makes the local e.p.s.p. decay more rapidly than a simple exponential with the time constant of the membrane.

In the real motoneurone, the majority of Ia synapses are fairly evenly distributed along the proximal dendrites (Jack *et al.* 1971) and the more peripheral a synapse the greater its synaptic efficacy (Iansek & Redman, 1973*b*). Thus, on synchronous activation of all the homonymous Ia fibres the effect of charge redistribution should be relatively moderate thereby making the behaviour of the real motoneurone closer to that of the simple R-C model than to that of the cable model with restricted input. Indeed, Ia action is sufficiently widespread to make the time constant of decay of the maximal Ia e.p.s.p. itself rather close to the time constant of a unit area of membrane. This all makes it reasonably appropriate to mimic transmitter action by injecting a constant charge for each input volley irrespective of the prevailing level of depolarization, rather than by attempting to introduce a brief conductance change. Moreover, for Ia synapses transmitter release appears to be relatively unaffected by volley spacing (cf. Curtis & Eccles, 1960; Phillips & Porter, 1964). It should finally be noted that the model derives largely from the study of quiescent motoneurones in the anaesthetized spinal cord. In a reflexly active cord individual motoneurones should if anything tend to have a shorter time constant than that presently employed.

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