

# Fatigue in mammalian skeletal muscle stimulated under computer control

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**Wise, A. K., D. L. Morgan, J. E. Gregory, and U. Proske.** Fatigue in mammalian skeletal muscle stimulated under computer control. *J Appl Physiol* 90: 189–197, 2001.—Functional electrical stimulation (FES) is used to provide paralyzed human subjects with postural support and a limited range of movements. Problems encountered with FES include jerky movements from tension oscillations during stimulation and rapid muscle fatigue. In this paper, we report experiments on anesthetized cats that test a new, computer-controlled method of stimulation of the muscle nerve supply, distributed across several inputs, which reduces these problems. After 5 min of continuous, distributed stimulation of the medial gastrocnemius muscle at 6 pulses per second (pps) across 6 channels, tension fell to  $55.9 \pm 3.9\%$  (SE) of its original value. In comparison, after 5 min of synchronous stimulation of one muscle portion at 36 pps, tension fell to  $11 \pm 3.7\%$ . At higher stimulation rates, 10 pps per channel (distributed) and 60 pps (synchronous), the differences in fatigue were even greater. Similar results were obtained when an intermittent, rather than a continuous, stimulation protocol was used. These findings indicate that distributed stimulation has important advantages over other methods for applications such as FES.

distributed stimulation; contraction; tension; rehabilitation

MEASUREMENTS OF MUSCLE FATIGUE have long been of interest to many different groups, including exercise physiologists and sports physicians. Aspects that are commonly considered are the energetic factors contributing to fatigue and the various sites in the motor pathway at which fatigue may occur. Thus, during prolonged exercise, fatigue may be variously attributed to central nervous factors (8), neuromuscular failure (7), failure of action potential propagation (13), and changes in sarcoplasmic release of  $\text{Ca}^{2+}$ , as well as a reduction in force generation by cross bridges (22).

Higher muscle force levels are reached with high rates of stimulation than with lower rates, but the onset of fatigue is more rapid (13). Previous studies have shown that smooth contractions can be achieved with low rates of stimulation by using sequential, distributed stimulation of the muscle (5, 18). With this method, rather than stimulating all motoneurons to the muscle synchronously, the nerve supply is sepa-

rated into several portions, each generating a similar tension, and they are stimulated sequentially (see Fig. 1). With low rates of distributed stimulation, rates at which synchronous stimulation produce unfused contractions, the amount of tension oscillation during each contraction is significantly reduced because different portions of the muscle are contracting at different times. The peak tension in one part will coincide with the minimum in another, tending to smooth the tension profile. In addition, internal motion in muscle and tendon is reduced, prolonging each contraction and further smoothing the tension profile. The effective stimulation rate of the muscle, in terms of the smoothness of the tension profile, becomes the product of the stimulus rate and the number of channels stimulated. From a functional point of view, distributed stimulation is more appealing than synchronous stimulation because it more closely resembles the natural pattern of muscle activation by the central nervous system.

In the past, distributed stimulation used three or more stimulus channels, with equal intervals between channels. This meant that, if the tension generated by one channel was significantly greater than the others, then, during repetitive distributed stimulation, the tension profile would not be smooth but would oscillate at the rate of stimulation. This limitation meant that distributed stimulation required close matching of force levels between channels and was used only with slow, fatigue-resistant muscles. In fast, fatiguable muscles or in mixed, fast-slow muscles, stimulus channels may be arranged so that each channel generates the same tension at the start of stimulation. However, during prolonged stimulation, as muscle fibers begin to fatigue, the fast, fatiguable motor units (6) will show a much more rapid fall in tension than the fatigue-resistant, fast and slow motor units. So what might have started as a number of channels, each generating approximately the same tension, may soon become unequal, thus leading to the development of tension oscillations at the stimulation rate.

In the present study, we added a sophistication to the method of distributed stimulation that overcomes the tension oscillation problem. If the tension gener-

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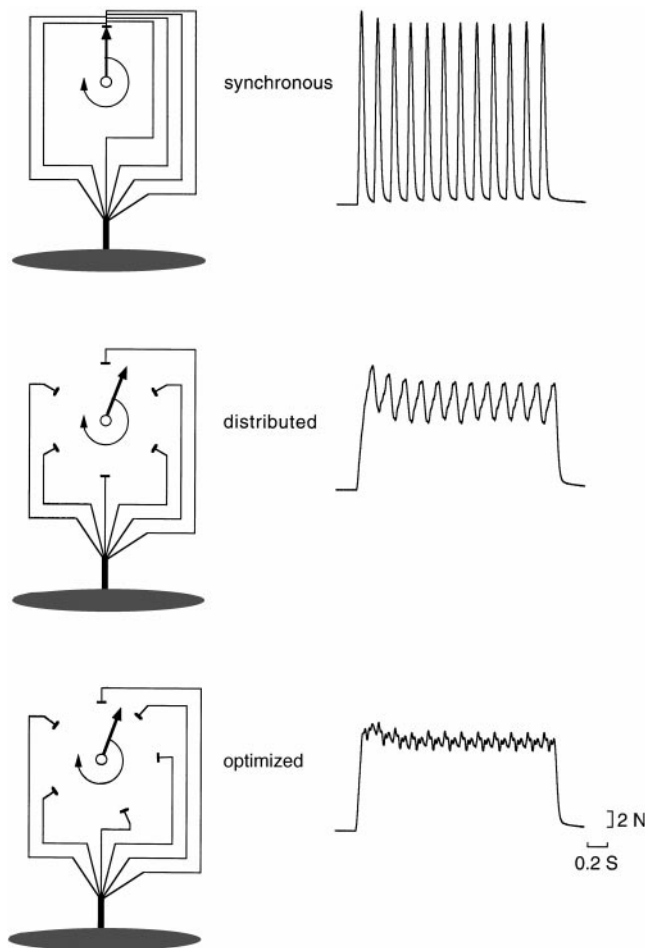


Fig. 1. *Left*: diagrams of stimulation arrangements. The nerve supply to the muscle was separated into 6 portions that were stimulated by the rotating contact. The speed of rotation or stimulation rate is the same for all stimulation protocols. *Right*: actual recordings of the isometric tension in the medial gastrocnemius muscle of the anesthetized cat for each stimulation protocol. Tension (2 N) and time (0.2 s) calibrations for all 3 traces are at *bottom right*. *Top*: 6 pieces of muscle nerve were simultaneously stimulated (synchronous) at 6 pulses per second (pps), leading to a series of twitch contractions. *Middle*: contacts were evenly spaced around the perimeter, generating a series of contractions with equal time intervals between them. Because nerve portions are stimulated one after the other, this is defined as distributed stimulation. Notice that the contraction generated includes a component of maintained tension that has superimposed on it a smaller tension oscillation than for synchronous stimulation. *Bottom*: distributed stimulation with unequal intervals. A computer calculated spacings that minimized tension fluctuations at the speed of rotation of the central contactor. The tension trace shows that this "optimized" stimulation further reduced the tension oscillation (<1% of ripple at stimulation frequency) and increased the amount of steady tension.

ated by different channels is not equal or if inequalities develop as a result of fatigue, the intervals between stimuli to each channel can be adjusted by a computer that controls the stimulus channels. Therefore, if, for example, the tensions generated by two channels are much lower than those of their neighbors, the inter-stimulus interval between the two channels would be decreased, whereas it would be increased for the larger tensions, until the tensions were closer to equal. The

process of minimizing tension ripple is iterative, with continuous adjustments to intervals being possible during stimulation. It means that the tension ripple at the rate of stimulation does not exceed a predetermined level at any time during a prolonged period of stimulation.

We report measurements of tension and electromyogram (EMG) to determine the rate of fatigue using computer optimization of distributed stimulation compared with synchronous stimulation. Our hypothesis is that this new method will allow for longer periods of stimulation with less fatigue, as well as enabling smooth gradation of tension over a wider range than has previously been possible. These are important goals for functional electrical stimulation (FES) of muscle in paralyzed human subjects.

## METHODS

The experiments were carried out on a total of 10 cats of both sexes weighing between 4 and 7.9 kg. All experiments were carried out with approval of the local ethics committee. Anesthesia was induced with an intraperitoneal dose of pentobarbital sodium (40 mg/kg body wt) and was maintained during the course of the experiment with additional doses, when necessary, administered via the cephalic vein. The trachea was cannulated and end-tidal  $\text{CO}_2$  concentration was monitored. Expired  $\text{CO}_2$  levels provided an indication of adequacy of ventilation and depth of anesthesia. Rectal temperature was measured, and body temperature was maintained at  $38^\circ\text{C}$  by using a feedback-regulated heating blanket.

A laminectomy was performed to expose ventral roots L6–S2. These were cut at their point of entry into the spinal cord and deflected onto a dissection plate. Electrical stimulation established where motor axons to the medial gastrocnemius (MG) ran in the ventral roots, typically at L7 and S1. The left hindlimb was dissected to expose the MG muscle. For this, it was necessary to free the MG from the lateral gastrocnemius and soleus and to cut and separate the tendons from the Achilles tendon, leaving just the tendon of MG attached to the calcaneum. All hindlimb nerves other than the MG nerve were cut, including those to the hip muscles. The hindlimb was fixed to a rigid metal frame by using steel pins in the pelvis and at each end of the tibia. Exposed tissues were covered with mineral paraffin oil retained in baths fashioned from skin flaps. Temperature in the paraffin pool was maintained within  $2^\circ\text{C}$  of core body temperature by using heating lamps.

The calcaneum was severed, and the portion attached to the tendon had a 2-mm-diameter hole drilled through it. A threaded rod was passed through the hole, and the calcaneum was clamped between a pair of nuts and washers. This meant that the MG tendon and its attachment to the calcaneum were left essentially undisturbed. Tension was measured with a U-shaped strain gauge bolted to the threaded rod. Compliance of the system was  $5 \mu\text{m}/\text{N}$ .

Muscle EMG was measured with pairs of flexible wires that were bared at their tips and inserted into the belly of the muscle with a fine hypodermic needle. The EMG was filtered with a bandwidth of 10–1,000 Hz and recorded digitally. The rectified, integrated signal was analyzed spike by spike, in response to each stimulus. Measurements were made of spike width, height (peak-to-peak), and integral. When EMG was analyzed during distributed stimulation, it was normalized to account for recording differences. Normal-

ization expressed the EMG relative to its value during the last minute of stimulation. The EMG for the six channels was then averaged, giving one average, normalized value for distributed stimulation.

Because the object of the experiments was to compare the effects of distributed and synchronous stimulation on fatigue, the ventral root supply to MG was divided into two portions (estimated, on stimulation, at approximately one-third and two-thirds of the whole muscle twitch tension) at the start of the experiment. The one-third portion was used for synchronous stimulation. The two-thirds portion was further subdivided into six portions, to be used later for distributed stimulation. Two measurements of fatigue were taken, the first with the two-thirds portion using distributed stimulation. The remaining piece was then fatigued with synchronous stimulation.

A computer-controlled stimulator was constructed and used six of eight available output channels (5). The software was designed so that the recorded tension could be used to calculate intervals between stimuli. Stimuli were delivered by means of an array of bipolar platinum electrodes. Stimulus strength for each output channel was adjusted to produce a maximum contraction for that channel. It was important to avoid stimulus breakthrough, i.e., current from one stimulating electrode spreading to neighboring electrodes.

Evidence for stimulus breakthrough was sought by comparing the size of a contraction from one channel stimulated alone to the contraction size of that channel when stimulated together with the remaining five channels. If tension for each channel during individual and combined stimulation was similar, it was assumed that no stimulus breakthrough occurred. To achieve maximal contractions with each channel, stimulus strength was adjusted to be 3–4 times the stimulus threshold. It is our experience that there is no significant change in stimulus threshold at the end of a period of stimulation used to measure fatigue.

Stimulation rates of 6 and 10 pulses per second (pps) were used. This meant that the whole muscle, when stimulated through six channels, was being stimulated at 36 and 60 pps, respectively. These rates of synchronous stimulation generate near-maximal tension, with a faster rise time at the higher rate. In some practical situations involving FES, periodic bursts of stimuli are used to generate walking movements. Therefore, a second, intermittent stimulation protocol was set up using the same rates of stimulation. A cycle time of 10 s was established consisting of 3 s of distributed or synchronous stimulation, followed by 7 s of rest.

The optimization process began with equal intervals between stimulus channels at the chosen stimulus rate, delivered sequentially over the six channels. The distributor can be imagined as a rotating commutator making contact, successively, with each of the six channels, with the rate of rotation determining the frequency of stimulation of each channel (Fig. 1). With stimulation at 6 pps, when the intervals between stimuli are equal (distributed; Fig. 1), the tension in MG fluctuates with a period representing that rate and an amplitude depending on the difference in tension between the portions, i.e., ~36% of peak tension for the example in Fig. 1.

An algorithm in the program then calculates the amount of tension ripple at the stimulation rate. Any steady tension is subtracted, and a sinusoid is fitted to the ripple trace. For the next sequence, the stimuli for each channel are delayed by an amount that depends on the amplitude of the fitted sinusoid at the time the stimulus pulses occurred for that channel. Therefore, the next set of intervals between stimulus trains will no longer be equal. In Fig. 1, this is shown by making the

spacings between the commutator contacts unequal (optimized).

The computer then sends these new intervals to the stimulator, tension is once more recorded, and new intervals are computed in a convergent iterative process that quickly reduces the fundamental ripple to <1% of mean tension. The number of iterations required for stimulus optimization varied between experiments and depended on the differences in tension generated by each channel at the start of the exper-

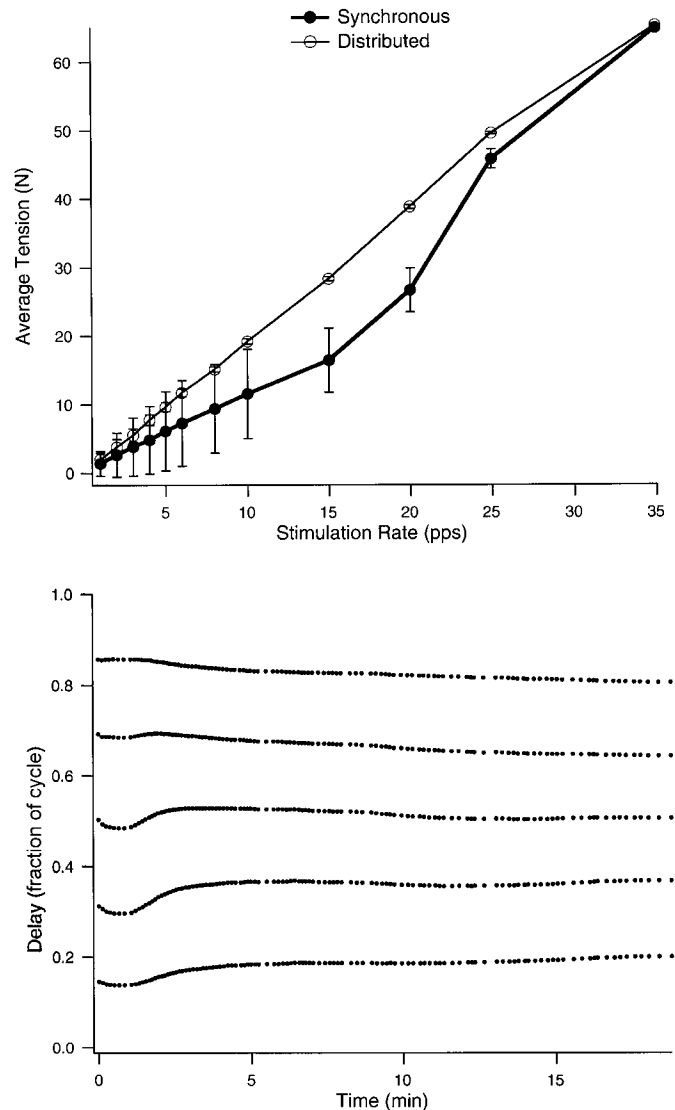


Fig. 2. *Top*: plots of mean tension levels calculated from tension traces and comparison of optimized, distributed stimulation (thin line) with synchronous stimulation (thick line) over a range of stimulation rates from 4 to 35 pps. Error bars indicate the mean peak-to-peak fluctuations in tension at each stimulation rate. *Bottom*: changes in delays between stimulus channels as the tension generated by each channel changes during continuous stimulation at 6 pps over 20 min. There are 6 stimulus channels with 5 sets of intervals between them. For each interval trace, new interval values are indicated by circles. During this 20 min period, there were 111 updates. The number of updates could be controlled manually. The timing of each stimulus is given as the delay between it and the stimulus to channel 1. Notice that, early in stimulation, intervals were changing quite rapidly in response to the period of tension potentiation followed by the onset of fatigue.

Table 1. Comparison of stimulation rates for distributed and synchronous stimulation of the ventral root

	<i>n</i>	Distributed			Synchronous		
		Stimulation rate	Potentiation	Fatigue, 5 min	Stimulation rate	Potentiation	Fatigue, 5 min
Continuous	3	6×6	133.26 ± 2.80	55.94 ± 3.67	36	106.56 ± 0.58	11.61 ± 3.66
	2	6×10	163.05 ± 6.14	61.74 ± 7.96	60	103.25 ± 1.78	4.23 ± 2.90
Intermittent	2	6×6	111.24 ± 1.04	99.55 ± 1.30	36	100.00 ± 0.00	53.16 ± 0.84
	3	6×10	115.88 ± 5.66	75.35 ± 4.84	60	100.66 ± 0.66	46.92 ± 3.22

Values are means ± SE. *n*, No. of experiments.

iment, as well as on any changes in tension during stimulation. For tension differences of ≤30%, between 5 and 20 iterations were typically required to achieve the 1% level.

Theoretically, there are no limits to the differences in tension that can be optimized, provided the biggest portion is no larger than the sum of the other portions. In practice, as differences between portions increase, the amount of higher harmonic ripple increases and limits the smoothing process.

Figure 1 compares the three methods of stimulation. In the first (*top*) section, the six channels were synchronously stimulated. Notice that the tension fluctuations are so large that tension at the end of each twitch is almost zero. The second section (*middle*) shows the effects of distributed stimulation with equal interstimulus intervals. The fluctuations here are much smaller, and tension no longer falls to zero, with the minimum value being 60% of peak tension. Adjusting the spacing between stimulus channels (Fig. 1, *bottom*) so that the fundamental ripple is <1% causes a further reduction in fluctuations, although some ripple persists at higher harmonics of the stimulus frequency. Peak-to-peak ripple for distributed stimulation fell from 46 to 21%. Therefore, the whole muscle is being stimulated at 36 pps, but stimulation is distributed optimally through six channels. Each muscle portion (channel) is stimulated at only 6 pps. This method allows stimulation of muscle portions at rates as low as 6 pps while keeping tension ripple at manageable levels. There is less fluctuation of tension than with synchronous stimulation due to 1) the distributed nature of the inputs and 2) the optimization of the interstimulus intervals.

Figure 2 shows a comparison of the average tension generated and the peak-to-peak fluctuations over a range of stimulus frequencies. Synchronous stimulation produces less average tension and larger fluctuations than distributed, optimized stimulation for all rates of stimulation, until rates are >25 pps, at which the muscle was close to being maximally activated.

The advantage of the optimization procedure is that it becomes possible to generate smooth contractions at low stimulus rates with unequal inputs. Even if the inputs are equal at the start of the experiment, sizeable tension inequalities are likely to develop during the course of stimulation because of different rates of fatigue or potentiation of tension by motor units included in the ventral root portions. Therefore, it is necessary not only to optimize at the start of the experiment, but to repeat the optimization procedure at regular intervals as fatigue sets in. In a typical experiment, during 20 min of continuous distributed stimulation of each of six portions at 6 pps, 111 updates of the optimization process were carried out, which led to progressive changes in the intervals between stimulus channels as fatigue progressed. An example is shown in Fig. 2. There are five sets of delays, representing the intervals between each of the six channels. Notice that the bigger shifts in delays occurred early during the experiment, when twitch potentiation and

the onset of fatigue produced the largest changes between stimulus channels.

Statistical analysis of the data (Table 1) was carried out using an ANOVA. Comparisons were made between the effects of synchronous vs. distributed stimulation, on low (6/36 pps) vs. high (10/60 pps) rates of stimulation, and on continuous vs. intermittent stimulation.

## RESULTS

All distributed stimulation was done across six channels. The combined twitch tension of these six portions was ~70% of the whole muscle twitch. The remaining 30% was used for synchronous stimulation. Of the 10 experiments carried out, 5 measured fatigue using continuous stimulation, and 5 used an intermittent stimulation protocol. Within the continuous stimulation group, three experiments used distributed stimulation at 6 pps and synchronous stimulation at 36 pps. In the remaining two experiments, rates of stimulation were increased to 10 and 60 pps. In the second series of five experiments, which used the intermittent stimulus protocol, the muscle was stimulated for 3 s followed by 7 s of rest. Again, a comparison was made between

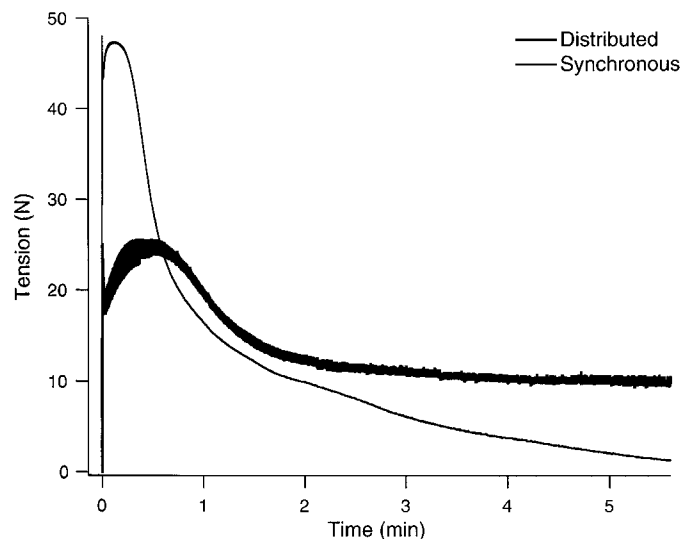


Fig. 3. Comparison from one experiment of the decline in tension during optimized, distributed stimulation of 6 channels, each at 6 pps, and synchronous stimulation at 36 pps over a 5-min period of continuous stimulation. Although the tension generated by synchronous stimulation reached a higher peak, it rapidly fell as fatigue took effect, finishing at a much lower level than that for distributed stimulation.

distributed and synchronous stimulation for rates of 6 channels  $\times$  6 pps and 6 channels  $\times$  10 pps (Table 1).

**Continuous stimulation.** An example of tension changes during continuous stimulation over 5 min is shown in Fig. 3. With both distributed and synchronous stimulation, there was some potentiation of tension during the first second after onset of stimulation. Tension rose to a peak and was followed by a small rapid fall and a second slower rise. Tension then fell progressively as fatigue took effect. In this experiment, despite a larger portion of the muscle being used with distributed stimulation, the tension reached a peak of only 25 N, whereas, with synchronous stimulation, it peaked at 47 N. The lower tension was the result of the much lower rate of stimulation of each portion (6 vs. 36 pps). The other striking difference is in the rate of fatigue. The initial tension was calculated as an average from the first 3 s of stimulation. After 5 min of continuous synchronous stimulation, tension dropped to 4% of its initial value, whereas, in the portion contracted using distributed stimulation, tension at 5 min was still 40% of its initial value. The rate of fatigue was therefore dramatically different. For three experiments, mean tension at 5 min was  $56 \pm 4\%$  (SE) of the initial value for distributed stimulation and  $12 \pm 4\%$  for synchronous stimulation. When stimulation rate was increased from 6 to 10 pps per distributed portion (2 experiments), the 5 min value was  $62 \pm 8\%$  for distributed stimulation and  $4 \pm 3\%$  for synchronous stimulation. The lower value for fatigue with distributed stimulation at 10 pps, compared with 6 pps, was due to the large amount of potentiation present at the onset of stimulation at 10 pps (Table 1). The precise amount of fatigue varied between experiments, as, inevitably, the distribution of fatigue-resistant and fatiguable motor units in the different stimulated portions was never the same.

We posed the following question: was the decline in tension attributable entirely to fatigue processes within muscle fibers or was there some breakdown in neuromuscular transmission as well? To help answer this question, intramuscular EMG was recorded with a pair of flexible wire electrodes during the fatigue procedure. An example of tension and EMG together is shown in Fig. 4 for both synchronous and distributed stimulation. For distributed stimulation, the EMG is the average across the six channels, and, for both forms of stimulation, it was normalized with respect to its value at the end of stimulation.

For synchronous stimulation, after an initial peak, the normalized EMG fell steeply, together with tension, after  $\sim 1$  min of stimulation, briefly steadied, and then fell further. Importantly, it was found that, after several minutes of stimulation, interrupting stimulation for periods of 1 s or more led to significant recovery in both EMG and tension. When stimulation recommenced, both fell rapidly again.

For distributed stimulation, there was an early increase in both tension and EMG, followed by a slow fall, which was less for EMG than for tension (Fig. 4, bottom; note different time scale). After 10 min of

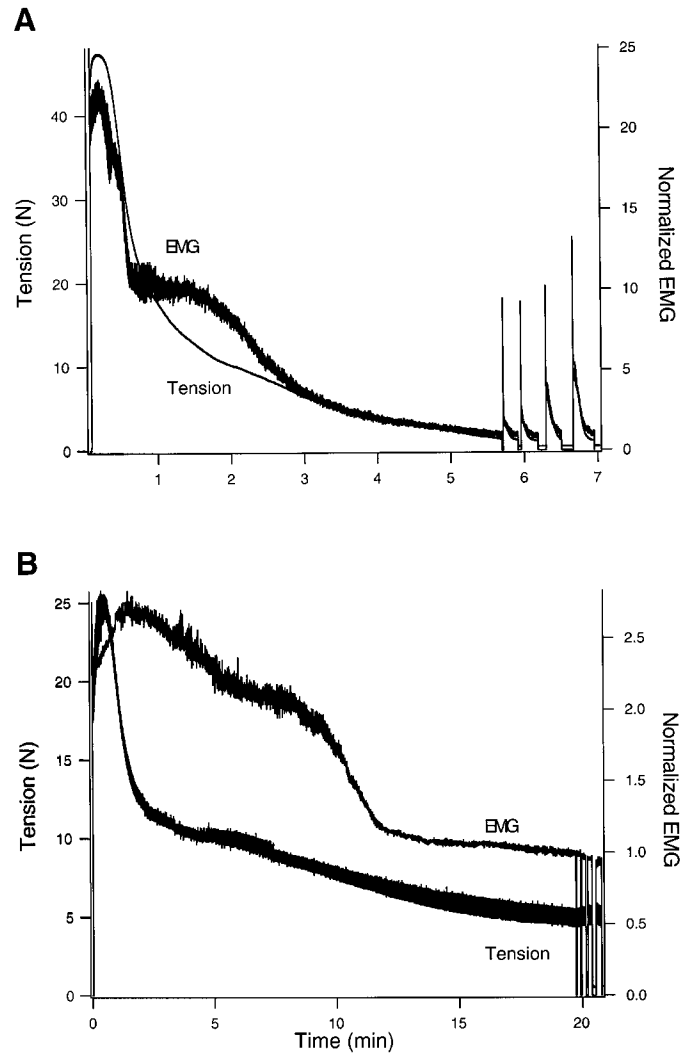


Fig. 4. A: tension (thin line) and electromyogram (EMG; thick line) during synchronous stimulation at 36 pps. After an initial but brief period of potentiation of both tension and EMG, both fell precipitously to finish at 50% of their original value after 1 min of stimulation. The subsequent fall over the following 5 min was more gradual. After 5.7 min, when tension had fallen to 4% of its initial value, stimulation was interrupted for periods of 1–10 s. After resumption of stimulation, there was significant recovery of both tension and EMG that was followed by a rapid decline. B: tension and EMG during continuous, optimized, distributed stimulation at 6 pps in each of 6 channels. There was some potentiation, followed by decline of both tension and EMG. Notice the much longer period of stimulation. At 19 min, stimulation was interrupted for periods of 1–10 s. There was no overshoot of tension or EMG after recommencement of stimulation.

stimulation, EMG fell more steeply; however, this was not reflected in the tension trace. After 20 min of stimulation, interruptions in stimulation of 1–10 s caused both tension and EMG to fall to zero, but both returned to their preinterruption values when stimulation was recommenced. However, EMG had dropped by 50% of its original value, and, therefore, another mechanism, one with a slower recovery time course, had to be responsible for this observed fall.

**Intermittent stimulation.** At a stimulus rate of 6 pps across six channels, for each 3-s burst of stimulation, a

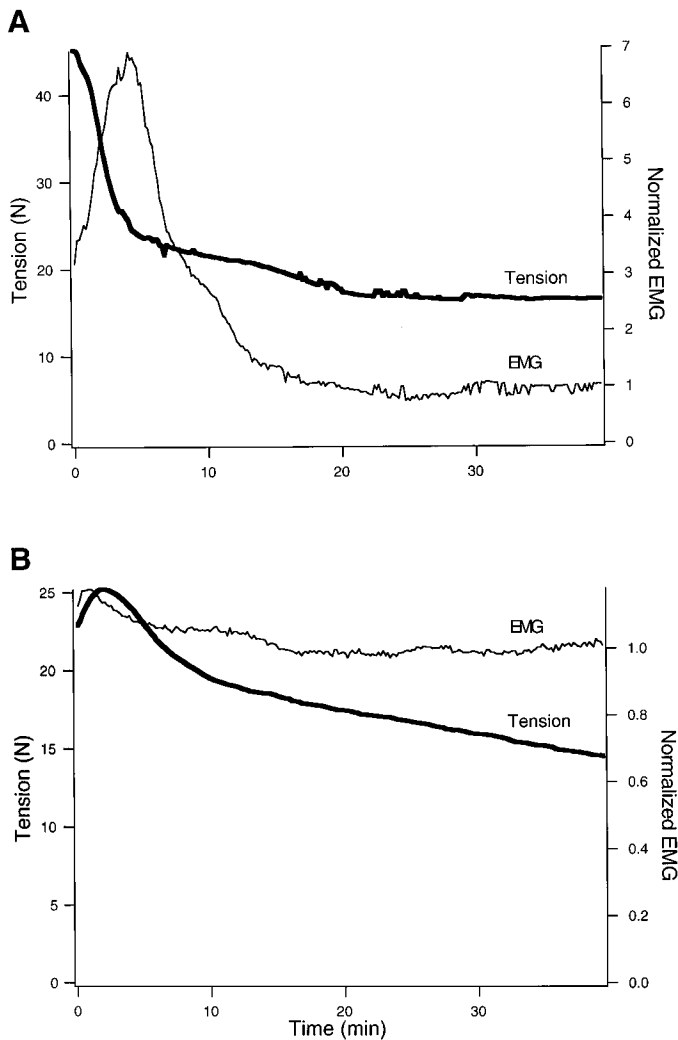


Fig. 5. A: EMG and tension for intermittent, synchronous stimulation at 36 pps. Over the 40-min stimulation period, tension fell by 64% and EMG by 69% of its initial value. B: EMG and tension for intermittent, distributed stimulation at 6 pps in each of 6 channels. There was virtually no drop in EMG and only a 35% drop in tension during the 40 min of stimulation.

channel received 18 stimuli. For synchronous stimulation, it received 108 stimuli. Tension was measured during each contraction. Examples of mean tension and EMG generated using intermittent distributed and synchronous stimulation are shown in Fig. 5. Because tension changes during stimulation were more gradual, recording was continued for a longer time, typically 40 min. With distributed stimulation, tension at the end of 30 min of stimulation was still at 70% of its initial value, whereas, with synchronous stimulation, tension dropped to 37% of the initial value.

During two of the intermittent synchronous stimulation (36 pps) experiments, tension at 5 min dropped to  $53 \pm 1\%$  of its initial value, and there was no potentiation of tension (Table 1). After 5 min of distributed stimulation at 6 pps per channel, tension was still at  $100 \pm 1\%$ , due to an 11% potentiation during the first minute of stimulation. When 10 and 60 pps were

used (3 experiments), tension at 5 min was down to  $47 \pm 3$  and to  $75 \pm 5\%$  with synchronous and distributed stimulation, respectively. There was some early tension potentiation with distributed stimulation, particularly for rates of 10 pps (Table 1).

There was, therefore, considerable fatigue present with synchronous stimulation, even when an intermittent stimulus protocol was used. This again posed the question, was the fatigue entirely muscular or was there a neuromuscular component as well? To determine the answer, it was necessary to look at EMG as well as tension (Fig. 5). It is apparent that, during distributed stimulation, there was very little change in mean EMG levels during each contraction. For synchronous stimulation, after an early peak, the EMG dropped rapidly to a low level.

To determine how well tension was maintained during each contraction with intermittent stimulation, the amount of "sag" (the decline in tension during each contraction) was measured. Sag was expressed as the fall in tension from the start to the end of stimulation, expressed as a percentage of the average tension. Measurements of the amount of sag over the 40 min of stimulation for 6 pps per channel (distributed) and 36 pps (synchronous) stimulation are shown in Fig. 6. With both forms of stimulation, tension was well maintained for the first few contractions but then began to sag. For distributed stimulation, sag increased from an

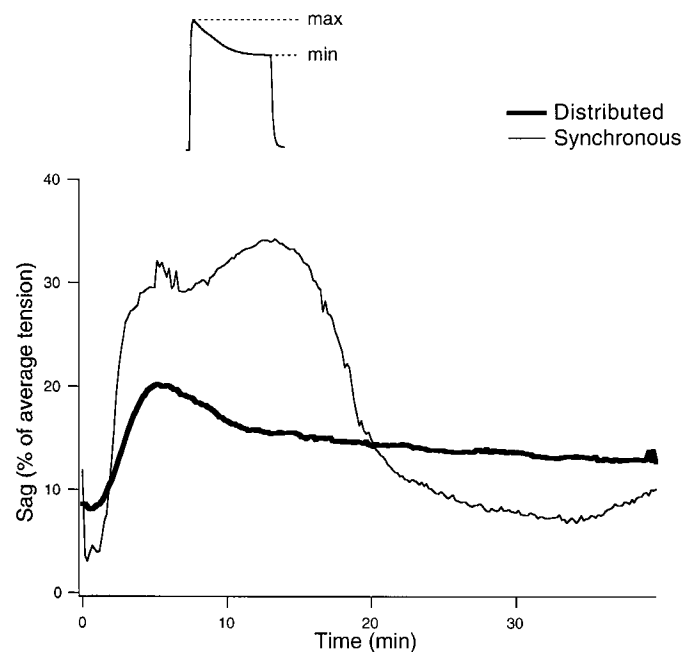


Fig. 6. Sag is the fall in tension from maximum to minimum during each contraction, using the intermittent stimulation protocol. An example of sag during a contraction is shown (*inset*). Sag is quantified by expressing it as a percentage of the average tension during each contraction. During optimized, distributed, stimulation at 6 pps in each of 6 channels (thick line), there is an early increase in sag, which then declines and remains relatively steady after 10 s of stimulation. For synchronous stimulation at 36 pps (thin line), there is a much larger increase in sag that begins to decrease after 15 min of stimulation, reaching a final value below that for distributed stimulation.

initial 8% to a peak of 20% after 5 min of stimulation. The amount of sag then fell to a final value of 12% at 40 min. For synchronous stimulation, the changes were much more dramatic. The initial sag of 12% increased to 30% at 5 min and reached a peak of 34% at 13 min. After 20 min, the amount of tension sag for synchronous stimulation became greatly decreased, ending up, in fact, less for synchronous than for distributed stimulation. This was most likely the result of fatigue in the synchronously stimulated portion of motor units that are characterized by the presence of sag. However, there was some variation in the measured amount of sag in different experiments, presumably reflecting differences in the distribution of fatiguable and fatigue-resistant motor units in the stimulated portions.

**Statistics.** An ANOVA was used to test for significant differences in fatigue and tension potentiation with the different stimulation protocols used (Table 1).

After 5 min of stimulation, there was a significant effect ( $P < 0.001$ ) of the method of stimulation on remaining levels of measured tension. Similarly, there was significantly less fatigue with intermittent vs. continuous stimulation ( $P < 0.001$ ) and with the 6/36 vs. the 10/60 pps stimulation rate ( $P = 0.04$ ).

The amount of potentiation was significantly less with synchronous than distributed stimulation ( $P < 0.001$ ). Continuous stimulation produced significantly more potentiation than intermittent stimulation ( $P < 0.001$ ), and potentiation was greater with lower stimulation rates (6/36 vs. 10/60 pps,  $P = 0.02$ ).

## DISCUSSION

The main object of this study was to test a new method of distributed stimulation, in which intervals between stimuli are under computer control. The method is based on that first used by Rack and Westbury (18; see also Ref. 4), but the computer control of intervals removes the restriction of requiring equal tensions and equal fatigability between stimulus channels.

We used distributed stimulation across six channels in one part of the ventral root and compared it with synchronous stimulation of another part. The method is subject to the criticism that there may be sampling errors. The MG is composed of both fatigue-resistant and fatiguable motor units. If the axons supplying the different unit types are not distributed at random throughout the ventral root, and one subdivided portion contains a proportionately larger number of axons to fatigue-resistant units than another, misleading estimates of fatigue might result. The MG contains ~300 skeletomotor axons that are distributed across ventral roots L7 and S1 (3). In a study of single, identified motor units in this muscle (17), our laboratory showed that axons of different unit types are distributed at random throughout the two motor roots. In the series of experiments described herein, we made a conscious effort not to select root fragments that were always from the same part of the root for distributed or synchronous stimulation. In support of our view that ax-

ons of the fatiguable units were distributed evenly across all stimulated portions, there was always a large amount of fatigue seen with synchronous stimulation, and this varied relatively little from one experiment to the next (Table 1). Such a result would not be expected if there was a bias in the sampling of motor units.

An important issue in these experiments was whether the observed fatigue was attributable entirely to factors within muscle fibers or whether it included a component of neuromuscular transmission failure. The observation that interruption of continuous, synchronous stimulation for 1 s or more (Fig. 4A) led to rapid but short-lived recovery of both EMG and tension, which was followed by a further steep fall when stimulation recommenced, suggests that at least a component of fatigue was neuromuscular. When the experiment was repeated with distributed stimulation (Fig. 4B), there was no rapid recovery. Therefore, presumably, no breakdown in neuromuscular transmission occurred. However, because EMG dropped to 50% of its original value, another source of breakdown in transmission, one with a slower time course, must have been involved. Inspection of individual EMG traces showed that, along with a decline in amplitude, there was an increase in duration of individual potentials, suggesting a slowing of impulse conduction along muscle fibers.

It has been reported that a decline in EMG accompanies the drop in tension during fatigue tests, which was attributed to transmission failure at axonal branch points and at neuromuscular junctions (7, 14). Rapid recovery has been interpreted similarly in terms of both pre- and postsynaptic transmission (12, 20). It appears that fast fatiguable motor units are particularly prone to neuromuscular transmission failure, so that, for these units, the tension decline is not just the result of sarcolemmal or intracellular processes (9). It is interesting to note that the low stimulation rates used with distributed stimulation are very effective in avoiding this problem.

For continuous, distributed stimulation, our working hypothesis is that the decline in EMG is the result of conduction failure along the sarcolemma and into the transverse tubules. There is a buildup of extracellular  $K^+$  dose in the muscle, which leads to depolarization of the sarcolemma. This, in turn, triggers slow inactivation of the  $Na^+$  current (19), which reduces action potential amplitude and slows impulse conduction (13). Such slowing was, in fact, apparent in our records. The rate of removal of  $K^+$  depends on  $Na^+-K^+-ATPase$  activity and on the blood supply, processes that would be expected to be fairly slow and take longer than the 10-s interruption of stimulation.

For the intermittent stimulation protocol (Fig. 5), it was similarly concluded that, during synchronous stimulation, the drop in EMG included a component of neuromuscular fatigue. It is interesting to note that, with the intermittent stimulation protocol, 3 s of stimulation followed by 7 s of rest, for distributed stimulation, the rest period is long enough to avoid any decline

in EMG, because levels were well sustained throughout the 40 min of stimulation. It suggests that only minimal rest intervals are needed for maintenance of EMG levels. This deserves further study in future experiments.

Although stimulating ventral root portions is a convenient method for animal experiments, such an approach would not be feasible with human subjects. The way we plan to overcome this difficulty is to use surface electrodes and to stimulate the muscles percutaneously. We have recently shown that it is possible to achieve smooth, graded contractions of the human triceps surae over the range 0–25% of maximum by using distributed stimulation across 3 channels (10). One important difficulty with such an approach is that contractions evoked by stimuli from each channel are often different in size. Furthermore, with surface stimulation, there is the likelihood of spread of stimulus current from one channel to another, leading to further inequalities in the contracted portions. These problems can now be dealt with by means of the optimization process. In addition, as inequalities between the stimulated portions develop, it will be possible to repeat the optimization any number of times during the course of stimulation, according to the distribution of fatiguable motor units (Fig. 2).

A potential obstacle for human applications of a method of stimulation using distributed, optimized inputs, is the need to access muscle tension for the optimization process. We are currently experimenting with accelerometers, placed at different locations on the limb or over the muscle, to detect muscle-tendon movement during stimulation and to use these movements to obtain approximations of the derivative of tension. This would be an adequate signal for the optimization process.

In previous applications of distributed stimulation, the problem of differential rates of fatigue between stimulus channels was minimized, either by using fatigue-resistant muscle (18) or, in fast, fatiguable muscles by using only brief trains of stimuli separated by long rest periods (15). Now it is possible to stimulate for much longer periods and to use any kind of muscle, i.e., slow, fast, or mixed. Other methods of optimizing stimulus patterns, not based on distributed stimulation, have been used but took advantage of the muscle's catch property (2) or other nonlinearities in the tension summation process (21). However, these approaches use synchronous stimulation of the muscle, which, apart from being a rather unnatural form of activation, brings with it the problems of tension ripple and rapid fatigue. In addition, in any human application, synchronous stimulation leads to a degree of discomfort for the subject.

For low stimulation rates, optimizing the intervals between stimulus channels does not reduce tension fluctuations to zero. However, ripple at the stimulus frequency is brought down to 1%, and all that is left is harmonic ripple (compare Fig. 1, *middle* and *bottom*). The ripple will be at higher frequencies than the stimulation rate, and, taking the inertia of the limb into

account, it will lead to proportionately less tremor. This is, therefore, a specific advantage of the method over distributed stimulation with equal intervals.

The principal finding made here was that, with optimized, distributed stimulation, fatigue was less than with synchronous stimulation by 5- to 20-fold when the muscle was stimulated continuously and by ~2-fold when intermittent stimulation was used (Table 1). In other words, synchronous stimulation, especially if applied continuously, is particularly prone to the development of fatigue. In making these observations, we do not want to claim any remarkable new result, as it would be expected that stimulating a muscle portion at 36 pps leads to more rapid fatigue than stimulation at 6 pps. The importance of the observation is that, in present FES applications, synchronous stimulation is always used. In this paper, we point out the significant advantages of distributed stimulation. It might be argued that any advantage is achieved at the expense of a reduced level of tension output from the muscle. However, in FES applications such as adoption of a given posture and the generation of slow movements in a paralyzed subject, it is not necessary to generate maximum tensions, and a smooth, submaximal contraction is all that is needed.

We chose 6 and 36 pps as appropriate rates of stimulation because they represent the lower and upper limits of the normal range of motor unit firing rates (1). We included some experiments using 10 and 60 pps because this would lead to an increase in the level of tension and rate of rise of tension. One consideration for an FES application is the rate of rise of tension during each period of stimulation. A more rapid rise means that, during intermittent stimulation, higher repetition rates are possible and shorter-duration stimulus trains can be used (2). It also reduces the delay between stimulation and the rise of tension, which would be an advantage for closed-loop control of muscle. We included experiments using intermittent stimulation because of the possibility of a future application of FES for locomotion in paralyzed human subjects. In our experiments, the muscle was stimulated for 3 s in a 10-s cycle, giving a duty cycle of 0.3. This compares with the normal duty cycle for human locomotion of 0.3–0.5.

We predict that computer-controlled stimulus patterns, like those used in this study, will be used in future FES applications. This development will be helped by further advances in computer miniaturization and increases in computational power. One obvious goal is to regulate muscle length and force using a closed-loop control system with afferent feedback from goniometers and force sensors (16). An important consideration for such a systems approach is that, during stimulation, muscle force output must remain reasonably constant for a constant level of stimulation. In the experiments using intermittent stimulation, we measured not only mean tension levels during each contraction but the amount of sag that was present as well (Fig. 6), as this would have implications for any systems application. Sag becomes particularly prominent

when continuous stimulation is briefly interrupted (Fig. 4), presumably because of the involvement of neuromuscular fatigue. Sag represents a change in the muscle transfer function within a contraction, requiring the control system to adapt during each contraction instead of only between contractions. This would represent a significant complication for application to closed loop control of human locomotion. Our data suggest that sag is considerably less when distributed stimulation is used. Thus, apart from other advantages, in situations using a systems approach, distributed stimulation will be the method of choice.

In summary, we introduced a method that permits distributed stimulation of unequal portions of the muscle. The resulting tension is smoother over more of the physiological range of stimulation rates than with synchronous stimulation. Intermittent, distributed stimulation, as might be used in FES for human locomotion, is also more suitable for closed-loop control of muscle. Given that distributed stimulation is better tolerated by human subjects than synchronous stimulation (10), this approach represents a significant step forward in helping to rehabilitate paralyzed human subjects.

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