Orderly stimulation of skeletal muscle motor units with tripolar nerve cuff electrode

Orderly Stimulation of Skeletal Muscle Motor Units with Tripolar Nerve Cuff Electrode

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Abstract—An electrical nerve stimulation technique, using single tripolar electrode, was shown to be capable of recruiting motor units according to their size, while allowing simultaneous but independent control of firing rate in the active units. Test paradigms consisting of established fundamental physiological concepts of soleus-gastrocnemius architecture, motor units conduction velocity, firing rate behavior of motor units of different sizes, and their susceptibility to fatigue were employed to validate the technique and demonstrate its utility as a basic and applied research tool.

INTRODUCTION

ELECTRICAL nerve stimulation may be a highly useful tool for the *in vivo* study of skeletal muscle contractile properties in experimental animals. Unfortunately, currently available stimulation techniques fail to provide contraction under conditions which are similar to the known motor unit recruitment strategies of various muscles. An often used technique delivers to the nerve brief rectangular pulses of suprathreshold amplitude while varying the pulse frequency (rate coding). In essence, all the motor units are always active, which is significantly different from the "size principle" mode [7] under which units are recruited according to their size, small ones first and then progressively larger ones. Attempts to elicit a more favorable condition by increasing the stimulus pulse amplitude from subthreshold to suprathreshold are also deficient since the larger axons have lower excitation threshold [2] and are always activated first in a "reverse recruitment" mode. Such an approach yields large initial force increments and results in fast onset of fatigue which is characteristic of large motor units. The problem, therefore, is the lack of a method which can allow excitation of small units first and progressively larger ones with capabilities to simultaneously modify the action potential firing rate of the active units at various strategies similar to that observed in various skeletal muscles [1], [4], [6].

The early work of Wedensky [11] showed that high frequency (20 kHz) electrical stimulation of the muscle nerve block its contraction. Tanner [10], who studied Wedensky's high frequency technique, demonstrated that inhi-

bition of motor units was dependent on their size as the stimulus amplitude varied. Lowering the stimulus amplitude allowed axons of small size, which have high excitation thresholds to electrical stimulus, to escape the inhibition and conduct action potentials elicited by a proximal stimulus. Further reduction of the high-frequency stimulus amplitude allowed progressively larger axons to escape the inhibition and conduct action potentials to the muscle. Tanner's convincing demonstration was not surprising since it only confirmed Erlanger's Nobel Prize (1944) winning work [2], in which he shows, with single axon recordings from a whole nerve trunk, that the excitation threshold to an electrical stimulus was dependent on the axon diameter (or its conduction velocity). Large diameter axons were shown to have low excitation thresholds, while progressively smaller axons had progressively higher thresholds. Erlanger's work is recognized today in the well-known strength-duration curves for axons of various diameters. Tanner, therefore, extended Erlanger's findings to stimuli of high repetition rates, which inhibit muscular activity. Our previous work [9] with high frequency stimulation demonstrated that rates of 600 Hz provided the most efficient block (i.e., requiring the lowest stimulus amplitude), and that the stimuli could consist of short rectangular pulses of 100 μS duration, instead of a sinusoidal waveform. In the many studies performed, the high-frequency block was applied for over 9 h on and off without significant alteration of muscle properties. The literature provides support to the notion that such high frequency maintains the endplates in a temporary refractory state, thereby preventing the muscle from contracting [9].

The objective of this paper is to describe and validate a new method, employing high frequency stimulation, which can recruit motor units according to their size, while maintaining fully independent control of the firing rate. The technique is based on dual stimuli applied to the nerve with cuff electrode, and may serve as a useful tool to study various electrophysiological and biomechanical properties of muscles under more favorable physiologic conditions than currently employed stimulation techniques.

METHOD

Preparation

Six adult cats were anesthetized with chloralose (60 mg/kg). All hind limb muscles in the leg were dener-

vated except for the muscle nerves to the soleus and m. gastrocnemius. Later, the muscle nerve to one of the muscles above was cut in order to study the response of a single muscle. The calcaneal tendon was cut at its insertion and connected to a force transducer. Pins were inserted in the distal femur and mid-tibia, and attached to a rigid frame including a pelvic clamp to provide isometric conditions for the muscles under study (with hip, knee, and ankle at 90° to each other).

Instrumentation

A tripolar cuff electrode consisting of a 1 cm long cuff with 3 mm spacing between the three stainless steel poles, was mounted on the sciatic nerve as shown in Fig. 1. The center pole was used as common for the two stimulators, while the stimuli were delivered to the distal and proximal poles.

The instrumentation of the stimulation system was described in detail elsewhere [13]. Briefly, an IBM-XT computer delivered two voltage envelopes. The first envelope served as the input to a voltage-controlled oscillator (VCO), the output of which yielded 100 μS rectangular pulses at suprathreshold amplitude with rate corresponding to the input voltage to the VCO. Rates from 0–100 pulses per second (pps) were available. This constituted the firing rate (FR) stimulus.

The second voltage envelope was modulated by 100 μS rectangular pulses at 600 pps. The pulse amplitude was governed by the voltage envelope input and normally decreased linearly from the just-above-threshold of the smallest axon to the just-below-threshold of the largest axon. This constituted the recruitment (R) stimulus. Since suprathreshold high-frequency (600 pps) stimulation blocks the muscle from contracting as response to the FR stimulus [9]–[11], [13], gradual reduction of the pulse amplitude (as shown in Fig. 2), allows the smallest motor axons (which have high excitation thresholds) to escape the blocking effect of the R stimulus first, and conduct the action potentials generated by the FR stimulus. Progressively larger axons escape the R stimulus as its pulse amplitude decreases and allows their orderly recruitment similar to that seen in voluntary contraction [7].

EMG was recorded with two fine, insulated stainless steel wires except for a 1 mm exposed tip which were inserted with hypodermic needles in each muscle at about one half the distance between its motor point and the distal tendon. The EMG was amplified differentially with 110 DB CMRR and bandpass filtered (10–500 Hz). Force was recorded with a Grass FT-10 transducer. Force and EMG were recorded by an IBM-XT computer at 6000 Hz sampling rate.

Protocol

Three test categories were applied once the FR stimulus was calibrated at 1.5 × suprathreshold, and the R stimulus values set at just-above-threshold of the smallest axon and just-below-threshold of the largest axon. All threshold values were determined experimentally.
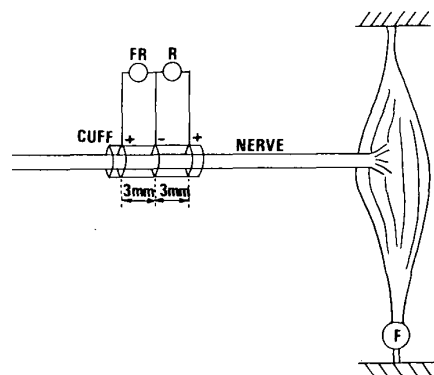

Fig. 1. Schematic of the tripolar electrode mounted on the nerve-muscle preparation, showing the set-up of the two stimulators. (FR—firing rate, R—recruitment, F—the force transducer.)
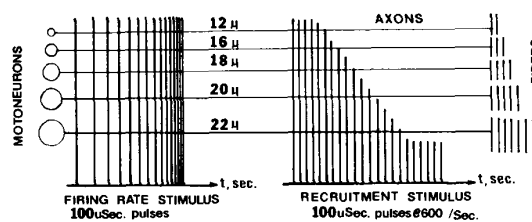

Fig. 2. Schematic of a sample of motor units of increasing size, their increasing axon diameter and innervation ratio, and the two stimuli. Each stimulus spike represents a 100 μS pulse and both stimuli have a common time base. Note the increase of firing rate with time and the simultaneous decrease of pulse amplitude in the recruitment stimulus, which allows progressively larger axons to escape its blocking effect and conduct the FR action potentials.

In the first test, with both soleus and m. gastrocnemius muscle nerves intact, the R stimulus pulse amplitude was linearly decreased, while the FR stimulus rate was fixed at 45 pps. Since the soleus predominantly consists of small motor units [5], [8], while the m. gastrocnemius has primarily large motor units [3], [12], as shown in Fig. 3, then M-wave activity should be evident from the soleus well in advance of any M waves in the m. gastrocnemius, if recruitment of motor units was accomplished according to the size principle. Conversely, applying a stimulus of linearly increasing pulse amplitude at constant rate [shown schematically Fig. 5(b)] via two poles of the electrode with the FR stimulus set at zero should induce *reverse recruitment*, exciting the large axons first and smaller ones later, as the pulse amplitude increases [2]. Under such conditions, the EMG from the m. gastrocnemius should be evident well in advance of any M waves in the soleus, if reverse recruitment indeed occurred.

The second test category was performed on a single muscle, with the nerve branch to the soleus or m. gastrocnemius cut. The FR was fixed at a constant value while the R stimulus was applied. FR values of 5, 10, 20, 30, 40, and 50 pps were used each in a separate trial. Since the M wave is the response to each single pulse of the FR stimulus, the number and rate of the recorded M waves for any time period should be identical to that of the FR
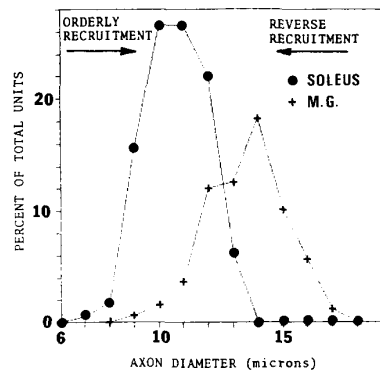
Fig. 3. The distribution of soleus and m. gastrocnemius motor units versus their axon diameter (compounded data from 3, 5, 8, and 12). Note that an orderly recruitment stimulus will excite axons from left to right (as shown by the arrow), activating the soleus first. A reverse recruitment (shown by a right to left arrow) will, however, excite the m. gastrocnemius first.

stimulus, pointing out that there is no interaction between the two stimuli.

Since the conduction velocity of action potentials in the axons and muscle fibers of small motor units is slower relative to the conduction velocity in larger motor units [2], one should expect the latency from each applied FR stimulus pulse to the resulting M wave to decrease if motor units were recruited according to the size principle. To demonstrate that recruitment was orderly, we selected several sequentially recruited M waves recorded at constant firing rate and displayed the duration of their occurrence from the time of their respective FR stimulus pulse application.

Furthermore, if recruitment is indeed orderly, the force trace should exhibit the gradual appearance and then increase of unfused force late in the recruitment phase (i.e., late in the force rise phase), as the large motor units are recruited in trials when the firing rate is fixed below their frequency of fusion (5–20 pps).

Another test category consisted of validating the orderly recruitment from the fatigue standpoint. With the FR set constant, the R stimulus was adjusted to decrease slightly such that only a few small motor units were recruited within 1 s and then remained active for 10 s. Since small motor units are fatigue resistant compared to larger motor units, they were expected to demonstrate relatively little reduction of force at the end of the trial. In consecutive trials, the R stimulus was adjusted to recruit larger motor units, with the expectation of larger decrease of force at the end of 10 s of activity due to the gradual decrease in resistance to fatigue, which is characteristic of larger units. The percent fatigue was calculated, representing the amount of force reduction at the end of 10 s, as compared to the peak force.

It is well established that the median frequency (MF) of the electromyograms power density spectrum is linearly related to the average conduction velocity (CV) of the active motor units in the muscle [1]. In order to quantitatively assess rate of increase of the average CV, the MF of each M wave during orderly recruitment of motor
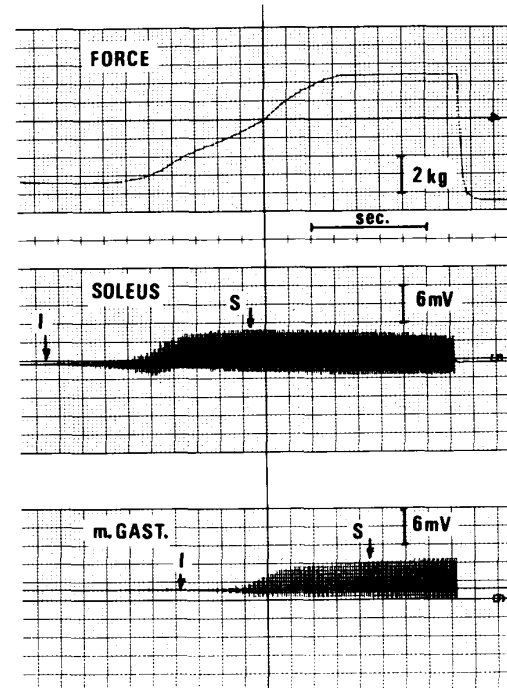

Fig. 4. The force, soleus EMG and m. gastrocnemius EMG responses to the orderly recruitment stimulus of Fig. 2. The EMG (M-wave train) from the soleus was seen first, about 1 s before any significant EMG in the m. gastrocnemius, confirming the ability of the stimulus to excite small units first and progressively larger ones later. Note the low level initial discharge in the m. gastrocnemius, confirming the existence of some small motor units in that muscle. (I—initiation of the M wave. S—saturation of the M wave.).

units at constant firing rate of 40 pps was calculated and plotted versus time. Each M wave was multiplied by the tukey window, the fast Fourier transform (FFT) was then taken, and the power density spectrum determined. The MF was then calculated according to the following relationship:

$$MF \rightarrow \int_0^{mf} P(f)\, df = \int_{mf}^{\infty} P(f)\, df = 1/2 \int_0^{\infty} P(f)\, df$$

where $P(f)$ is the power density spectrum and f is the frequency.

RESULTS

M-Waves During Orderly and Reversed Recruitment

Fig. 4 shows the force, soleus EMG, and m. gastrocnemius EMG recorded during orderly recruitment of motor units while the FR was constant at 45 pps. It is evident that initiation and increase in the M-wave amplitude was seen in the soleus nearly 1 s before any significant activity in the m. gastrocnemius was detected (excluding the low level discharge of some small units known to exist in the M.G.), attesting to the fact that the soleus's small motor units became active first. Furthermore, significant M-wave initiation in the m. gastrocnemius was evident shortly before the M-wave amplitude of the soleus saturated (once all its motor units were recruited), confirming the partial
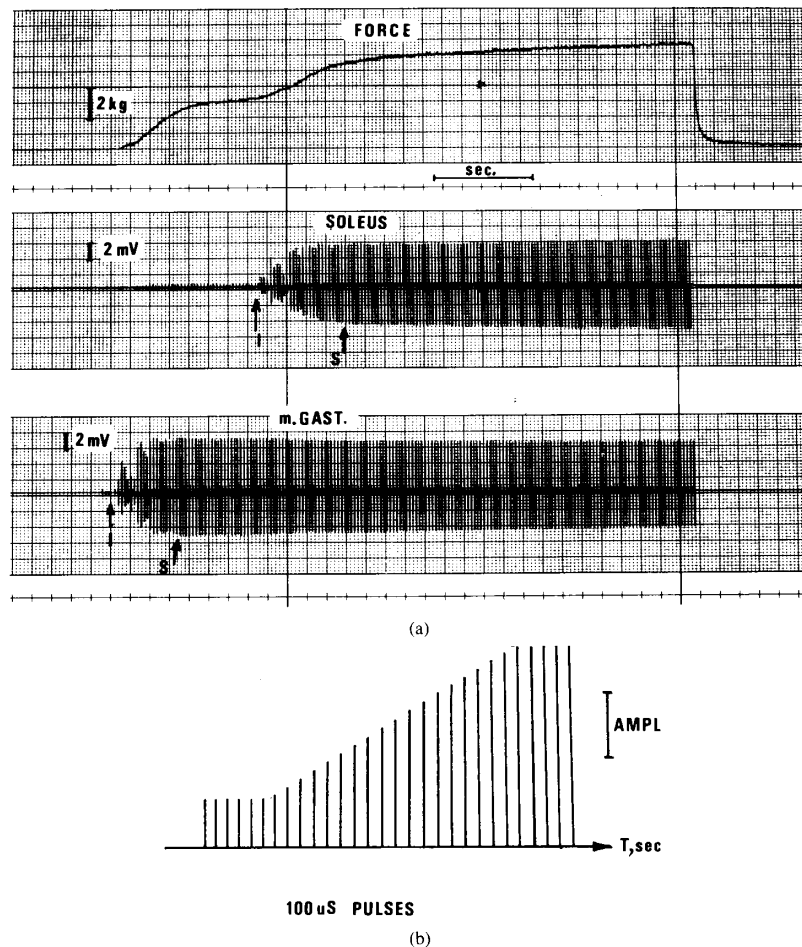
(a)

100 uS PULSES

(b)

Fig. 5. In this figure (a) shows the force, soleus EMG, and m. gastrocnemius EMG responses to the reversed recruitment stimulus shown schematically in (b). Note that the m. gastrocnemius initiated EMG discharge 1.1 s before any EMG discharge was seen in the soleus, confirming Erlanger's strength-duration curves for axons of different diameter or the reverse recruitment properties of pulses of increasing amplitude. In (b), each vertical spike represents a 100 μS pulse, and the pulses are increasing in intensity to recruit large diameter axons first and smaller ones later. The rate of the pulses was set at 55 pps.

overlap of some motor units in both muscles, as reported by Burke and Tsairis [3] and also shown in Fig. 3.

A similar response pattern was obtained from all six preparations in which EMG discharge from the soleus was seen well in advance of any discharge in the m. gastrocnemius. For a 3 s full recruitment period, the lead time of the soleus discharge over that of the m. gastrocnemius ranged from 0.92 to 1.04 s and the average lead time of all preparations was 0.98 s. The minor variability of the lead time was attributed to minor anatomical variations which exist from animal to animal.

Fig. 5(a) shows the force, soleus EMG, and m. gastrocnemius EMG during *reverse recruitment* [shown schematically in Fig. 5(b)] applied via two poles of the electrode (common and positive) at a fixed rate of 55 pps (the FR stimulus was set at zero). It is evident from the

traces that M-wave activity in the m. gastrocnemius is seen nearly 1.2 s prior to any M-wave activity in the soleus, confirming that the larger motor units were excited first in this reverse recruitment stimulation paradigm, as was first described by Erlanger [2]. It can be suggested, then, that the control paradigm shown in Fig. 2 indeed recruited small motor units first and larger ones later, in an orderly manner, as was confirmed by the consistently reproduced response in the six preparations.

Force Response During Orderly Recruitment

Fig. 6 shows force traces which are the response to orderly recruitment applied while the FR was constant at various rates during each trial. Two important facts are evident from the traces: the gradual appearance and then
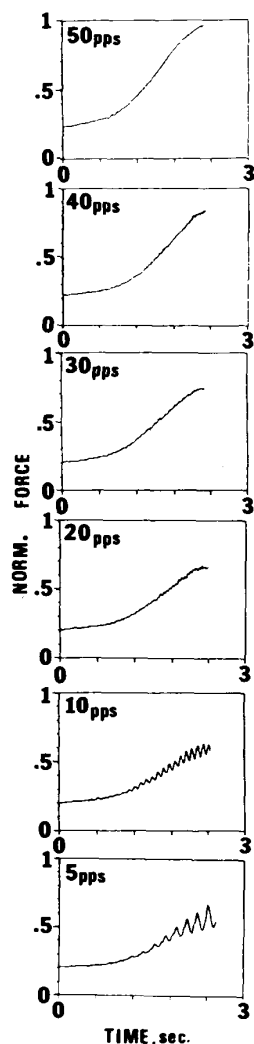
Fig. 6. Normalized force versus time traces from a m. gastrocnemius preparation subjected to the orderly recruitment stimulus, while the firing rate was constant at several values. Note that in the traces where the firing rate was fixed at 5, 10, and 20 pps throughout the recruitment cycle, the initial increase of force was smooth, but as larger units (which require higher initial firing rate) were recruited, the force shows gradual appearance and increase of unfused twitches, clearly demonstrating that orderly recruitment took place. (Force was normalized with respect to the maximal tetanic force at 50 pps.)

firing rate was kept constant, below the initial activation rate of such large units, they responded with unfused force upon their recruitment. The important fact is that the initial force was smooth, indicating that small motor units (of low initial firing rate) were recruited first. The gradual appearance of twitches, as well as the gradual increase of their amplitude, indicated the progressive recruitment of larger motor units which require progressively higher initial firing rate to yield fused force. Since the FR stimuli were held constant, at low rates (5, 10, 20 pps), the medium size motor units contributed their force in a partially fused manner while the larger ones, recruited last, responded with pure twitches. The unfused force seen only at the late phase of the force rise also suggest that motor units were recruited according to the size principle.

The Independence of the Firing Rate from the Recruitment Stimulus

Fig. 7 shows six traces (corresponding to the force traces of Fig. 6 but in reverse order) of EMG (M-wave trains) recordings from the m. gastrocnemius during orderly recruitment, while the FR stimulus was set constant at 5, 10, 20, 30, 40, and 50 pps, respectively. It is evident from the figure that the rate of the M waves is identical to the FR stimulus rate, demonstrating total independence of the R from the FR stimuli. Furthermore, the amplitude of the M wave demonstrated gradual increase as larger motor units were recruited and their corresponding MUAP's spatially added to those of the previously active motor units.

Fig. 8(a) shows the M-wave train recorded from a trial in which motor units were recruited in order of their size while the firing rate was simultaneously increased. In this trial, the last motor unit was recruited as the FR just reached its highest value. Such control strategy is similar to that employed by the biceps and deltoid [1]. Careful evaluation of the figure confirms that the inter M-wave interval was indeed shorter and its rate identical to the firing rate as it increased, while each consecutive M-wave amplitude was larger as newly recruited MUAP's were spatially adding. The last few M waves, two of which are shown, were of the largest amplitude, indicating that all units were fully recruited.

Fig. 8(b) was recorded from a trial in which all the motor units were fully recruited according to the size principle to generate 50 percent of the muscles maximal force, while the firing rate was simultaneously increasing throughout the full force generation cycle, a control strategy similar to that employed by the FDI muscle [1]. It is evident from the figure that the M-wave amplitude was indeed increasing during the initial period, then saturated and remained constant in the final force generation period. The inter M-wave interval, however, was progressively shorter throughout the trial, a manifestation of the simultaneously increasing firing rate.

In summary, Figs. 7 and 8 demonstrate the independence of the firing rate from the recruitment stimulus, as well as the ability of the control scheme to increase the

increase of unfused force in the final segment of the force rise for trails in which low firing rates were applied (5, 10, and 20 pps), and the progressive reduction of the maximal force as the FR was reduced. The gradual appearance and increase of the unfused force in the middle and terminal phases of the force rise are the manifestation of the gradual recruitment of larger motor units at rates much below their rate of force fusion. This observation is in agreement with the well-founded firing rate behavior of motor units in voluntary contraction, with large units, which are normally recruited later in the force generation phase, having higher initial, as well as higher maximal, firing rates than small sized units [1], [4], [6]. Since the

5pps

10pps
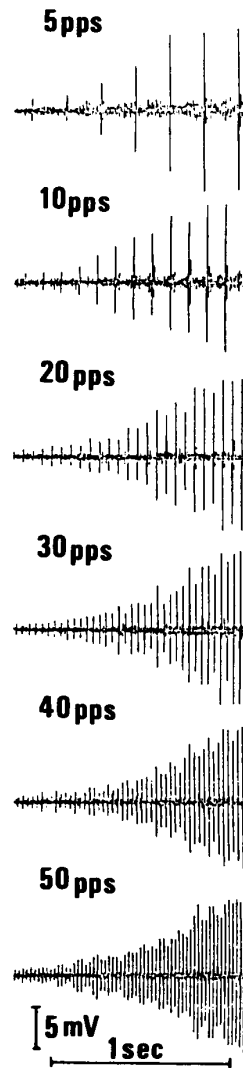
20pps

30pps

40pps

50pps

[5mV 1sec

Fig. 7. *M*-wave trains recorded from a m. gastrocnemius preparation during the application of orderly recruitment, while the firing rate held constant at various frequencies. Note that the number of *M* waves per second is identical to the FR frequency, confirming the independence of the two stimuli. Also note the gradual increase of the *M*-wave amplitude due to the spatial addition of the muscle action potentials from the newly recruited units.

firing rate simultaneously with recruitment and at various control strategies, in which all the motor units are fully recruited to generate any portion of the initial force increase [13].

Fatigue

Fig. 9 shows typical force traces from an isolated soleus preparation in which motor units were recruited over 1 s and then left active for a 10 s duration. The figure clearly shows that when only a few small motor units are recruited (trace *a*), the force displays minimal fatigue (12 percent decrease from the peak force), confirming the greater fatigue resistance of such small units. As larger units (which are more susceptible to fatigue) are re-
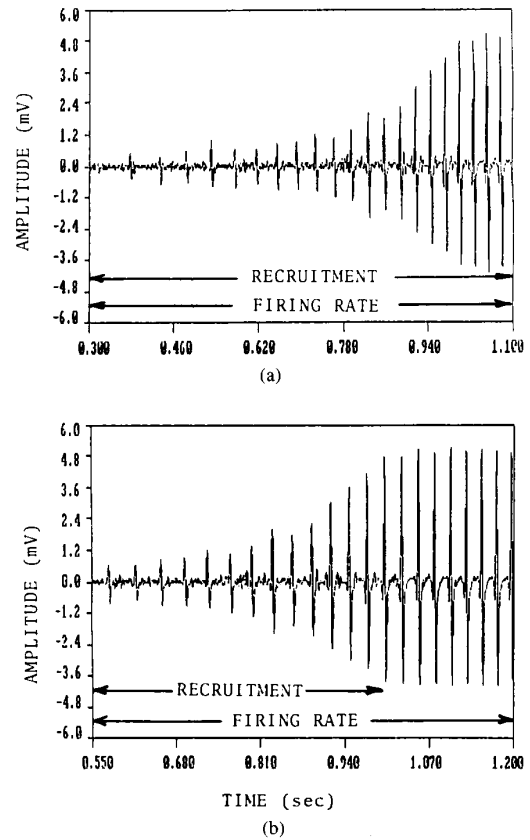
Fig. 8. Showing the *M*-wave train during (a) increase in the firing rate throughout the orderly recruitment cycle, and (b), increase in firing rate during and after all the motor units were fully recruited. Note that the constant amplitude of the *M* wave after the completion of the recruitment cycle in (b), which attest that no further addition of MUAP's occurred.
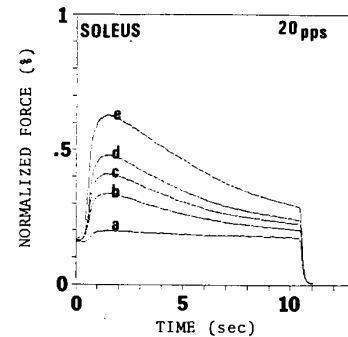
Fig. 9. Force traces recorded from a soleus preparation at various recruitment levels. In trace *a*, only a few small, fatigue-resistant units were recruited, demonstrating little fatigue, while in traces *b*, *c*, *d*, and *e*, progressively larger motor units, which are more susceptible to fatigue, were recruited. The force traces corroborate that orderly recruitment occurred by the gradual increase in fatigue as larger units were recruited. (The force of each trial was normalized with respect to the maximal tetanic force of the muscle at 47 pps.)

cruited, the force traces display gradual increase in fatigue. The fatigue calculated for the force traces *b*, *c*, *d*, and *e*, indicated 36, 46, 51, and 56 percent reduction of force, respectively. The traces, therefore, demonstrate the
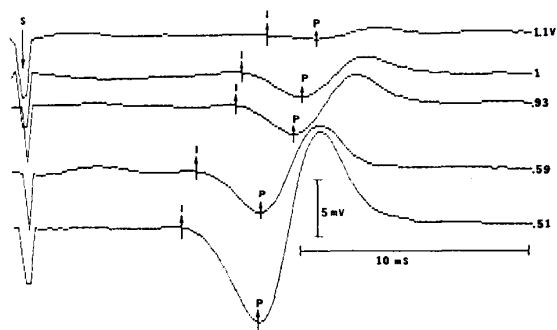
Fig. 10. Five sequential traces of FR stimulus pulses and their respective M-waves as the R stimulus voltage was decreased. Note the decrease in the duration between the FR stimulus pulse (S) and the initiation (I) and peak (P) of the resulting M wave as larger motor units with faster action potential conduction velocity were progressively recruited. The R stimulus pulse amplitude shown to the right of each trace.
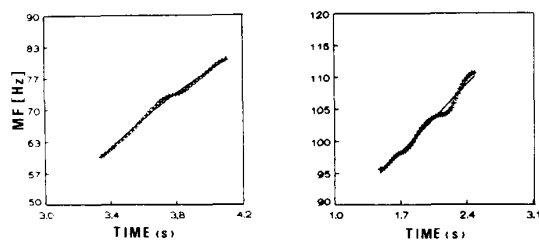

Fig. 11. MF versus time plots during orderly recruitment of motor units with the FR kept constant. Note that the data from two separate preparations rises nearly linearly as the average conduction velocity increases during the orderly recruitment phase.

gradual decrease in resistance to fatigue associated with the increasing size of motor units. This, in turn, further suggests that recruitment was accomplished according to the size principle, exciting the small, fatigue-resistant units first, and the fast-to-fatigue, larger units later.

Conduction Velocity

Fig. 10 displays five traces of the duration between the FR stimulus pulses and their resulting M waves as the R stimulus pulse amplitude was gradually decreased. It is evident from the figure that the initiation as well as the negative peaks of the M waves occurred earlier as motor units were recruited. This, in turn, points out that the conduction velocity of the early recruited units were rather slow when compared to the conduction velocity of the motor units which were recruited later, as the R stimulus pulse amplitude was reduced. (Note the R stimulus pulse amplitude voltage on the right of each trace.)

It could be concluded, then, that based on the conduction velocity of the active motor units, the data further suggest that recruitment took place according to the size principle [7].

The Median Frequency (MF) Response

Fig. 11 shows MF versus time plots calculated for two separate M-wave trains in which all the motor units were

orderly recruited while the firing rate was constant at 40 pps throughout the trial.

It is evident that the MF increased nearly linearly during the recruitment phase of the trials, indicating that the average CV increased as progressively larger, faster conducting units were excited. The consistent similar response from several preparations further confirms that the stimulation method employed in this paper allows orderly recruitment pattern of the motor unit pool of a muscle.

CONCLUSIONS

The data presented above suggest that on a macroscopic level the stimulation technique used in this paper has the capabilities to recruit motor units according to their size, small ones first and progressively larger ones later (as the R pulse amplitude is decreased). This fact was established from test paradigms including: the differential motor unit architecture of the soleus and m. gastrocnemius, the faster conduction velocity of larger motor units, the increasing MF during recruitment, the gradually increasing initial firing rate of progressively larger motor units, and the gradually increased susceptibility to fatigue of such larger motor units.

Combining the above evidence with the time-proven classical strength-duration curves established by the single fiber recordings of Erlanger [2], it could be concluded that the technique presented in this paper allows one to recruit motor units according to the size principle.

Based on the M-wave recordings presented in this paper, it is clear that complete independence in the control of the motor units firing rate and their orderly recruitment is available when using the described technique. Furthermore, the technique developed here allows the activation of a muscle with a large choice of firing rate and recruitment control strategies similar to that of various skeletal muscles by proper adjustment of the two stimuli.

The reader should recognize, however, that in the technique described here, all the motor units fired synchronously, while under voluntary control, asynchronous discharge is evident. Unfortunately, attempts to induce asynchronous discharge may require improvement in technology which are not currently available, but are well worth looking forward to.
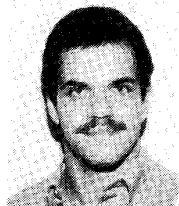
In summary, the technique described here, using single tripolar electrodes, allows recruitment of motor units according to their size independently and simultaneously with changes of firing rate at various control strategies similar to the voluntary control of various muscles. Such a technique provides a highly useful tool for basic and applied investigations into the biomechanics and electrophysiology of skeletal muscles [14]-[17].

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on the Biomechanics of Movement and the external control of the neuromuscular system.

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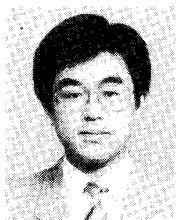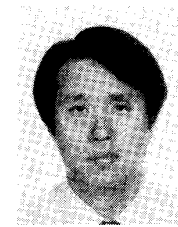
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