Journal of Neurology, Neurosurgery, and Psychiatry 1986;49:677–685

Axonal stimulation for end-plate jitter studies

JOŽE V TRONTELJ,* MARJAN MIHELIN,† JOSE M FERNANDEZ,‡ ERIK STÅLBERG§

From the University Institute of Clinical Neurophysiology, the University Medical Centre of Ljubljana, Yugoslavia,† Department of Clinical Neurophysiology, Valle de Hebron Hospital, Barcelona, Spain,‡ and Department of Clinical Neurophysiology, University Hospital, Uppsala, Sweden§*

SUMMARY This single fibre EMG study compares the standard method of neuromuscular jitter measurement in voluntarily activated muscle to that by intramuscular electrical stimulation of motor axons in a group of normal subjects. The latter method avoids the interdischarge interval-dependent jitter, as well as a possible failure to recognise split muscle fibres. The mean MCD on axonal stimulation was only 5·2 μs less than in the voluntary activation study and was thus 8% more than theoretically expected for single motor end plates. The difference could be due to an axonal jitter and some other factors. Axonal stimulation has proved to be a relatively easy and reliable method for routine estimation of neuromuscular jitter, provided that the resolution of time measurement is better than 2 μs, so that low jitter due to occasional direct muscle fibre stimulation is not mistaken for a normal reading. The upper normal limits for the extensor digitorum communis muscle suggested by the present study are 40 μs (individual muscle fibres) and 25 μs (mean of 30 muscle fibres).

In single fibre electromyography, the jitter of the motor end plates is usually measured during slight voluntary contraction of the muscle. This however, is not always practicable, for example in unco-operative or unconscious patients, young children and even in severely paretic muscles, whether there is an upper motor neuron lesion or very severe myasthenia gravis or weakness due to some other peripheral pathology. Electrical stimulation has been suggested for such cases and has actually been used in research,[1] but only seldom in clinical practice. Some of the reasons have been lack of relevant experience, the need for a high resolution jittermeter to eliminate the possibility of mistaking the low jitter due to direct muscle fibre stimulation for a normal reading, and unavailability of normal data. The purpose of this work was to collect normal values and compare the technique to the well established method with voluntary activation.

Material and method

Fifteen normal volunteers participated in the study. Their ages were between 17 and 39 years, mean 25 years; there were seven males and eight females, all in good health and without evidence or past history of a neurological disease. The left extensor digitorum communis muscle was used for the study. The subject was seated comfortably in a reclining chair with his or her forearm resting relaxed on a support. The motor point was determined for the part of the muscle extending the third finger with a surface stimulating electrode. A teflon coated monopolar needle with a 1 mm bared tip (Teca MF37) as a stimulating cathode was then introduced into, or just proximal to, the motor point, while another needle of the same type was placed subcutaneously a few cm laterally as an anode. A constant voltage stimulator (Medelec type SC6 stimulator with an IS/V stimulus isolation unit) was used to deliver pulses of 50 μs duration and 25 to 100 V amplitude. The stimulus amplitude was adjusted to a strength which produced small twitches most often visible only as jerking of the needle cathode, and only occasionally as small movements of the middle finger. Then a single fibre EMG electrode was inserted 2 to 2·5 cm distally in the extensor digitorum communis muscle and a position was found from which good recording from responding muscle fibres could be obtained. The stimulus frequency was then raised from 2 or 3 Hz (used in finding the recording position) to 10 Hz and the amplitude was adjusted to about 10–30 V above the threshold for the studied motor unit. At threshold, the jitter was considerable. Gradual raising of stimulus strength from the threshold resulted in a progressive reduction of the latency and of the jitter to a point when further increases produced no change in either the latency or the jitter. Before starting computation, the

Address for reprint requests: Dr J Trontelj, University Institute of Clinical Neurophysiology, University Medical Centre of Ljubljana, Zaloske 7, 66105 Ljubljana, Yugoslavia.

Received 19 July 1985.
Accepted 25 August 1985

stimulus strength was carefully adjusted well above this point. Occasionally, the stimulus used was close to threshold of another motor axon and intermittently recruited action potential of its muscle fibre could partly interfere with the studied spike. Such recordings were naturally discarded. Sometimes, the observed muscle fibre's action potential appeared at two or even three different latencies, with a stable jitter at each, due to an axon reflex mechanism.[2] In such cases, the earliest latency obtained with the strongest stimulus was preferred for the jitter measurement.

The recording equipment was a Medelec MS6 electromyograph and the filters were set to 3·2 KHz for the high pass and 16 KHz for the low pass filter. The rather high setting of the high pass filter increased the accuracy of the jitter measurement by providing a more stable baseline and a profound attenuation of action potentials of the more distant muscle fibres.

The jitter was measured on-line by means of a jittermeter[3] with a resolution of 0·1 μs. The jitter was measured as variation of successive latencies from the stimulus to the selected point on a single muscle fibre action potential. Care was taken to have clear-cut recordings with no superimposed action potentials of other fibres having an independent jitter of their own (evident as changeable shape or amplitude of the spike). The jitter was computed as mean consecutive difference of latency (MCD) from a series of 50 consecutive discharges, and six series of 50 were obtained from each muscle fibre. Only exceptionally the MCDE value (computed after eliminating of data exceeding MCD +4 SD) was also obtained to eliminate the errors due to an occasional disturbing potential.[1 3] The mean latencies were also recorded for each spike. In each subject, 26 to 41 (mean 34·4) muscle fibres were analysed in this way, whereby the position of the stimulating cathode was changed several times. From any one recording position, one or up to five different muscle fibres could be studied, when their spikes were separated well enough to preclude any mutual interference. Often these spikes belonged to different motor axons, which was easily detected by finding different stimulating thresholds. Care was taken in this case to make the stimulus well above threshold for each of the axons,. Occasionally, particularly when a motor unit was observed for a longer period of time, the effectiveness of the stimulus decreased, resulting in an increase in the latency and in the jitter, and necessitating a readjustment of the stimulus. Conversely, an increase in the effectiveness of the stimulus resulted in recruitment of additional, often disturbing motor units. The whole study usually took about one hour, and most of the time it did not cause any discomfort to the subject who often could not feel the stimulus. As regards the electromyographer, the study was considered easier and was performed faster than the jitter study on voluntary activation.

The electrical stimulation jitter study was preceded or followed by a conventional jitter study in the same muscle, about 1–2 cm laterally from the stimulating and recording sites used in the electrical stimulation study. The jitter was measured in 18–35, mean 22·4, muscle fibre pairs as MCD value obtained from five to six series of 50 consecutive discharges.

In both types of the studies, the series with the highest MCD value of any one fibre was discarded and a mean value of the remaining MCDs was computed for each fibre, each subject, and each of the two types of the studies. Also discarded were all muscle fibres with at least one MCD value less than 4·0 μs or 2 values between 4·0 and 5·0 μs. Such recordings were eliminated because they were taken to represent direct stimulation of muscle fibres.

In a few cases, responses from two or three muscle fibres innervated by the same motor axon were recorded on tape for subsequent computation of the jitter to the stimulus as well as between the individual action potentials in the identical sequences of responses. A HP 3960 tape recorder was used at high speed, 15 ips, at which the jitter of the tape did not exceed 1–2 μs over segments of 10 ms. A Tektronix 5113 oscilloscope with "triggerable after delay interval" facility was used in conjunction with the delay line of the MS6 electromyograph to trigger on any selected action potential.

Results

The MCD values in each of the two types of the studies for individual subjects are shown in fig 1. Figure 2 shows distribution histograms, also in comparison to previously published normal material for jitter in voluntarily activated extensor digitorum communis.[1] In any one subject, the mean value of MCDs was lower on axonal stimulation than on voluntary activation (fig 3) and the difference is highly significant ($p < 0·001$). Even when all fibres (pooled data) are compared, the difference is significant at the same level (table 1). As can be seen from fig 3, there is relatively large intersubject mean MCD variability, which however is similar for both types of the study. In other words, subjects with relatively low mean jitter on voluntary activation also tended to have low mean jitter on electrical stimulation and vice versa. In no case was the mean jitter on axonal stimulation larger than

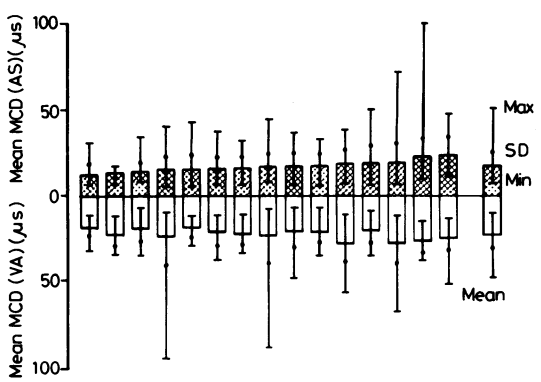

Fig 1 *Graphic representation of the mean MCD values for all recordings from individual subjects arranged in the order of increasing values on axonal stimulation. Each of the columns above the horizontal line indicates the mean value on axonal stimulation (AS) and below the horizontal line the mean value for the same subject on voluntary activation (VA). The bars indicate the ranges of individual values (between dashes) and 1 SD (closed circles). The mean values for all subjects are indicated on the far right.*

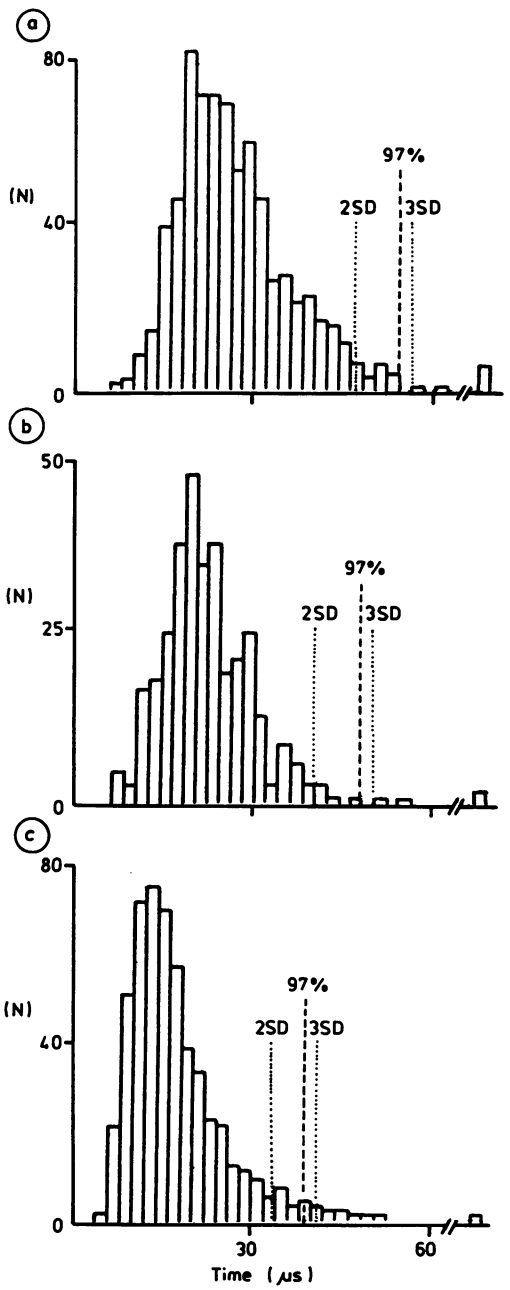

Fig 2 *MCD histograms for individual muscle fibres (or pairs of muscle fibres in case of voluntary activation studies). Top: normal material from reference (1); middle: from voluntarily activated muscle in the present study; bottom: from electrically activated muscle in the present study. Pooled data from all subjects. The vertical lines indicate mean + 2 SD and mean + 3 SD, as well as the 97th percentile limit.*

that on voluntary activation. Variability of the jitter of individual muscle fibres in repeated computations estimated as range of individual MCD values obtained from five consecutive series of 50 discharges was found to be correlated to the mean MCD ($p < 0{\cdot}005$ in 12 subjects on axonal stimulation and in four on voluntary activation). The ranges were slightly narrower on axonal stimulation (mean 5·3 as compared to 6·5 μs), however the difference was not significant.

In one type of experiment, response from two or three muscle fibres innervated by single motor axons were recorded on magnetic tape and their jitter was then measured both to the stimulus as well as between each other in the identical 10 sequences of 50 responses. This measurement was performed in order to compare the jitter between pairs of muscle fibres to the values expected theoretically[1] and computed from the jitter of single fibres using the expression:

$$\text{jitter of pair} = \sqrt{(\text{jitter of end plate 1})^2 + (\text{jitter of end plate 2})^2}$$

One of the actual recordings is shown in fig 5 and the results are presented in table 2.

A proportion of responding fibres (about 10%) had low jitter, that is between 0·9 and 4·4 μs (fig 6). These were taken to represent responses to direct stimulation of the muscle fibres, that is not via the motor

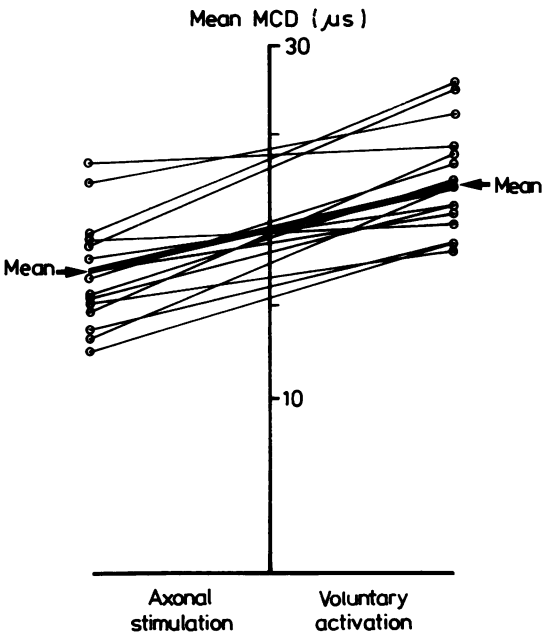

Fig 3 *Lines connecting mean MCD values on axonal stimulation and on voluntary activation in individual subjects illustrate that the latter value was higher in each subject.*

axon and the motor end plate and were not included in the statistics on axonal stimulation. Such responses were not uncommon even with the stimulating needle cathode in the middle of the motor point, but were less frequent when the cathode was moved a few mm proximally. Apart from the low jitter these responses were also characterised by large jitter and considerably longer latency on threshold stimulation, as well as by generally lower resistance to higher stimulation rates. For example, when the rate was raised from 10 to 20 Hz, the latency showed a considerable progressive prolongation, typically for up to 5 or even 10 ms, and large increase in jitter, followed by complete blocking for a short while. The resulting resting period was followed by reappearance of the

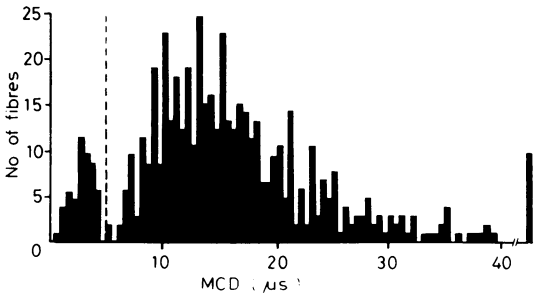

Fig 4 *Histogram of all MCD data obtained with electrical stimulation (569 muscle fibres), including those with low jitter considered to represent direct muscle fibre stimulation. The vertical dotted line indicates the arbitrarily defined border between the axonal and direct muscle fibre responses at 5 μs. Only few data are seen close to the dividing line between the two populations. The direct responses (53 muscle fibres) have a mean MCD of 2·82 μs, SD 0·90 μs. Mean + 3 SD limit would be at 5·5 μs.*

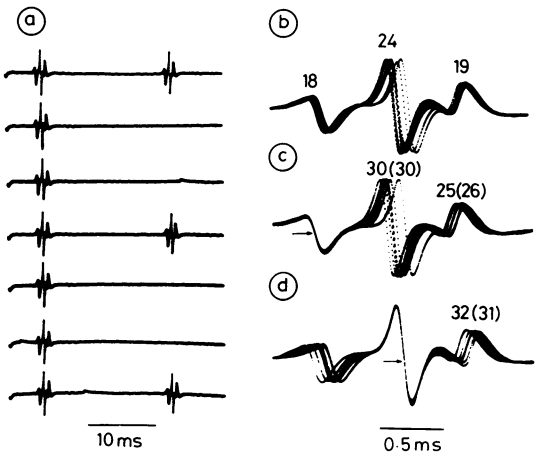

Fig 5 *A recording from three muscle fibre innervated by one motor axon, as proved by the occasional appearance of recurrent responses (A). The inverse polarity of the first two action potentials is due to the use of bipolar recording with a double surface SFEMG electrode. The jitter was measured as shown for each fibre from the stimulus (B) and then from the first (triggering point marked with arrow) to the second and third fibre (C) and from the second to the third fibre (D). The actual values of MCD as well as the theoretically expected values (numbers in brackets) are indicated (see also table 2).*

Table 1 *Comparison of MCD values obtained in 15 subjects on voluntary activation and on axonal stimulation*

	Axonal stimulation	*Voluntary activation*	*p*
Total no of muscle fibres	516	336	
No of muscle fibres per subject			
mean	34·4	22·4	
range	26–41	18–35	
MCD—pooled data/μs/			
mean, SD	17·1, 8·2	22·3, 9·5	<0·001
range	5·0–72·2	6·4–93·2	
MCD—individual subjects/μs/			
mean of mean MCDs	17·2	22·4	<0·001
SD of mean MCDs	3·0	2·9	
range of mean MCDs	12·8–23·4	18·4–27·7	

Table 2 *Jitter of four different multiple potentials belonging to single axons, as measured to stimulus and between the individual single fibre action potentials in identical sequences of 500 responses. The theorectically expected values in brackets*

No of multiple potential	*Jitter measured to stimulus*			*Jitter measured between action potentials*		
	MCDa	*MCDb*	*MCDc*	*MCDa–b*	*MCDa–c*	*MCDb–c (μs)*
1, Triple	18	24	19	30 (30)	25 (26)	32 (31)
2, Double	17	7		20 (18)		
3, Double	11	13		17 (17)		
4, Double	17	12		19 (21)		

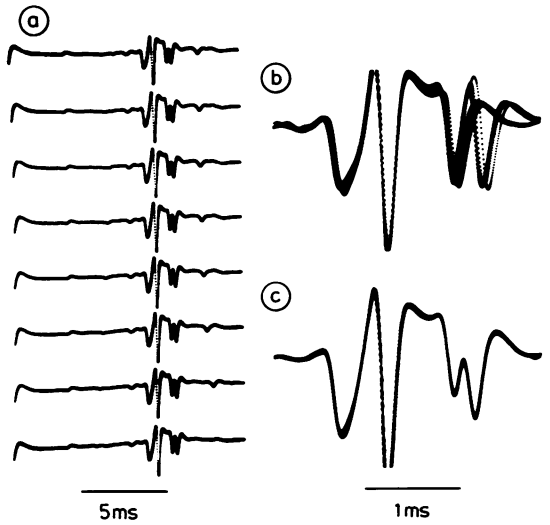

Fig 6 *A recording with low jitter (A). When the stimulus is at or just above the threshold (B), the jitter may be large or, occasionally, in the range of that normally obtained on suprathreshold axonal stimulation, but falls below 5 μs when stimulus strength is raised (C).*

responses, whose latency was usually still prolonged at first but rapidly normalised. After a variable period of relatively stable responses the whole cycle repeated itself. The cycles then tended to become progressively shorter and the periods of block longer. They could be partly overcome by an increase in stimulus strength or decrease of stimulation rate.

Another characteristic of responses with low jitter was that they usually, although not always, occurred at shorter latencies than those with jitter above 5 μs. The MCD values of fibres with low jitter are indicated in table 3 and in the left part of fig 4. The mean MCD value of the 53 muscle fibres believed to be directly activated was 2·8 μs, SD 0·9 μs. The two populations of data in fig 4 appear to be well separated with only a few data near the border of 5 μs which has been suggested as a criterion to distinguish between axonal and direct muscle fibre stimulation in earlier work.[1] However, some of these fibres showed an intermittent increase of their jitter, usually between 5 and 15 μs, which lasted for a few seconds to a few tens of seconds, was not associated with any change of the mean latency, and could not be reversed by increase in stimulus strength.

Table 3 *The jitter in directly stimulated muscle fibres*

No of fibres	53
Mean latency	7·01 ms
Mean MCD	2·82 μs
Range of MCD	0·9–4·4 μs
SD of MCD	0·9 μs

Two kinds of more exceptional phenomena in fibres with low jitter have to be mentioned. In some fibres there was a progressive drop of action potential amplitude and increase of its duration which occurred gradually during prolonged periods of stimulation at intermediate rates (for example 10 Hz) but much faster at higher rates. These changes were reversible after rest but not by an increase in the stimulus strength. An exceptional type of recording was from two muscle fibres of different subjects which showed large jitter and blocking at low stimulation rates (3 or 5 Hz), not reducible by increases in stimulus strength, but becoming low (<5 μs) on higher stimulation rates. These recordings were believed to be due to ephaptic driving of muscle fibres by other fibre action potentials. The fibres with these two phenomena were not included in any analysis.

On axonal stimulation, the latencies of the responses of different muscle fibres varied considerably, from as early as 2 ms to as late as 16 ms. From a given recording electrode position, however, the multiple spike recordings were not usually dispersed so greatly. A small change of the recording position, by about one or two mm, frequently produced responses grouped at much shorter or considerably longer latencies.

There was no definite correlation between the latency and the jitter on axonal stimulation, in individual (with the exception of two) data; however, a significant correlation was found in the pooled data ($r = +0{\cdot}29$, $p < 0{\cdot}001$). There was no significant correlation between the mean interpotential interval and MCD on voluntary activity. There was also no correlation between the jitter of directly activated muscle fibres and their latency.

Discussion

THEORETICAL BACKGROUND

Aims of the study

The estimation of the end-plate jitter by single fibre EMG is a sensitive test of neuromuscular transmission, with which it is possible to detect even minor defects, for example in clinically unaffected muscles of myasthenic patients. Furthermore, even the values within the normal range have a physiological significance, since the magnitude of the jitter is correlated to the safety factor of neuromuscular transmission, as has been demonstrated by experiments with blocking agents.[5]

Usually the jitter is measured between action potentials of two muscle fibres of the same motor units firing in voluntarily activated muscle. One of the

two action potentials is used to trigger the oscilloscope and serves as a time reference from which the interpotential interval (IPI) is measured to a selected point on the other action potential. Under these conditions, the measured IPI variability, the jitter, consists mostly of the variable neuromuscular transmission time across the two end plates, whereby the contribution of each is unknown. Other possible sources of IPI variability include variation of conduction times along the two axonal branches and along the two muscle fibres from the end plates to the recording electrode. This variation is due to changing axonal and muscle fibre conduction velocities following preceding discharge. Even at rates of about 10 Hz there is some residual subnormality of the axon and subnormality, or, more often, supernormality of the muscle fibre. If the consecutive interdischarge intervals (IDI) are constant, the next discharges fall in the identical points of the recovery function, and the resulting change in the conduction times, if any, is a smooth function, the influence of which upon the jitter is largely eliminated by expressing it as mean consecutive difference (MCD) instead of standard deviation.[1] Theoretically, even with nonuniform firing rates, no additional jitter is expected, if the lengths of the two axonal branches and the muscle fibre segments to the electrode are equal and if the recovery functions are identical. In practice they are not; however at relatively even firing rates and with IPI less than 4 ms (which is true of the great majority of recordings obtained in the usual way), the contribution of axonal and muscle fibre jitter due to unequal recovery functions is assumed to be small. However, a systematic study of this contribution has so far not been available.

One of the aims of the present work was to study jitter under conditions when the axonal and muscle fibre jitter due to their recovery functions may be assumed to be largely eliminated, that is, at constant interdischarge intervals. The other aim was to assess jitter measurement by axonal stimulation as a practicable routine test of neuromuscular transmission.

TECHNICAL CONSIDERATIONS OF AXONAL STIMULATION

The introduction of regular IDI is aimed at minimising any axonal or muscle fibre jitter due to their recovery functions (it may not eliminate a jitter due to weak points in the pathway of the impulse, such as presumably occur in regenerating axons or damaged muscle fibres). However, could stimulation itself give rise to some additional jitter? On threshold stimulation there is a jitter of stimulation site which may be much larger than that of a normal motor end plate. One of the sources is a large jitter of the start of the propagated nodal potential as it arises from the local nodal response. The other source is uncertainty as to the node at which the propagated response starts. On suprathreshold stimulation the contribution from both sources appears to be minimal, at least less than 2–4 μs as judged from the very low jitter of a single axon potential recorded with microneurography from the human median nerve with intrafascicular stimulation (Trontelj, unpublished observation). Great care was taken to avoid stimulation just above the threshold since the efficiency of the stimulus tends to change slightly even with most carefully controlled conditions.

A further uncertainty may be introduced by the shape of the stimulus itself. In tissues, the rectangular stimulus pulse becomes distorted owing to the resistive and capacitive properties. The use of a rather short pulse, just 50 μs, is considered to reduce the likelihood of the response being triggered by the quite slowly rising slope near the top of the pulse and increase the possibility of fine adjustment of the amplitude. At the same time, thresholds of axons within the reach of the stimulating cathode are more widely separated with short stimulating pulses. Thus it is easier to stimulate the selected axon well above its threshold and have the recording undisturbed by near-threshold stimulation of other axons.

A stimulator with fine adjustment of the stimulus amplitude is considered necessary since the range between the minimum safe suprathreshold value and the threshold strength for other axons disturbing the recording is often narrow.

The bimodal jitter due to the axon reflex mechanism is usually easily seen and its cause identified by small changes in stimulus strength.[2] It may however be difficult to detect occasionally when the jumps between the earlier and the later latencies are rather short, exceeding only slightly those of the end-plate jitter. This further demonstrates the need for careful individual adjustment of stimulus amplitude for every studied axon.

High resolution jitter measurement is required in order to distinguish between normal end plate jitter and low jitter of directly stimulated muscle fibres. The resolution should be in the range of 1 or maximum 2 μs. Manual measurement from paper recordings, in the best circumstances, just allows such resolution. The resolution of 0·1 μs used in this study made it possible to accurately determine the normal range of jitter of directly stimulated muscle fibres, but is not indispensable for routine diagnostic studies.

Comment on results

The jitter in 336 pairs of muscle fibres measured during voluntary activity is slightly, although significantly ($0{\cdot}001 < p < 0{\cdot}005$) lower than the published normal material.[3] The probable reasons

include the young age of most of our subjects (mean 25 years) and the selection of data (elimination of the highest MCD of the six consecutive series of 50 discharges in each potential pair), as well as careful control of recording conditions, especially uniformity of discharge rate.

The jitter measured between two muscle fibres includes random variation of transmission times across two motor end plates, and theoretically equals:

$$\text{jitter of pair} = \sqrt{(\text{jitter of end plate 1})^2 + (\text{jitter of end plate 2})^2}$$

The jitter of an average single motor end plate should thus be

$$\text{jitter of single end plate} = \sqrt{\frac{(\text{jitter of pair})^2}{2}}$$

In our study, theoretically expected jitter on axonal stimulation should thus be

$$\text{Mean MCD (axonal stim)} = \text{Mean MCD (vol activ)}/\sqrt{2} = 15{\cdot}8\,\mu s$$

The actual result was 17·1 μs, that is 1·3 μs or 8% higher than the theoretically expected value. The difference is small, although statistically significant. The fact that it is small indicates that the following factors theoretically accounting for it are of relatively minor importance:

(1) Jitter of the stimulated node of Ranvier and the axon distal to stimulation site, which is estimated at about 1–3 μs. In the pooled data, there was a significant correlation ($r = +0{\cdot}29$, $p < 0{\cdot}001$) between the latency and MCD, which suggests that longer (or thinner) axons may also contribute a jitter, although small, probably not exceeding 5 μs. Alternatively, and more likely, the end plates supplied by thinner axons may have slightly larger jitter. On the other hand the longer latency may partly be due to a longer delay at the end plate, because of a less steep end plate potential. Such end plates have a lower safety factor and large jitter.[1]

(2) Occasional appearance of the recurrent response introduces a disturbance in the IDI intervals, often producing a large latency change of the following response (due to supernormal muscle fibre propagation velocity) and thus increasing the calculated MCD.

(3) In the case of voluntarily activated pairs of muscle fibres, the randomness of variation of neuromuscular transmission time may not be completely independent. In other words, there may be a degree of co-variance, for example due to a summation effect of the electrical field of the passing action potential of the first fibre with the end plate potential of the second fibre. This would clearly tend to reduce the combined jitter measured between the two fibres' action potentials.

(4) The two types of studies probably did not involve identical populations of motor units. Low grade voluntary contraction preferentially activates low threshold small motor units, while electrical stimulation most likely activates both small and large ones (electrical stimulation of the nerve trunk preferentially activates large motor axons, however our needle cathode was placed near an intramuscular nerve bundle in which most of the axons were already branched; furthermore, the proximity of the needle is presumably more critical than the actual electrical threshold of the individual axons for their respective order of recruitment in the elicited response). The large motor units may have a different (greater?) jitter than the small ones.

The experiment in which the jitter was also computed between individual muscle fibres of a stimulated motor axon showed remarkable similarity between the obtained values and the theoretically expected values (table 2), which in the average differed by only 1 μs. If this finding, observed on a rather small sample of nine muscle fibres, may be taken to represent a general rule, then it seems to suggest (1) that the jitter of the stimulated site and of the distal portion of the axon is less than 1–2 μs, and (2) that the jittering of the different end plates in the motor unit follows an independent random pattern.

Thus, although the IDI dependent jitter may be assumed to have been eliminated, the jitter on axonal stimulation is nevertheless not smaller but is slightly larger than theoretically anticipated from the values in voluntarily activated muscle fibre pairs.

In the normal muscle, the IDI dependent jitter does not appear to constitute any significant proportion of the total jitter in voluntarily activated muscle fibre pairs. This impression is also supported by the fact that trial-to-trial variability of MCD in the same muscle fibres at different degrees of variability in discharge rate was not significantly larger on voluntary activation, although some of the six series did contain considerable IDI variation. Also, there was no consistent correlation between the mean interpotential interval (MIPI) and the magnitude of the jitter.

The present study showed considerable inter-individual differences in jitter, their mean MCD ranging from about 13 to 23 μs on axonal stimulation and from 18 to 28 μs on voluntary activation. As shown in fig 3, there was a fairly good correspondence between the relative magnitudes of jitter on either type of activation. This is in agreement with previous observations which led to the contention that the magnitude of jitter, even when within the normal range, reflects the safety factor of the neuromuscular transmission, and thereby also to the concept of "high" and "low safety" individuals.

Responses with low jitter

The responses with MCD below 5 μs also had other characteristics on the basis of which they could be recognised as being produced by direct stimulation of the muscle fibres. These included very large jitter and much longer latency on threshold stimulus, and smooth decrement of both on gradual increasing of stimulus strength. At higher stimulation rates, the latency gradually lengthened, typically by a few milliseconds, which was followed by blocking and, after a fraction of a second, by restoration of the response at the original (or somewhat longer) latency, after which the whole sequence repeated itself. This behaviour is presumably due to the recovery cycle of the stimulated site on the muscle fibre. Responses with the same characteristics are also seen in completely denervated muscle.[1 4] Similar behaviour, except for low jitter, is also seen with responses to axonal stimulation, but is usually much less pronounced.[1] The stimulating threshold of muscle fibres was in the same range as that of the axons, and the type of responses obtained seemed to depend mostly on position of the stimulating cathode. Occasionally both types of responses were elicited at one stimulation and recording site and even their latencies could partly overlap, although those of the direct responses tended to be shorter. Direct responses were quite often obtained while stimulating in the motor point. It is concluded therefore that low jitter is the most reliable criterion for differentiation between the direct muscle fibre and axonal stimulation, which is also substantiated by the combined MCD histogram of both types of responses in fig 4 showing virtually no overlapping. Thus the 5 μs limit seems to be adequate, but it should be borne in mind that short episodes of temporary increase of jitter to between 5 and 10, rarely up to 15 μs, may occur in some fibres for unknown reasons. This implies that for reliable differentiation, responses with jitter between 5 and 10 μs have to be observed for at least half a minute.

Computation of muscle fibre propagation velocity just from the latency and the distance between the stimulating cathode and the recording needle electrode may not be quite accurate, since the actual starting point of the propagated action potential along the muscle fibre is unknown, however the error probably does not exceed 5–10%. The values obtained range from 2·4 to 6·2 m/s, mean 4·5, SD 1·2 m/s, which is similar to the values obtained by calculating propagation velocity across a multielectrode.[5]

Responses with large jitter

When axonal stimulation is used in a diagnostic study and an action potential with a large jitter is recorded one should first exclude a possibility of threshold stimulation. If after an increase in stimulus strength the jitter remains large it can be considered most likely to result from an abnormal motor end plate (an exception is the rare case of ephaptically driven muscle fibre, which however can be identified by increasing the rate of stimulation, for example to 20 Hz; this will usually dramatically reduce the jitter and stop the blocking).

In a patient with a neuromuscular transmission problem, axonal stimulation study has the advantage of easy differentiation between myasthenia gravis type and Lambert-Eaton syndrome type of involvement of the motor end plate. In this case the initial stimulation rate should not be as high as 10 Hz, but rather 3 or 5 Hz. When abnormal jitter is recorded, the rate is increased to 10 Hz. A reduction of jitter would then indicate the Lambert-Eaton type of abnormality, while in the case of myasthenic involvement the jitter and degree of blocking increase further.

Conclusions

In conclusion, electrical stimulation of intramuscular motor axons is a practicable method of studying the motor end plate jitter in routine clinical work. A sample of 30–40 motor end plates can be accurately evaluated in about one hour, which is similar to the time required to evaluate 20–30 potential pairs in voluntary activation study. The advantages of the electrical stimulation include perfect control of discharge rate and little need for co-operation of the patient. The additional discomfort of electrical stimulation is minimal and a majority of our subjects liked it better than the voluntary activation study. Thus it is suitable for clinical evaluation of the neuromuscular jitter particularly in patients who for any reason are unable to cooperate. It may also prove useful in animal studies. The mean jitter measured on axonal stimulation is on the average about 5 μs lower than that obtained on voluntary activation but the two studies are in good correlation.

The upper normal limit of MCD on axonal stimulation suggested by the present study is 40 μs for individual extensor digitorum communis motor end plates (close to 97th percentile, that is with one value out of 30 allowed to exceed this limit). The upper normal limit of mean MCD in a sample of 30 motor end plates in a subject is 25 μs (which is about 3 SD above the mean). The corresponding limits for voluntary activation in this study are 51 and 30 μs. This is slightly lower than the previously published normal material, presumably due to more rigid control of the stability of innervation rate and perhaps to the relatively young mean age of the volunteers in the study.

The present study suggests that only a small part of the total jitter measured between a pair of voluntarily

activated muscle fibres is contributed by an IDI dependent variability due to the axon and muscle fibre recovery functions in the normal muscle. It may be considerably larger in pathology, such as muscular dystrophies and other conditions with long interspike intervals. In these cases voluntary activation is liable to produce false abnormal readings, which cannot even be completely avoided by computing MSD (mean sorted difference) instead of MCD, since the former only eliminates the effect of completely stochastic IDI variation, but not that of short trends.[1] Axonal stimulation however would give correct results. Furthermore, it can reveal low jitter between split muscle fibres with long interpotential interval, where voluntary activation study would produce an IDI dependent jitter and thus obscure the low jitter phenomenon. Naturally in this case the jitter must be measured between the two spikes.[6]

This study was supported by the Slovene Research Council, Grant No. C3-0178-306-85 and by the Swedish Medical Research Council, Grant No. 305.

References

[1] Stålberg E, Trontelj JV. *Single Fibre Electromyography*. Old Woking, Surrey: Mirvalle Press, 1979:1–244.

[2] Stålberg E, Trontelj JV. Demonstration of axon reflexes in human motor nerve fibres. *J Neurol Neurosurg Psychiatry* 1970;**33**:571–9.

[3] Trontelj JK, Mihelin M, Pleteršek T, Antoni L. Jittermeter: a microcomputer-based system for single fibre electromyography. *Int J Biomed Comput* 1979;**10**: 451–9.

[4] Trontelj JV, Stålberg E. Responses to electrical stimulation of denervated human muscle fibres recorded with single fibre EMG. *J Neurol Neurosurg Psychiatry* 1983; **46**:305–9.

[5] Stålberg E. Propagation velocity in human muscle fibres in situ. *Acta Physiol Scand* 1966;**70 suppl 287**:1–112.

[6] Hilton-Brown P, Stålberg E, Trontelj JV, Mihelin M. Axonal stimulation in dystrophic muscle. *Muscle Nerve* 1985;**8**:383–8.