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RESEARCH ARTICLE

# A SIMULATION STUDY ON NEUROMUSCULAR FACTORS AFFECTING CONSECUTIVE MOTOR UNIT ACTION POTENTIAL WAVESHAPE

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## ABSTRACT

Quantification of consecutive motor unit potential (MUP) is used to diagnose and monitor the progress of neuromuscular pathologies in clinical applications. In this study, a detailed motor unit simulation was conducted to reveal and understand the factors affecting MUPs. Using a volume conductor model and real muscle parameters, normal and pathological MUPs were created. The shape changes observed in consecutive MUPs, called jiggle, are calculated with a quantification method. Increased jitter duration and re-innervation percentage commonly observed during motor unit loss increase the jiggle value proportionally. Moreover, increasing fiber density changing different regions of a muscle bundle decreases the jiggle value. The blocking phenomena generally observed in re-innervated fibers affects the jiggle value similar to jitter duration. But, higher blocking levels (50%) of re-innervated motor fiber do not have an effect on jiggle value as lower levels of blocking (20%). In conclusion, simulation of pathological MUPs showed that it is useful for clinicians to understand the progress of a neuromuscular pathology and the factors affecting consecutive MUP wave shape.

**Keywords:** Motor Unit Action Potential, Neuromuscular Pathology, Motor Unit Simulation

## 1. INTRODUCTION

The analysis of motor unit potential (MUP) recorded during voluntary muscle contraction provides important information to help in the diagnosis and characterization of neuromuscular pathology. Besides the analysis of such classical parameters as duration and amplitude, the degree of change in MUP shape at consecutive discharges can be analyzed [1]. This variability depends on the behavior of the multiple SFAPs of MUP, which is seen in single-fiber electromyography (SFEMG). The variability of the intervals in SFAPs, which is called the jitter, is between 10-30 μs in normal muscles [2]. However, the jitter is increased for disturbed neuromuscular transmission such as in early re-innervation and myasthenic disturbances. If there are more severe disturbances, some SFAPs can be lost as a result of an intermittent failure of transmission in the motor unit (MU) endplate. All these disturbed neuromuscular transmission conditions cause instability or variations in consecutive MUPs and this is called "jiggle" by Stålberg and Sonoo [3].They proposed a method to express the quantification of shape variability and defined two parameters: the normalized mean of median consecutive amplitude differences (CAD) and the median of the cross correlational coefficient of consecutive discharges (CCC). The mathematical expression of CAD and CCC is [4]:

$$CAD = \frac{\sum_{t=1}^{n} \left\{ median \begin{bmatrix} |y_1(t)-y_2(t)|, |y_2(t)-y_3(t)|, \ \dots, \\ |y_{m-1}(t)-y_m(t)| \end{bmatrix} - C \right\}}{\sum_{t=1}^{n} |y(t)|} \quad (1)$$

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$$CCC = median\left\{ \frac{\sum_{t=1}^{n}[y_1(t)-\bar{y}_1][y_2(t)-\bar{y}_2]}{\sqrt{\sum_{t=1}^{n}[y_1(t)-\bar{y}_1]^2}\sqrt{\sum_{t=1}^{n}[y_1(t)-\bar{y}_1]^2}}, \ \ldots, \ \frac{\sum_{t=1}^{n}[y_{m-1}(t)-\bar{y}_{m-1}][y_m(t)-\bar{y}_m]}{\sqrt{\sum_{t=1}^{n}[y_{m-1}(t)-\bar{y}_{m-1}]^2}\sqrt{\sum_{t=1}^{n}[y_m(t)-\bar{y}_m]^2}} \right\} \quad (2)$$

where $\boldsymbol{y_i(t)}$ = amplitude of $i^{th}$ waveform at time $t$, $\boldsymbol{m}$ = number of waveform, $\boldsymbol{n}$ = number of sample in a waveform, $\boldsymbol{C}$ = noise calculation parameter and $\boldsymbol{\bar{y}_i}$ is the mean of $\boldsymbol{y_i}$.

For the calculation of jiggle parameters, a total 5 ms analysis window centered at the maximum negative peak was used from 30 ms MUP trace. CAD is actually the abbreviation of "normalized mean of median consecutive amplitude differences" and it expresses the ratio between the area of amplitude difference of the MUP waveform at consecutive discharges and the area of the averaged MUP (Figure 1).

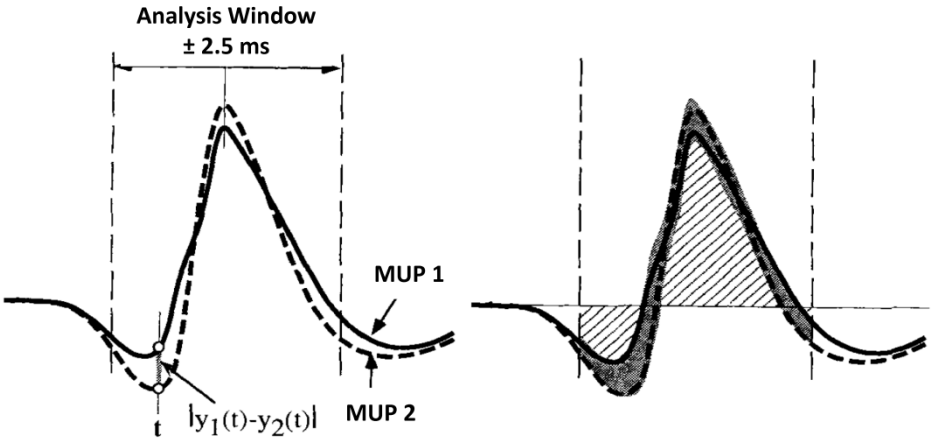


**Figure 1.** Jiggle calculation schematic representation [3]

The method and the mathematical functions were based on simulation studies and tested with real electromyographic signals [3, 4]. The alignment of the waveforms, the choice of the reference point and the sensitivity of the method were tested with real electromyographic signals. Furthermore, for the biological and the technical noise, the segment of 5 ms nearest to the right endpoint in each trace was used to get $\boldsymbol{C}$ value using the points contained in an interval of ±20 % of the acquisition gain (Figure 2). $\boldsymbol{C}$ was designed here to compensate selectivity for smooth baseline fluctuations by excluding the activity from nearby MUPs (recruited MUPs other than the analyzed ones) [4].

The relationship between the jiggle and the jitter, the temporal dispersion of the waveforms were also tested with simulations [3]. Simulation studies indicated that the jiggle assessed by this method is proportional to the jitter of the SFAPs. But the effect of blocking phenomena, percentage of re-innervated fibers and the fiber density on the shape variability of the MUP has not been tested with simulation. For this purpose, a detailed muscle bundle was simulated with real muscle parameters to reveal the effects of blocking, re-innervation and fiber density.

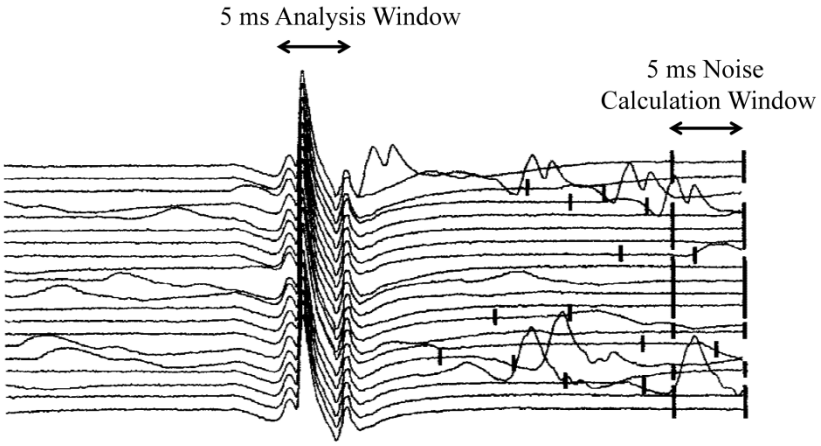


**Figure 2.** Selection of analysis and noise (C) calculation window [4]

## 2. MATERIALS and METHODS

It is impossible to experimentally change the MU properties of a living muscle. Hence, MUP simulation is essential to understand, quantitatively express and interpret the shape changes observed in consecutive MUPs. For this reason, a MUP simulation was created based on the MU parameters obtained by previous studies [5-9].

### 2.1. Simulation Parameters

The muscle fiber density, muscle fiber length, number of muscle fibers and neuromuscular junction (NMJ) positions in a MU were set to create specific MUP to the specific MUs in each muscle group. In addition, the number of healthy and pathological or reinnervated fibers in a MU was set as variable. The jitter value, NMJ delay, was set as 20µs for normal muscle fibers and 20-300µs for pathological muscle fibers [10]. The concentric needle electrode was placed at the midpoint of the distance between the NMJ and the muscle fiber end (tendon connection) of the MU. Standard muscle and fiber parameters used in simulation are given in Table 1.

**Table 1.** Parameters used in simulation model [7]

| Type | Parameter | Interval | Current value |
|---|---|---|---|
| Motor Unit | Number of Muscle Fiber | 0-1000 | 150 |
| Motor Unit | Diameter | Size | 2.5 mm |
| Motor Unit | Minimum Contraction Rate | 0-10 | 5 |
| Motor Unit | Maximum Contraction Rate | 10-100 | 40 |
| Motor Unit | Fiber Density (fiber/mm$^2$) | 1-10 | 5 |
| Motor Unit | Contraction | % 0-100 | % 30 |
| Muscle Fiber | Radius | 20-90 µm | 50 µm |
| Muscle Fiber | Radius Distribution | ± 30 µm | ± 5 µm |
| Muscle Fiber | NMJ Position (From Center) | -20 - 20 | 0 mm |
| Muscle Fiber | NMJ Position Distribution | ± 20 mm | 2 mm |
| Muscle Fiber | Latency | 0-1000 µs | 500 µs |
| Muscle Fiber | Jitter | 0-300 µs | 20 µs |

### 2.2. Formulation of MUP

In this study, the volume conductor model was used to create SFAP [11]. The muscle fiber volume conductor model is generally expressed as the convolution of the transmembrane current of a

cylindrical muscle fiber and the electrode transfer function. The muscle fiber is assumed to be straight and cylindrical. The extracellular environment is assumed to be infinite with cylindrical anisotropy. The origin of the cylindrical coordinate system is in the cross section of the fiber in the end plate (NMJ) and in the center of the fiber (Figure 3A). The action potential is formed in the endplate and travels along the muscle fiber in both directions as a depolarization wave and ends in the tendon. Depolarization wave in a muscle fiber has to flow through the membrane and transmembrane current density is proportional to the second derivative of the intracellular potential [12]. Therefore, it can also be thought that the transmembrane current originates from the endplate and spreads towards the tendons.

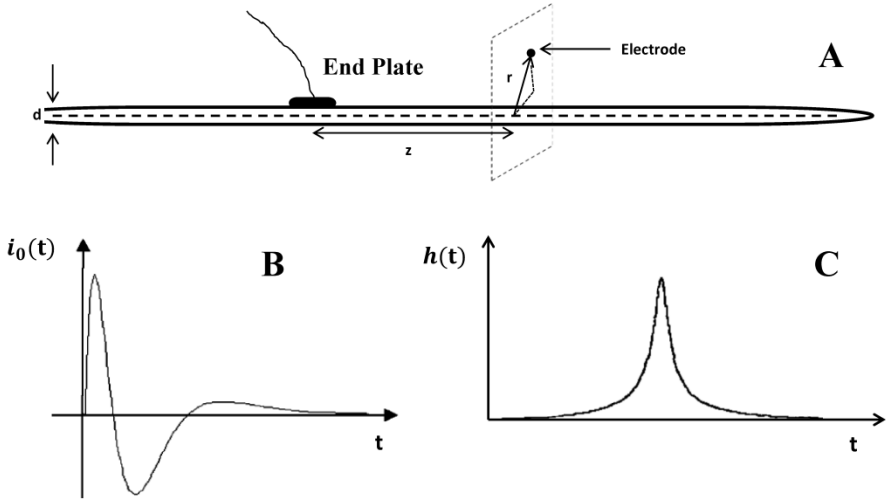


**Figure 3.** SFAP formation at the electrode. A. Schematic representation of muscle fiber and electrode position. B. Transmembrane current waveform in a muscle fiber. C. Electrode transfer function [6].

The end plate (z = 0) current, $i_0(t)$, as shown in Figure 3B and a potential generated by the point current source located at the beginning of the cylindrical coordinate system is expressed by a mathematical equation [13];

$$P(r,z) = \frac{1}{4\pi\sigma_r}\left(\frac{I}{\sqrt{\frac{\sigma_z}{\sigma_r}r^2+z^2}}\right) \tag{3}$$

where $I$ = Intensity of the current source, $\mathbf{r}$ = radial distance, $\sigma_r$ = extracellular conductivity (0.063 $Sm^{-1}$) and $\sigma_z$= intracellular conductivity (0.33 $Sm^{-1}$).

It is assumed that the unit current source emerges in the end plate at t=0 and moves towards the tendons at a constant speed, the potential generated at the electrode by this source is $h(t)$ (Figure 3C). If $i_0(t)$ (Figure 3B) is divided into $n$ sources of different amplitude, the first source will appear at $t = 0$ and each subsequent source at the interval $\Delta t$. The amplitudes of these sources can be expressed as $a_1, a_2, \ldots a_n$. The first current source emerging at $t = 0$ propagates and reaches the tendons and creates $a_1.h(t)$ potential at the electrode. The potential created by the second current source at the electrode will be $a_2.h(t-\Delta t)$. So, the total potential formed at the electrode can be expressed as;

$$SFAP(t) = a_1.h(t) + a_2.h(t-\Delta t) + \ldots + a_n h(t-(n-1)\Delta t) \tag{4}$$

where $\Delta t$ approaches to zero, $n$ approaches to infinity.
$h(t)$ and $i_0(t)$ are expressed as convolution (*) as follows.

$$SFAP(t) = i_0(t) * h(t) \quad (5)$$

As a result, SFAP can be expressed as the output of a linear system whose input is transmembrane current, $\boldsymbol{i_0(t)}$, and stimulus response, $\boldsymbol{h(t)}$.

MUP of a MU now can be expressed as summation of SFAP waveforms (8) created by muscle fibers and mathematical formulation of MUP;

$$MUP(t) = \sum_{i=1}^{N} SFAP_i\,(t - \tau_i).s_i \quad (6)$$

$\boldsymbol{MUP(t)}$: The potential of the MU, $\boldsymbol{SFAP_i(t)}$: SFAP of the $i^{th}$ muscle fiber, $\boldsymbol{N}$: the number of muscle fibers, $\boldsymbol{\tau_i}$: delay of $\boldsymbol{SFAP_i(t)}$ to the recording point and $\boldsymbol{s_i}$: randomly assigned value of 1 or 0 representing blocking of muscle fiber.

### 2.3. Pathological MUP Simulation and Jiggle Calculation

While simulating consecutive MUPs, firstly, a muscle bundle containing the characteristics of a certain muscle group (fiber density, number of MUs, the number of muscle fibers contained in a MU, muscle fiber length, etc.) was created. The MUs and muscle fibers within this muscle bundle were randomly positioned within anatomical boundaries. Next, the probabilities of jitter generation or blocking of muscle fibers in these MUs were randomly adjusted. The electrode position was placed at a point between the NMJ and the tendon after the concentric electrode parameters [6] were adjusted. Then, the neurons in the MUs forming the muscle bundle were stimulated at a frequency of 6-8 Hz, and sequential MUPs were created.

To observe the effect of jitter on CAD and CCC values, MUs with 10% and 20% re-innervation rate were created. In addition, the probability of blocking of reinnervated muscle fibers was assigned as 0%, and the jitter level of these reinnervated muscle fibers was changed between 50-300μs in steps of 50μs. For normal muscle fibers, the jitter level was set as 20μs. After the MUP waves were created, 30dB intensity white noise was added, which is the most common background noise level in needle electrode recordings in the clinic.

When a motor nerve is severely damaged or lost its function, it cannot innervate the muscle fibers it is attached to. In this case, the neighboring motor nerve begins to form new NMJs by extending new terminal ends to the non-stimulated muscle fibers and it is called re-innervation, the stimulation of the muscle fiber also takes longer time than normal because the NMJ connection is not fully formed [14]. The number of reinnervated muscle fibers in a MU will also change the shape of MUP. In order to reveal the effect of re-innervation on the jiggle value, the percentage of re-innervated muscle fibers was changed between 10% and 50%, and pathological MUP waves were created. While forming pathological MUP waves, jitter levels of re-innervated muscle fibers were determined as 100, 200 and 300μs.

In the process of re-innervation formation, NMJ formation (the connection of motor neuron terminal end and muscle fiber) takes time. During this time, when there is no complete fusion in NMJ, some of the consecutive stimuli cannot create an action potential in the muscle fiber [15]. Therefore, while some of the muscle fibers in a MU form SFAP, some cannot form, and in this case, which is called blocking, shape change occurs in consecutive MUP waves. In order to reveal the effect of the blocking percentage on the jiggle value, the percentage of blocking of re-innervated muscle fibers was changed between 10% and 50%, and pathological MUP waves were created by simulation. While forming pathological MUP waves, jitter values of reinnervated muscle fibers were set as 20, 100, 200 and 300μs. The amount of re-innervated muscle fibers was set as 20% of all muscle fibers for the

pathological MUs. For normal muscle fibers, jitter value was determined as 20μs, re-innervation rate 1% and blockage probability 1% [16].

The number of muscle fibers per unit area varies according to muscle groups or muscle bundle regions. Changing the muscle fiber density will increase the number of muscle fibers in the recording area of the concentric electrode. Moreover, with increasing muscle fiber density, MUP amplitude also increases [5]. Therefore, normal and pathological muscle models with different muscle fiber density were created to reveal the effect of muscle fiber density on jiggle parameters. In order to simulate the human muscle structure appropriately, the simulation was performed by changing the muscle fiber density between 5-30 fibers/mm$^2$ [17]. During the simulation, 20% of all muscle fibers were set re-innerved. Also, 10% of these re-innerved muscle fibers were assigned to have the possibility of blocking. Pathological MUP waves were obtained by assigning the jitter probabilities of re-innervated muscle fibers as 150μs, and jiggle parameters were calculated using equation 1 and 2.

## 3. RESULTS and DISCUSSION

Discrimination of normal and pathological consecutive MUPs can be easy for a clinician by looking at the shape of the potentials but quantification of shape variability of MUP waveforms provides accurate information about the examined MU.

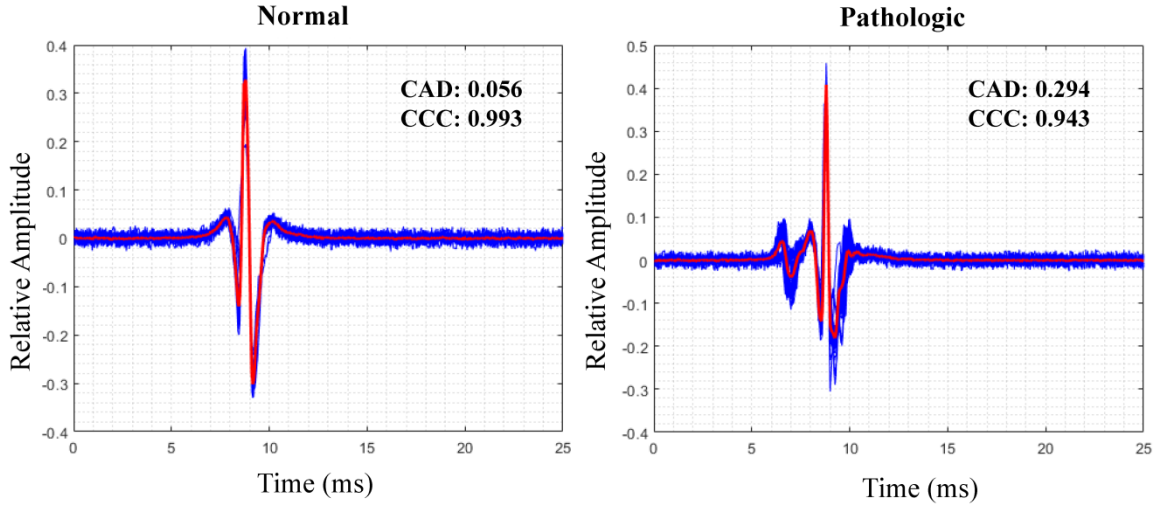


**Figure 4.** Simulated normal and pathological MUP waveforms. For normal MUP: Jitter value is 20μs, re-innervation ratio is 1% and blocking ratio of re-innervated fibers is 1%. For pathological MUP: Jitter value is 100μs, re-innervation ratio is 10% and blocking ratio of re-innervated fibers is 10%. Red waves are the mean of the consecutive MUPs.

As can be seen in Figure 4, the CAD value of the normal MUP wave is 0.056 while pathological MUP wave is 0.294. Also, the CCC values representing the cross-correlation between consecutive MUP waveforms support the CAD values. If there is no difference between the consecutive potentials CCC value should be 1. But, increasing difference between potentials will decrease the CCC value. In Figure 4, while CCC value is 0.993 for normal MUP waveforms, it drops to 0.943 for pathological MUP because of the increasing consecutive wave shape difference.

With increasing jitter duration, jiggle value (CAD) increases significantly for the case of 10 and 20 % of re-innervation as expected (Figure 5). The decrease in CCC with increasing jitter duration supports the acquired jiggle values. Increasing jitter value assigned to the SFAPs causes shape variability in consecutive MUPs because of the temporal dispersion between randomly created SFAPs. There is an increase between 20 and 50μs jitter durations for the 10% re-innervation rate but it is not significant.

With these results, increasing level of a neuromuscular pathology affecting NMJ or re-innervation rate of a MU will cause a directly proportional increase in CAD value.

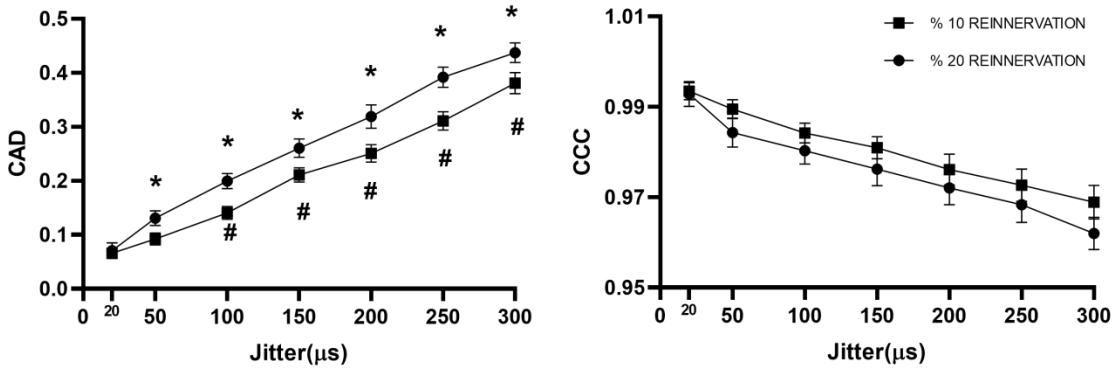


**Figure 5.** Simulation of the relationship between jiggle and jitter. Bars represent the SEM values. One-Way ANOVA (Dunnett) test was used for significance. Each jitter group is compared with following increased jitter group. (*: $p<0.05$, #: $p<0.05$)

When the re-innervated fiber percentage in a MU increased, calculated jiggle values increase significantly for all jitter durations (Figure 6). It is assumed that since the increased re-innervation percentage will cause new unstable NMJ formation, it is inevitable to have high jitter duration in MUs. The jitter duration, which was considered 300μs in the first stage of NMJ formation, will decrease to 100μs or lower as the NMJ connection becomes more stable [18]. Therefore, the obtained results support the assumption mentioned above because jiggle values for 100μs jitter duration (more stable NMJ) are smaller than 300μs jitter duration (unstable NMJ).

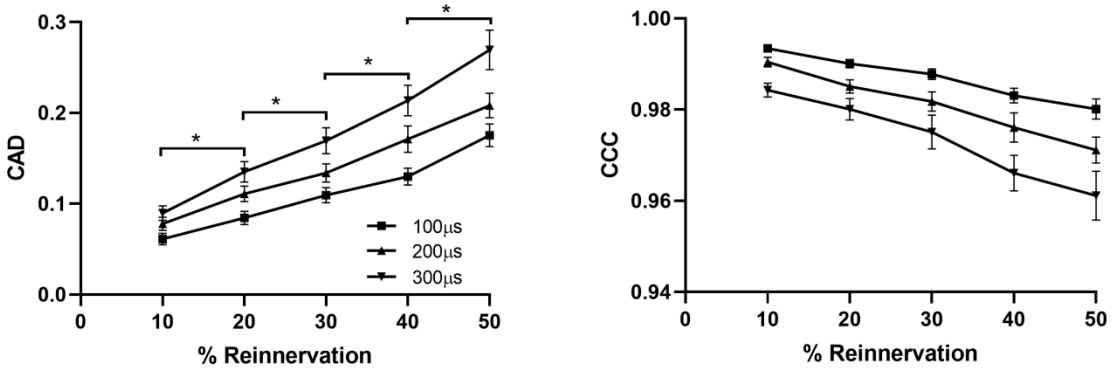


**Figure 6.** Simulation of the relationship between jiggle and re-innervated fiber percentage. Bars represent the SEM values. One-Way ANOVA (Dunnett) test was used for significance. Each re-innervation group is compared with following increased re-innervation group. (*: $p<0.05$, significance level is same for 100μs, 200μs and 300μs).

In the formation process of NMJ, from the initial state (unstable) to the stable state, it is believed that muscle fibers are not always stimulated in the case of consecutive stimulation [10]. This is called blocking, which causes shape change in successive MUP waves and significantly affects the consecutive MUP wave shape. The simulation results supports this assumption. The percentage of blocking, which is accepted as 1% for normal muscle fibers, increases up to 50% for pathological muscle fibers. When the jitter time is kept constant and the blocking rate increases significantly up to 30%, the jiggle value also increases, and remains constant at 30% (Figure 7). There is no significant

increase between 30 and 50 % of blocking. This situation can be explained as the high blocking rate of the fibers causes a drop in the number of SFAP waves that form MUP waves.

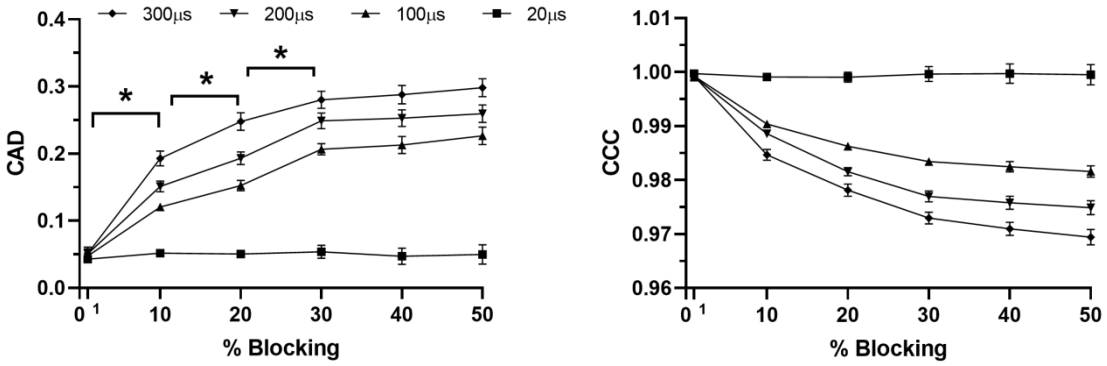


**Figure 7.** Simulation of the relationship between jiggle and blocking percentage. Re-innervation rate is 20% for the blocking rates 10-50%. Bars represent the SEM values. One-Way ANOVA (Dunnett) test was used for significance. Each blocking percentage is compared with following increased blocking percentage. (*: $p<0.05$, significance level is same for 100µs, 200µs and 300µs, no significance for the 20µs jitter duration).

Muscle fiber density (the number of muscle fibers per unit cross section) varies in certain regions of a muscle bundle [5, 17]. Changing muscle fiber density will change the number of muscle fibers in the recording area of the concentric electrode, and thus the number of SFAP that create the MUP wave. The increase in muscle fiber density in normal muscle groups does not make a statistical difference in jiggle value. Because the jitter duration and blocking percentage are very low in normal muscle groups, increasing the muscle fiber density does not create a significant change in consecutive MUP waves. However, as the muscle fiber density increases in pathological muscle groups, the jiggle value decreases statistically between 15 and 30 fibers/mm$^2$ according to the 5 fibers/mm$^2$ (Figure 8). This is due to the increased number of muscle fibers in the concentric electrode recording area, which relatively decreases the reinnervated muscle fiber density.

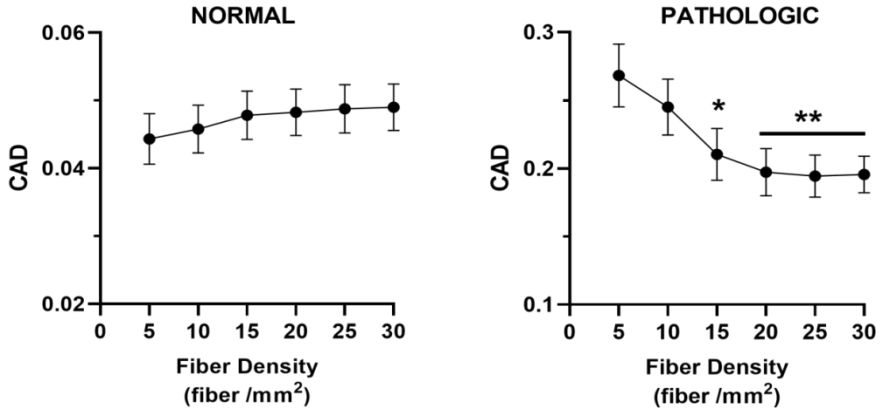


**Figure 8.** Simulation of the relationship between jiggle and fiber density. For normal group: Jitter value is 20µs, re-innervation ratio is 1% and blocking ratio of re-innervated fibers is 1%. For pathological group: Jitter value is 100µs, re-innervation ratio is 10% and blocking ratio of re-innervated fibers is 10%. One-Way ANOVA (Dunnett) test was used for significance. Each fiber density results are compared with the initial fiber density (5fiber/mm$^2$). (*: $p<0.05$, **: $p<0.01$).

## 5. CONCLUSION

Consecutive MUP simulation using real muscle parameters is necessary to understand the factors affecting the function of MU during neuromuscular pathology. In this study, quantification of consecutive MUPs was used to reveal the effects of jitter duration, re-innervation percentage, blocking percentage and fiber density to the shape of the MUP. Increasing jitter duration observed between pathological SFAPs of a MU composing MUP increases the jiggle value. Similarly, re-innervated motor fiber percentage in a MU also increases the jiggle value of consecutive MUPs. In the initial stage of the re-innervation process, the fusion of the NMJ is unstable and blocking phenomena occurs during consecutive stimulation of MU. Blocking of some fiber in a MU causes changes in MUPs and increases the jiggle value. Jiggle value increases when blocking ratio of re-innervated fibers rises up to the 30% but higher blocking ratios do not affect the jiggle value because of the decreasing number of re-innervated fiber percentage. Fiber density of a muscle fiber changes in some regions of the muscle bundle especially muscle fibers near the tendons are denser than middle of the muscle bundle. Simulation of this parameter showed that higher fiber density decreases the jiggle value because of the increasing number of fiber in the concentric electrode active area. Therefore, recording MUPs close to the muscle provides accurate and stable jiggle calculation.

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