A COMPARISON OF TECHNIQUES FOR

DECOMPOSITION SURFACE EMG SIGNALS INTO

MOTOR UNIT ACTION POTENTIAL TRAINS

By

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A COMPARISON OF TECHNIQUES FOR

DECOMPOSITION SURFACE EMG SIGNALS INTO

MOTOR UNIT FIRING ACTION POTENTIAL TRAINS

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Abstract:

Advancements in surface electromyography (sEMG) have led to many discrepancies in techniques used for signal decomposition. Specifically, the capabilities of well-established recording systems, and the methods involved in identifying motor unit (MU) action potentials and respective firing behaviors. **PURPOSE**: To examine the differences in MU identification and validation procedures, and firing behaviors between a four-channel (4-ch) sensor and a sixty-four channel (64-ch) high-density sEMG array. **METHODS:** Following 2 maximal voluntary contractions (MVC), ten (23 ± 3 yrs.; 178.64 ± 5.82 cm; 177.8 ± 17.37 kg) lower body resistance trained males performed 10 sec submaximal isometric ramp contractions of the knee extension exercise at 10%, 20%, and 50% MVC. During testing sEMG was recorded from the vastus lateralis using both 4-ch and 64-ch sensors. Signals were separately decomposed into their constituent MU action potential trains and were further validated for subsequent analysis of firing behaviors. The slope and *y*-intercept were calculated across the relationships between recruitment threshold versus mean firing rate (RT/MFR). A 2-way mixed factorial ANOVA (sensor [4-ch vs 64-ch] × contraction intensity [10% vs 20% vs 50%]) was used to examine mean differences in MU yield during all contraction. For validated MUs, the RT/MFR relationships were compared between sensors at each intensity and a paired samples *t* test was used to compare differences in RTs. **RESULTS:** There was a significant interaction between sensor and intensity, as well as a main effect for intensity, with follow up analysis revealing a significant difference between MUs validated at 10% and 50% MVC ($p < 0.05$). There was a significant difference in slopes at 10% and 50% MVC, and *y*-intercepts at 20% MVC for RT/MFR relationships ($p < 0.10$) and the RT of validated MUs were significantly different ($p <0.5$) between sensors at each intensity. **CONCLUSION:** MUs validated using the 4-ch sensor yielded a greater numbers during higher contraction intensities versus the 64-ch sensor. The inability of the 64-ch sensor to yield a greater amount of MUs at 50% MVC may have been due to the subjectivity of the manual editing procedures. However, both validation procedures eliminated a high amount of decomposed MUs.

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CHAPTER I

Introduction

Originally established using intramuscular electrodes, LeFever and De Luca (1982)[1] developed techniques to separate acquired electromyographic signals into individual motor unit action potentials (MUAPs) and identifying their respective discharge patterns. This method came to be known as decomposition electromyography. Decomposition of the electromyographic signal is performed by identifying and overlapping MUAPs into individual trains based on their initial and continuous discharges (i.e., shapes, firing instances). Over the past several decades these techniques have been expanded upon using non-invasive techniques from surface electromyography (sEMG) sensors[2-12]. However, varying approaches in decomposition techniques from sEMG recordings have become more controversial due to the ensuing challenges associated with the identification of MUAP firings[13,14]. Recently, several concerns regarding data acquisition and subsequent signal processing techniques of motor unit (MU) decomposition have been debated and described[13].

Recently, several concerns regarding data acquisition and subsequent signal processing techniques of motor unit (MU) decomposition have been debated and described[13]. Over the years, Dr. Roger Enoka and colleagues have stressed the importance of concurrent two-source recording techniques that can allow for a more accurate analyses for tracking action potentials within the same contraction[5,15]. These are common practices for researchers that utilize a high density surface electromyography (HDsEMG) electrode[16-18]. These electrodes have a large channel (32-64) grid arrangement and are capable of collected sEMG signals from a sizable portion of the muscle of interest. Conversely, for MU decomposition, the use of a five-pin/four-channel sensor (64-ch; Delsys, Inc., Natick, MA,USA) has been highly criticized for its validation procedures[15,19]. Specifically, the automated decomposition techniques of the 4-ch sensor that claim to validate a higher yield of MUs, as well as its ability to distinguish low-thresholds MUs during contraction at various intensities[2,20]. Therefore, the 4-ch sensor has the capabilities to differentiate overlapping MUAP firings of lower and higher-thresholds and categorize them into their constituent action potential trains for further analysis of MU firing behaviors.

Accordingly, over the past decade these advancements have provoked many topics for debate. Many of these are in regards to the accuracy of the commonly utilized algorithms of the 4-ch sensor. Unlike the processing techniques of the HDsEMG grid array (i.e., 64-ch), the decompositions techniques of the 4-ch sensor are provided by the manufacturer which employ proprietary acquisition and processing techniques for the identification of MUs and subsequent tracking of their respective firing behaviors. Specifically, these provided procedures are achieved using automated programing and does not allow visual inspection or re-identification of the MUAPs within the software itself. The contemporary techniques concerning MU decomposition using the 4-ch sensor were first described by De Luca et al. (2006)[12] and further improved upon by Nawab et al. (2010)[20] and De Luca and Hostage (2010)[2] through the application of the newly developed algorithms. The results of these studies emphasized the algorithms ability to accurately (92.5 - 97%) decompose, as well as discriminate, the firings of ≤ 40 MUs into their constituent motor unit action potential trains ($MUAP_T$). Decomposing signals acquired from contractions performed at intensities/levels up to maximal force production, De Luca and Hostage (2010)[2] confidently reported the promising advantages of using the Precision Decomposition III (PD III) algorithm to accurately detect and track the a high number of MUs. Although promising, the inability of researcher to see these algorithms and procedures perform in real time has created much skepticism that has led to further validation requirements.

Preliminary reports using earlier version of the PD III algorithms were proposed using a two-source method to acquire and reconstruct sEMG signals[1,12,21], for initial validation. A two-source method is performed via concurrent recordings involving intramuscular electromyography (iEMG) which uses indwelling techniques such as fine wire or needle electrodes that are inserted into the muscle itself. Although these allow for a more accurate representation of MU discharges, there are several limitations associated with these applications (i.e., small pick-up area, low-contraction forces, uncomfortable invasive procedures) [6,20]. In essence, the technological advancement of 4-ch sensors for the use of noninvasive sEMG was created to overcome practices of two-source methods. These recent developments have provided the PD III algorithms the ability to accurately decompose sEMG signals through the assistance of the artificial intelligence-based Integrated Processing and Understanding of Signals concept[2,20]. The PD III algorithm begins by extracting all identified MUAPs from the sEMG signal and assigning them into their constituent templates (i.e., trains). Within these templates, superpositioned (i.e. duplicates of a singular MU and/or overlapping MUs) and unidentified MUAPs are classified in signal regions which are required to satisfy specific criteria for ultimate inclusion (i.e. constructive and destructive interference effects, inter-pulse interval < 0.35s). Thereafter, the algorithm continues to identify additional MUAPs and either updates an existing template or creates a new one based on the initial firing of the MUAP. This process known as Decompose-Synthesize-Decompose-Compare (DeLuca and Contessa 2012), is deemed completed once each MUAP has been categorized into its respective template and removed in order to determine any potential superimposed firings. However, this task can be daunting due to factors related to the high number of firings in each MUAP, filtering, and the high variability (i.e. shapes) and/or overlapping between firings[3,15].

As such, various conflicting opinions regarding these concerns have consequentially required researchers using these sensors to adhere to more rigorous analyses and evaluation techniques[22-27]. The results of these studies illustrated the many inconveniences that can arise during concurrent acquisition of intramuscular signals and their methodological limitations. Specifically, per recommendation for further validation[5,13,15], the concurrent use of the dEMG sensor with indwelling techniques have reported the limitations may be due to the small pickup area and/or amount of MUs recorded[12,26]. Potentially, these limitation could be due to the differences in validation and processing techniques[28]. As well as, limitations involved with intramuscular electrodes which hinder the ability to perform contractions at higher intensities, and restrict information regarding MUs of higher thresholds[14].

Since early investigations classifying the "onion-skin phenomenon", researchers commonly report on MU firing times (i.e., discharge rates) and shapes of individual MUAP under conditions that are dependent on the muscle and contraction type. As reported by Fuglevand et al. (2015)[29] … "the relation between naturally occurring synaptic input and firing rate responses in motor neurons can be indirectly assessed." Thus, muscle force production can serve as an indicator of the synaptic excitation (i.e., neural drive) during voluntary contractions[29] and as a noninvasive technique to support characteristics of muscle fiber types[30,31]. These are commonly reported in literature using either the 4-ch or 64-ch sensors, which follow the aforementioned processing recommendations for the 4-ch sensor, and use isometric constant force trajectories to record and analyze MU firing patterns[2,20].

Generally, as an evaluation of neural modulation across force, the relationship between the mean firing rates (MFR) of a MUAP and its relative recruitment threshold (RT) are described using linear regression[2]. As previously mentioned, earlier recruited low-threshold MU have a greater firing rate at higher intensities than those recruited later. Thus, an inverse relationship can be plotted using MFR and their respective RT (i.e., slopes and intercepts) of decomposed action potential. However, in opposition to the relationships associated with peak firing rates, Enoka (2019)[13] describes the differences between studies that utilize signal processing of the 64-ch sensor versus those using the 4-ch sensor. To corroborate the focus of Dr. Enoka's review, the comparisons discussed focus on decomposition studies that have been able to replicate findings of those recorded with intramuscular electrodes. For example, similar to reports using intramuscular recordings, Enoka discusses a recent investigation by Del Vecchio et al. (2017)[32], which uses a HDsEMG electrode recordings to report contrasting results in mean firing rates between low-and high-threshold MUs. Specifically, the behaviors of mean firing rates between low-and high-thresholds MU were not as distinctive as those reported using the dEMG sensor during a high intensity contraction.

Although similar in reporting MU firing behaviors, investigations using the 64-ch sensor follow unique decomposition techniques which also use algorithms and manual editing techniques during two-source methods[9,10]. The details of the signal processing reported by Del Vecchio et al. (2017)[32], utilizing the 64-ch sensor, are thoroughly reported by Negro et al., (2016)[33] and Martinez-Valdez et al. (2016)[17]. Briefly, MUs are identified using a method of convolutive blind source separation (BSS). This method distinguishes between different MUs by using the absence of firing times (< 50 pulses per second) relative to the sampling rate (2 kHz) to identify respective action potentials. A silhouette measurement comparing the amplitudes of the deconvolved MU spikes, relative to the background noise, is then used to assess decomposition accuracy. The mutual time between consecutive action potentials is used to calculate instantaneous firing rate. Specifically, the initial firing rate, calculated as the ratio of the change in firing rate from the minimum rate to that at the force constant, divided by the force constant. These are then low-pass filtered with a first order Butterworth filter and a cut-off frequency set at 0.5 Hz[13]. In essence, BSS begins by estimating individual spike trains of a single MU and repetitively updating that MUs separation filter and applying it to the original signal. The filter, in turn, is continuously updated by improving the amount of sparseness for the MU trains of the predefined time intervals (Del Vechio 2020)[38].

In a recent debate, a letter to the editor entitled: “In Regards to Motor Unit Decomposition, Are We Caring about the Right Information?”, Dr. Jason DeFreitas (2019)[14] notes the disparities of interpreting IFR's, due to their short windows (i.e. < 2 sec) for averaging firing rates. Dr. DeFreitas goes on to explain how IFR's are commonly reported in studies in which the initial firings are calculated from contractions using quick increases, or oscillations of force. This is uncommon in most decomposition studies, specifically those that utilize steady state muscle contractions in order to assess deviations of inter-spike interval (ISI) distributions that can signify potential identification errors[26]. Additionally, Dr. DeFreitas expresses his concerns regarding the unlikelihood of detecting a vast number of low-threshold MU during low intensity force production, using decomposition approaches reported by Enoka (2019)[13] (i.e. two-source method). These can limit researchers' ability to assess muscle contractions at high-levels of force production and therefore limit the ability to recruit high-threshold MUs. If the technologies exist and are at our disposal, why not formulate new approaches to examine MU behaviors.

Additional concerns that may hinder recordings of MU decomposition are greatly considered throughout analysis and validation procedures. Several include doublet discharges or superimpositions that can hinder the ability to discriminate between firings of MUs. Doublets are pairs of short ISI (< 10 ms) that can occur during at the initial onset or sporadically throughout the contractions[34]. These doublet discharges can effect signal processing and automated MU decomposition techniques (i.e. PD III, BSS) by either missing initial firings or not including these into respective MUAP trains. Accordingly, several investigation have reported statistical methods to further evaluate signal accuracy and processing techniques of decomposed MU's, via analysis of ISI distribution and spike-triggered averaging[22-27]. Although rigorous, these can be performed routinely and provide information regarding the decomposition accuracies and the nature of the error during different experimental conditions.

1.2. Purpose of the Study

Despite the many limitations involved with sEMG, the controversy regarding the analyses of MU firing behaviors and the techniques of well-established recording procedures warrant concurrent examinations. Therefore, the purpose of this study is to simultaneously record muscle activation, using the 4-ch and 64-ch sensors, to compare decomposed signal recordings and respective MU firing behaviors.

1.3. Research Question

Information obtained from this investigation has the potential to provide insight into MU firing behaviors simultaneously recorded from two of the most commonly used decomposition techniques. Therein, the following research questions have been established to potentially address concerns within literature that need to be answered:

- Are there differences in the number of MUs yielded from each of the two sensors during contractions of the leg extensor muscles?
    - If so, are these associated with the location of the sensor placement
        - signal processing of decomposed MUs?
        - MU identification from low and high intensity contractions?
- Are the firing patterns of recorded MUs different between the two sensors:

  - mean firing rates (MFR)
  - recruitment threshold (RT)
  - relationships between these as expressed via linear regression coefficients
- Are their differences in MU firing behaviors during the various intensities of isometric ramp contractions?
  - will the two decomposition techniques be able to differentiate the low and high threshold MU to yield similar regression slopes and intercepts (i.e., MFR vs RT)

1.4. Hypotheses

- Following respective validation of MUs collected from both sensors, the number of MUs yielded from each sensor should not be significantly different.
- All MU firings during isometric ramp contractions will have similar firing properties (i.e., MFR vs RT relationships) following respective recordings and analysis procedures.

1.5. Significance of the Study

This study has the potential to report similarities or differences in the firing behaviors of MUs recorded using two highly utilized decomposition techniques. Including a variety of submaximal contractions commonly performed in laboratory testings' and will offer a robust evaluation and potentially address concerns of recording high-threshold MU.

1.6. Delimitations

The following are the delimitations for this study:

1. Approximately 10-15 males are needed to complete this investigation.
2. Participants must be between 18 to 35 years of age.
3. All participants must be healthy, recreationally active, and free from any neuromuscular disease.
4. Participants will be required to visit the laboratory on 2 separate occasion and be able to perform knee extensions of various force levels and contraction types.
5. Participants will be asked to refrain from physical activity or exercises involving the lower-extremities during the duration of this study.

1.7. Limitations

1. Participants being recruited for this study will come from either classroom visits, a posted flyer, or from the laboratory website. Thus, participants will likely be students from the School of Kinesiology, Applied Health, and Recreation.
2. Many of the affirmation limitations with the technology and equipment used to assess motor unit firing behaviors can potentially restrict analyses.
    a. Debated inaccuracies of the algorithms and decomposition methods using the 4-ch sensor.

b. Caution when pooling firing of higher-threshold motor units into their constituent action potential trains with the 64-ch sensor.

3. Absence of concurrent recordings from indwelling iEMG.

    a. Currently recommended as the "Gold Standard" for recording EMG signal

1.8. Assumptions

1. Participants answer health questionnaire honestly and accurately
2. Each maximal contraction is elicited under respective criteria.
3. Both sensor locations accurately detect sEMG signals and represent motor unit firing behaviors of the whole muscle
4. The independently established validity of both sensors and processes techniques is accurately depicting the relationships between MU firing behaviors.
5. The sensor location are accurately depicting activation of the whole muscle.

1.9. Threats to Validity

Listed below are the potential threats to validity and the actions that will be taken to account for them:

1. *Potential of induced fatigue*
    a. Due to the amount of contractions within a single visit, optimal rest time between contractions and between recording procedures will be given in order to limit the risk of fatigue.
2. *Order Effect*

a. Force tracings for each submaximal contraction will require a small amount of skill acquisition. Due to potential learning effects, all contractions under both conditions will be performed during a familiarization visit. All contractions will be in a randomized order and thoroughly instruction for performance outcomes.

3. *Intra-subject variability* –

   a. Due to potential inconsistently in subject performance and electrode placement all conditions will be performed within the same visit for subsequent analysis.

CHAPTER II

Review of Literature

The review of literature is organized into subsections, each in chronological order summarizing studies that are most relevant to its respective section.

2.1 Motor Unit Firing Properties

Liddell and Sherrington, 1925

The purpose of this investigation was to examine the occurrences of inhibitory relaxation following stimulation of the ipsilateral afferent nerve. Although this is not directly related to the present study, it was the first to use the term *motor unit.* The authors are accredited with being the first to recognize all of the fibers innervated by a motor neuron behave as a single entity.

Adrian and Bronk, 1929

The purpose of this investigation was to examine motor neuron firing properties and was the first study to detect action potentials from a single motor unit (Duchateu and Enoka, 2011). Of major significance, the authors report the changes in discharge frequency in fibers and the number of active fibers directly influence force gradation.

These highly contributable findings recognize the two primary influences for increased force production: increase firing rates for active motor units and recruit more motor units with increases in force.

2.2 Techniques for Motor Unit Decomposition

Mambrito and De Luca,1984[21]

The purpose of this study was to provide a basic demonstration of the decomposition system and the techniques involved for signal detection and recording of EMG signals for successive decomposition. Additionally, this study provides references for detailed presentation of the algorithms involved, as well as statistical techniques for analyses of decomposed MUAPTs. Using a quadripolar needle electrode, Mambrito and De Luca describe 4 main sections for EMG signal processing and decomposition techniques to accurately extract as many MUAPs from the acquired signal.

1. The first, describes a methodological approach for signal acquisition and quality verification using a quadripolar electrode. This electrode was designed to enhance the discrimination between different MUAPs acquired from 3 channels of EMG signals. Additionally, due to the inconveniences placed on the experimenter, an automated experiment control system was devised to assess EMG signal quality appropriate for decomposition.

2. The second, a recommended sampling and processing for EMG signals (bandpass filter of 1 kHZ and 10 kHz) for the present conditions.

3. The third, the introduction of a computer assisted interactive algorithm to extract MUAPs and match them into their respective MUAPTs. This is done by the algorithms ability to continuously update template matching and firing statistics to identify MUAPS in the EMG signal. This also allows the templates to be updated so that the algorithm can function even when the shapes of the MUAPs begin to vary.

4. The forth, discusses ways a researcher can analyze and display the results in time domains of the MUAPTs.

    a. By displaying the wave forms (shapes) of MUAPs

    b. Impulse trains representing MUAP firings

    c. Interpulse interval (i.e., ISI) plots, representing time intervals between motor unit firings vs. the time of the muscle contraction

    d. Firing rate plots estimated from the mean firing rates of detected MUs vs. the time of the muscle contraction

Additionally, this study provided tester reliability for the discussed procedures; accuracy of the evaluation techniques when recorded from a synthetic EMG signal; and accuracy measures from real EMG signals recorded independently and simultaneously from two different electrodes. Specifically, the comparison of the result from signals that were able to detect and categorize the similar MUAPTs.

Farina et al, 2008[6]

The purpose of this study was to investigate the number of identifiable MUAP within simulated and experimental sEMG recordings. Using simulated MUs from a cylindrical anatomical system (electrode grid 11 x 11 with pre-distinguished collection channels for comparison), Farina and colleagues compared the number of MUs that could be identified from respective location using intramuscular recordings under low intensity contractions (2.5, 5, 7.5, 10, and 12.5% MVC force) from the abductor digiti minimi. The results of this indicated that relatively few MU are distinguishable when only few channels of sEMG recordings are used to discriminate the same MUs in both techniques. Thus, the researchers suggest the use of a larger multichannel grid arrangement (i.e. HDsEMG array) in order to discriminate a high proportion of MUs rather than a detection arrangement of only a few channels for recording (i.e. dEMG sensor).

Holobar et al., 2009[9]

The purpose of this study was to systematically examine a recently developed approach for the approximation of complete MU discharge pattern that was developed by the researchers, called Convolution Kernel Compensation (CKC). Using an HDsEMG array, Holobar and colleagues examined the capabilities of the CKC to decompose sEMG recordings of low-intensity force varying contractions. Specifically, they wanted to test the potential capabilities of the CKC method to;

1. Identify a relatively large number of MU sampled from a population of various concurrently active MUs

2. Track early-recruited (low-threshold) MUs when higher-threshold MUs are later recruited

3. Identify MUs from different muscles with diverse anatomies

4. Identify MUAP trains that match those collected by intramuscular EMG recordings

This study was the first to provide a comprehensive performance analysis for methodology using sEMG decomposition and validation of individual MUs using the HDsEMG array. The authors do however conclude that although the CKC technique does provide support, the decomposition of sEMG should continue to be concurrently recorded with iEMG recordings in order to increase the number of identified MUs.

<u>Holobar et al., 2010[10]</u>

The purpose of this study was to compare decomposition results from both the HDsEMG array and concurrently recorded iEMG from 3 separate muscles during low-intensity (between 5% and 20% depending on the muscle). As a follow up study to the previous (Holobar et al 2009), this study also used the previously mention CKC technique to decompose sEMG recordings, as well as the use of EMGLAB for concurrent iEMG decomposition. The authors do stress the extensive manual editing required using EMGLAB for intramuscular recordings, and the difficulties associated with MUAP superimpositions for the inclusion/exclusion criteria for identifying MU discharges from iEMG. The average discharge rate (firing rate) and the coefficient of variation (CoV) for the ISI were computed for each identified motor unit for both decompositions methods. Concurrently identified MUs from the two decomposition methods were compared using the rate of agreement (RoA). The results of this study indicate a relatively high percentage (84%-89%, muscle and force level specific) of MU discharge times that were identified by both decomposition methods for each muscle across force levels. Although the state the index of agreement between these methods was linearly correlated with a self-consistency measure of MU discharge patterns (based on CoV of ISI) (R^2 = 0.38 – 0.68, for the 3 muscles examined), the authors do state;

> "Dispite the relatively small number of common motor units per contraction, because of the large number of contractions, the total number of motor units identified by both decomposition techniques was in the order of hundreds and allowed for a systematic validation of the decomposition results on a large data set…. The results on discharge statistics and on the high rate of decomposition agreement, and the observation that the errors in surface EMG were probably overestimated in the current validation because of the potential errors in intramuscular EMG decomposition, indicate that the analysis of motor unit behavior in the conditions analyzed can be performed with equivalent accuracy using intramuscular or surface EMG."

Thus validating the use of the HDsEMG array and decomposition methods during static, low-force contractions and bringing forth further concerns with concurrent use of intramuscular recordings.

<u>Newab, Chang, and De Luca, 2010[20]</u>

The purpose of this study was to report the recent technological advancement for the estimates the firing patterns of active MUs, as previously reported by De Luca et al. (2006). Then newly enhanced system uses artificial intelligence based algorithms (i.e., PDII) to decompose sEMG signals acquired from the four channels of the 5-pin surface electrode. sEMG signals were recorded from five muscles during isometric contractions at force levels up to 100% MVC. The accuracy of the decomposition was measured using a decompose-reconstruct method, and further validated for accuracy using concurrent indwelling EMG. The results of this investigation highlight the ability of the enhanced algorithms to yield a high number of motor units, occasionally up to 40, among various muscles and force levels. Additionally, the firings of the MUAP trains were shown to average 92.5% accuracy and at time reach up to 97%. The claims made regarding the reliability of the reported technological advancements for high-yielding decomposition sEMG has since begun an ongoing discrepancy between many researchers.

De Luca and Hostage, 2010[2]

The purpose of this study was to characterize the relationship between motor unit recruitment thresholds and mean firing rates during isometric contractions. The behaviors of these relationships were formulated from sEMG signals from the VL, FDI, and TA during muscle contractions at 20, 50, 80, & 100% MVC. These were recorded and decomposed from previously mentioned techniques (Newab, Chang, & De Luca, 2010), into constituent MUAP trains. The linear relationships represented as the coefficient of determination (R^2) between mean firing rate and recruitment threshold was shown to be much higher for individual subjects as compared to the entire group. Furthermore, the pooling of MUs from the multiple subjects reduced the R^2 value. Thus, R^2 should first be determined on an individual basis per contraction, then averaged along with other R^2 values from the same contraction. The results of this study report the “operating point” for the motoneuron pool that was shown to be consistent throughout the hierarchical inverse relationship between the recruitment thresholds and mean firing rates of the calculated MUs. Additionally, the modulation of excitation from the firing rates of recruited MU's across the increases of force levels. Therefore supporting the "onion skin" phenomenon and "common drive" of the motoneuron pool.

Farina and Enoka, 2011[15]

In this Letter to the Editor, the authors address the concerns with the reported analyses and employed techniques from the investigation by De Luca and Hostage (2010) and Newab, Chang, and De Luca (2010). The authors described, in their professional opinion, the difficulties associated with discriminating between overlapping action potentials in MU firings, especially those at higher force levels when recorded with intramuscular techniques. They further postulated what they viewed as inaccuracies existent in the PD III algorithm. Specifically, the authors' state;

"the ability to solve the global optimization of overlapping action potential using polynomial complexity algorithms is unlikely because it is a non-deterministic polynomial-type hard problem."

Moreover, the authors continue to rationalize the disparities between validation methods and procedures (i.e. *reconstruct-and-test procedure*), exemplifying that missed discharges from first or second order decompositions may, indeed, elicit inaccurate MUAP trains, in addition to insinuating that limitations in the signal processing and comparative analysis to classical two-source test (i.e. concurrent iEMG). Therefore, the authors request a more rigorous evaluation before claiming that the PD III algorithm can accomplish what was reported.

De Luca and Nawab, 2011[3]

In this reply to Farina and Enoka (2011), Drs. DeLuca and Newab thoroughly offered their in-depth defense of the decomposition algorithms ability to differentiate overlapping action potentials through the combined use of their PD III along with the IPUS concept. Additionally, the defense of the mathematical and methodological approach of the reconstruct-and-test, which was developed to overcome the short comings of a more commonly used "*generic test*" from mathematically synthesized signals and two-source methods.

Farina, Merletti, & Enoka, 2014[5]

In this update from their original literature review (Farina, Merletti, & Enoka, 2004), the authors discuss several important features of the potential benefits from extracting information about neural activation in the muscle. Of primary importance, these discuss the many challenges associated with retrieving the embedded neural code from sEMG signals that can be difficult to accomplish. Thereafter, several topics of debate are highlighted regarding limitations and the aforementioned concerns for the decomposition techniques from the 4-ch sensor.

“The action potentials of the active motor units can only be distinguished with adequate spatial information, which requires many recording channels of the EMG signal. An increase in the number of channels in which each motor unit action potential is represented will increase the number of motor units that can be uniquely detected at the skin surface, rendering the decomposition challenge theoretically possible.”

The review goes on to reason additional validation procedures performed by Hu et al., (2013) (discussed later), that use STA techniques to further interpret their rationale for not just the decomposition algorithms but the tests establishing the validity (i.e., reconstruct-and-test procedure). Finally, they conclude by suggesting the two-source approach, discussed earlier, which allows for an unbiased approach expressing the rate of agreement from separate approached of intramuscular vs. surface decomposition.

DeLuca, Nawab, and Kline, 2015[28]

In this Letter to the Editor, the authors request clarification from conflicting arguments made against their decompose-synthesize-decompose-compare strategies (DSDC) (formally *reconstruct-and-test procedure)*, as well as their suggested two-source method techniques. It seems that in these exchanges the conflicting arguments are misinterpreted by various statements. Specifically, the procedures involved with the decomposition validation via the reconstruction of synthetic signals that assess the accuracy of the decomposition algorithms.

Farina, Merletti, & Enoka, 2015[19]

In this reply to De Luca, Nawab, and Kline (2015), the authors clarify the issues regarding the proposed DSDC and address the misinterpretations of what is actually being debated. Therefore, generalized the argument that;

"sEMG decomposition is a source separation problem and a property of many source separation methods is that the residual noise decreases systematically with an increase in the number of estimated sources."

They conclude by remaining steadfast to their opinion of concurrent intramuscular and sEMG signal decomposition currently being the best practice for validation.

Enoka, 2019[13]

The purpose of this review was to compare results of investigations that have achieved decomposition of sEMG signals that agree with what is known from recordings obtained with intramuscular electrode. Specifically, surface decompositions that have been able to characterize discharge times of single motor units with rate coding characteristics that match those from iEMG. Those comparison of characteristic that are relevant to those of the present include; peak discharge rate, saturation of discharge rate during submaximal contractions, rate coding during fast contractions, and the association between oscillation in force and discharge rate. Although this review brings forward many of the replicated findings for agreeance between intramuscular and surface decomposition, it also identifies important focus for waveform editing from algorithms that can improve the understanding of motor unit physiology and its potential applications.

DeFreitas, 2019[14]

In this recent letter to the editor Dr. Jason DeFreitas (2019) provides a rational to concerns in Farinas' review asking the question; "are we focusing on the right information?". He begins by debating the issues regarding superimpositions when discriminating low- and high-threshold MUs, and continues to describe the innovations associated with the abilities to extrapolate a large population from the MU pool with the use of new technological advancement. Although in agreeance with some of the concerns from Farinas' review, DeFreitas argues the importance of the ability to yield and assess high-threshold MUs. Later he notes the disparities and inconsistent findings regarding the interpreting of instantaneous firing rates, due to their short windows (i.e. < 2 sec) for averaging firing rates and goes on to explain how IFR's are commonly reported in studies in which the initial firings are calculated from contractions using quick increases, or oscillations of force. This is uncommon in most decomposition studies, specifically those that utilize steady state muscle contractions in order to assess deviations of ISI distributions that can signify potential identification errors (Hu et al., 2014). Additionally, Dr. DeFreitas (2019) expresses his concerns regarding the unlikelihood of detecting a vast number of low-threshold MU during low intensity force production, using decomposition approaches reported by Enoka (2019) (i.e., intramuscular). These can limit researchers' ability to assess muscle contractions at high-levels of force production and therefore limit the ability to recruit high-threshold MUs. If the technologies exist and are at our disposal, why not formulate new approaches to examine MU behaviors.

2.3 Additional Validation Procedures for Motor Unit Firing Behaviors

Hu, Rymer, Suresh, 2013[23]

The purpose of this investigation was to thoroughly test the reliability of estimated MU parameters using spike triggered averaging (STA) of the sEMG signal. The authors investigated factors that could potentially induce amplitude bias when estimating MUAPs and shape variations using a reconstructed or simulated EMG signal derived from a 30% isometric contraction recorded from the FDI, using the 4-ch sensor. From the simulated sEMG recording, MUAPS were estimated from STA and five variables were examined;

1. Amplitude variations within the MUAP train
2. Varying duration of a MUAP train
3. Action potential super-position due to high firing rates
4. Synchronized firing effects
5. Spurious even classification during firing event discrimination

The issues for each these are thoroughly discussed. Briefly, the variation in MUAP duration led to an underestimation of the real MUAP amplitude. The synchronized firings led to and over-estimation of the amplitude. For small MUs, spurious firing resulted in the over-estimation in amplitude, while an under-estimation in amplitude was shown in large MUs. Amplitude estimation was minimally influenced by the variability in amplitude and high firing rates. There were large variations in MUAP shapes with higher firing rates and variations in MUAP duration. Finally, there was also a correlation between estimation errors and shape variations. Overall, this study was able to identify sources of STA biases that can arise from physiological properties of the MU pool and signal recording and processing procedures. Overall, STA can be used as a valid assessment if appropriate actions are used to remove unreliable estimates.

Hu, Rymer, Suresh, 2013[22]

The objective of this study was examine MU pool organizational properties by employing two separate sets of tests to examine and assess the validity of the decomposition results collected from the 4-ch sensor. The sEMG signals were recorded from the FDI using commonly practiced recording techniques recommend for the 4-ch sensor. For both subsequent examinations, participants performed 3 MVCs followed by submaximal isometric contraction utilizing trapezoidal force trajectories at 20%, 30%, 40%, and 50% MVC. The first test, STA, was used to reconstructs the shapes of the action potentials by using the MU firing times as triggers for the recorded raw sEMG signals.

Hu et al., 2014[26]

The objective of this study was to examine the firing statistics of the identified MUs in order to evaluate the accuracy of a decomposition algorithm from the 4-ch sensor. Decomposed MUs from both intramuscular and surface recordings EMG recordings were used for cross-validated of estimated ISI statistics. This investigation found that ISI distribution can provide information regarding the spurious errors and missed firing errors in the decomposition. Specifically, secondary peaks at the short or long ISIs, represents errors as shown in the deviation from the Gaussian distribution. Additionally, the authors report the inverse relationship between the decomposition accuracy and the variability (coefficient of variation) of the ISIs. Similar to the authors previous reports, ISI statics be used to ass spike timing accuracies of the identified MUs from the 4-ch sensor.

McManus et al., 2017[25]

Although the purpose of this investigation is not directly related to that of the present, the methodological procedures used extensively describe discrepancies within 4-ch validation techniques. To briefly describe the additionally validations used by McManus et al (2016); following the recording and analysis (Newab et al., 2010, DeLuca and Hostage, 2010) detected MUs and their respective firing times from their four MUAP waveforms (via four bipolar channels) were used to STA each respective, corresponding sEMG signal. Thus, four representative STA MUAP estimates are derived for each detected MU. The duration of each MUAP is estimated as the time between the zero crossing before the initial positive peak of the action potential and the zero crossing after the last positive peak. Using a moving average window, variations of STA MUAP trains were quantified over the duration of activation and shifted along the sEMG signal. Thereafter, the templates created from the STA analyses were then compared to detected MUs of from the original decomposed signal. Specifically, described in more detail by Hu et al (2013), two tests for reliability were administered in order to meet qualifications for acceptance. The first was performed by calculating the coefficient of variation for the peak-to peak amplitudes of MUAP templates in each window. Whereas, the second was performed by computing the maximal linear correlation coefficients between decomposed MU and the STA MUAP templates. The specifications for inclusion and further analysis were then determined depending on the length of the contraction, moving average window, and MU yield/validation per subject.

CHAPTER III

Methodology

3.1 Participants

Participants for this investigation consisted of 10 resistance-trained males (mean ± SD; height, 178.64 ± 5.82 cm; weight, 177.8 ± 17.37 lbs.; age, 23 ± 3 years). Inclusion criteria requires participants to have a minimum of 6 months lower-body resistance training experience. Individuals having experienced any current or recent musculoskeletal injury to the lower extremities or any neuromuscular disorders will not be allowed to participate in this study. All participants were required to complete a physical activity readiness questionnaire (PAR-Q), health history questionnaire, and a university Institutional Review Board approved informed consent form before testing procedures could take place.

3.2 Research Design

This study required 2 separate visits to the neuromuscular laboratory, separated by at least 48 hours. Following required documentation and inclusion criteria, the initial visit included familiarization of all testing procedures. Following a brief warm up, of 3 five-second knee extensions at perceived 25, 50, and 75% MVC, each subject then performed 2 MVCs.

Ultrasonography was used to identify location and muscle pennation angle, as well as measure subcutaneous tissue to confirm inclusion criteria. Each visit will consist of two maximal voluntary contraction (MVC) of the knee extension exercise, followed by 14 submaximal contractions. Visits 2 will serve as data collection for neuromuscular assessment of the VL muscle. Listed below is a summary for each visit, submaximal contractions (1.2 – 1.6) will be performed in a randomized order, as well as sensor recording conditions:

3.2.1 Testing Procedures

After obtaining the highest maximal force from the two randomized MVCs, all submaximal contractions and sensor recording conditions will be randomized with two minutes of rest between each contraction. Subjects will perform each of the listed contraction twice, separated by a 20 minute wash-out period.

1.1. Two MVCs

- Instructed to illicit rapid increase in the rate of torque development
- Instructed to illicit maximal force production

1.2. Sinusoidal contraction following a linear force trajectory increase up to 20% MVC with a 0.5 Hz waveform ± 5% then a linear decrease back to baseline

1.3. Sinusoidal contraction (same as prior) with a 1.5 Hz waveform ± 5%

1.4. Isometric trapezoidal contraction at 10% MVC

1.5. Isometric trapezoidal contraction at 20% MVC

1.6. Isometric trapezoidal contraction at 50% MVC

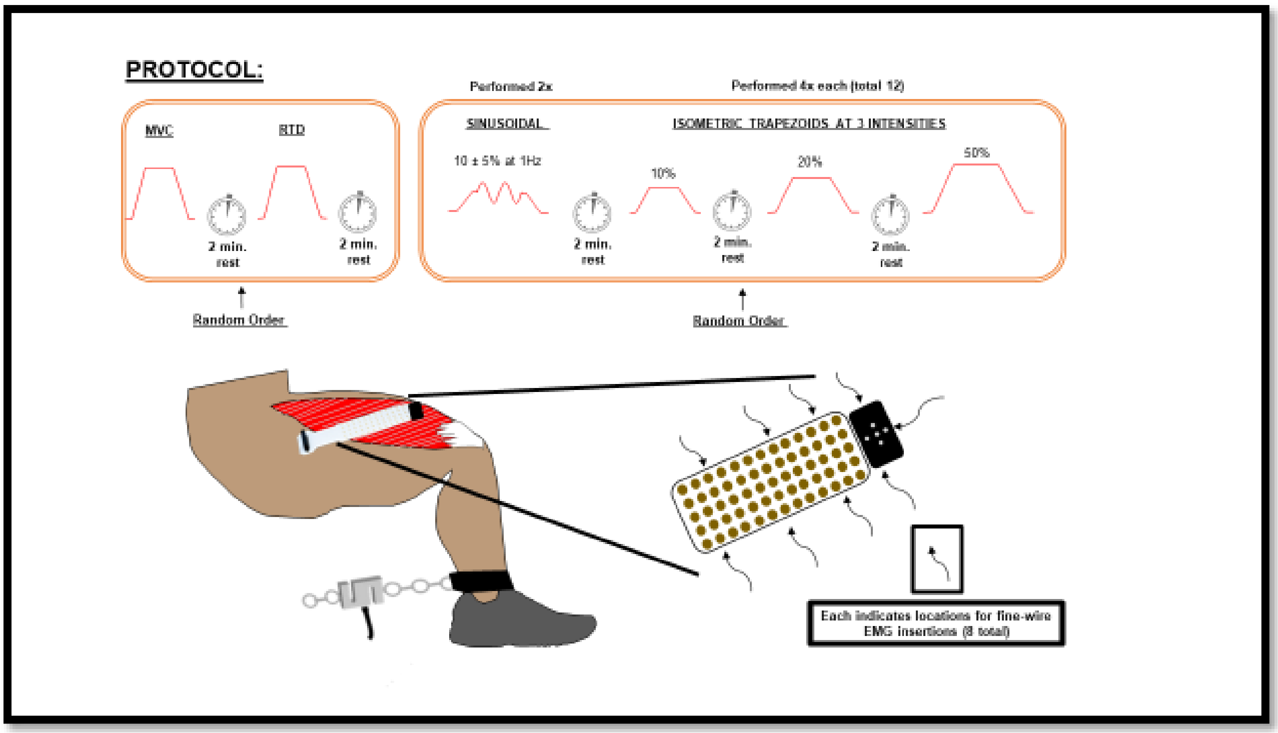

Figure 1 visual representation of the study protocol.

3.3 Instrumentation and Procedures

3.3.1 Ultrasonography

Muscle pennation angle of the VL will be assessed utilizing a brightness mode (B-mode) ultrasound imaging device (General Electric LOGIQ S8, Wauwatosa, WI, USA) and a multi-frequency linear-array probe (Model ML6-15-D 4-15 MHz, 50-mm field of view). Upon the initial familiarization visit, while lying supine, participants were asked to relax and lightly bend at their right knee (10-15˚) to obtain panoramic scans and help identify proper electrode placement and orientation. Panoramic scans of the VL will be taken at 50% the distance between the greater trochanter and the lateral aspect of the patella. To enhance images, a water-soluble transmission gel was applied to the probe and the skin. Three scans will be taken from the VL; ultrasound images were later analyzed using image analysis software (ImageJ, version 1.5i, NIH, Bethesda, MD, USA), with the average values utilized for statistical analyses.

3.3.2 Isometric Strength Testing

Participants were seated and secured in an upright position with their hip and knee joint angles fixed at 110° and 120°, respectively. Isometric force (N) was recorded using an S-beam load cell (Model SSM-AJ-500; Interface, Scottsdale, AZ, USA) attached to a cuff around the ankle. Following a warm-up of 3, 5 sec. self-determined submaximal isometric muscle actions at approximately 25%, 50%, and 75% MVC, the participants performed two separate, 3 sec. MVCs. The first MVC, instructed prior to performance, to "kick as hard as possible" in order to elicit maximal torque production. The second MVC, instructed prior to performance, to "kick as hard and as fast as possible" in order to elicit a rapid increase in force production. The highest force value from the two trails was designated as the MVC for that respective visit and isometric ramp contractions thereafter. The order in of each MVCs was randomized for both visits.

3.3.3 Submaximal Contractions

Submaximal knee extensions during was performed in a randomized order. Figure 1 depicts the sequence of contractions. Following MVCs and a 2 minute rest period, 12 separate trapezoidal tracings were performed using target force trajectories using a 10%/s linear increase to target force, a 10 second isometric hold, and a -10%/s linearly decreasing segment back to baseline. Each of these randomized contractions were performed at 10%, 20%, and 50% MVC. Visual feedback was provided by real-time force feedback, allowing participants to accurately produce force that follow each of the 3 different templates. For the trapezoidal contractions, duration and intensity of the ramp-up and constant force hold portions are set with special consideration for MU recruitment and synchronization. Additionally, 2-minutes of rest was given between each contraction.

3.3.4 Electromyography and Signal Processing

Two separate sEMG sensors were placed on the VL of the right leg during testing. Prior to electrode placements, the skin was shaved, lightly abraded, and cleaned with alcohol. Locations for each sensor will placed, according to respective recommendations.

The first sensors placed on the VL was placed two-thirds the distance between the center of the muscle belly towards the distal tendon (Zaheer et al., 2012). a five-pin, 4-ch surface electrode array (Delsys, Inc., Natick, MA, USA). A reference electrode (Dermatrode; American Imex, Irvine, CA, USA) was placed over the spinous process of the C7 vertebrae and both were secured using hypoallergenic tape. Signals from the four channels of the dEMG array sensor were differentially amplified, filtered between 20 - 450 Hz, and samples at 20 kHz using a sixteen channel acquisition system (Bagnoli system, Delsys Inc., Natick, MA, USA) and recorded for off-line analysis.

The second sensor for HDsEMG signals recorded from the VL with a semi-disposal adhesive grid of 64-ch electrode (13 rows x five columns); gold-coated; diameter 1 mm; inter-electrode distance 8mm; OT Bioelecttronica, Turin, Italy). Using a reference line marked between the lateral side of the patella and the anterior superior iliac spine, an additional line on the distal portion of the muscle belly oriented 20° with respect to the reference line will be used for sensor placement. Following skin preparation, the electrode cavities were filled with conductive paste (SPES Medica, Salerno, Italy) and positioned between the proximal and distal tendons with columns oriented along the muscle fibers. Two reference electrode were dampened with water and positioned on the right wrist. HDsEMG signals were recorded in monopolar mode and digitally converted using a 16 bit multichannel amplifier (EMG – Quattrocento, 400 cannel EMG amplifier; OT Bioelecttronica; 3 dB, bandwidth 10-500 Hz). Signals were amplified (150x), sampled at 10240 Hz and bandpass filtered (10-500 Hz) before being stored for offline analysis.

Signal recordings with the force transducer were amplified (200x) and sampled at 2048 Hz with the external analogue-to-digital converter linked to both recording systems.

Feedback of the force signal will be provided from Delsys software and displayed on a monitor position directly in front of the participant.

3.3.5 EMG Decomposition: 4-ch Sensor

From the 4-ch sensor, four channels of raw sEMG signals recorded during each submaximal contraction. These were then stored on a personal computer and later decomposed offline using the PDIII algorithm described by De Luca et al. (2006) and improved upon by Newab et al. (2010). All MU's that do not demonstrate at least 90% accuracy, as assessed by the Decompose-Synthesize-Decompose-Compare (DeLuca and Contessa 2012) test were eliminated. Further analysis for the remaining MU's (those with >90% accuracy) was performed using custom-written software (LabVIEW 2017, National Instruments, Austin, TX, USA), which calculates the mean firing rate (MFR), relative recruitment threshold (RT%), interspike intervals (ISI) between each firing (time in ms), and the coefficient of variance (CoV; standard deviation normalized by the mean) of ISI distribution for each MU. The ISI distributions was displayed in histograms and manually inspected by the primary investigator for further validation. Accordingly, accepted MUs must: follow a relatively normal distribution of the ISI histogram, a positive RT% (i.e., no firings before the onset of force), a CoV < 30%, a range of ISI < 100 ms, and no separate clusters before or after the main distribution, which may indicate additional or missed firings during the decomposition. Additionally, MUs will be immediately discarded if a bimodal ISI distributions was present or if there was an insufficient spread/distribution of detected MUs (e.g. range of RT% found in a contraction must span at least10% MVC). Figure 2 provides representative examples of ISI histograms along with the resulting decision regarding keeping or discarding those

MUs. Motor units that meet the above criteria (i.e. those that will be kept) will have the instantaneous firing rates smoothed with a 1-s Hanning window, and the following variables will be calculated: MFR (in pulses per second) of individual MUAPs were calculated as the mean during the plateau of the smoothed curve; RT%, calculated as the relative force level (% MVC) at the onset of firing.

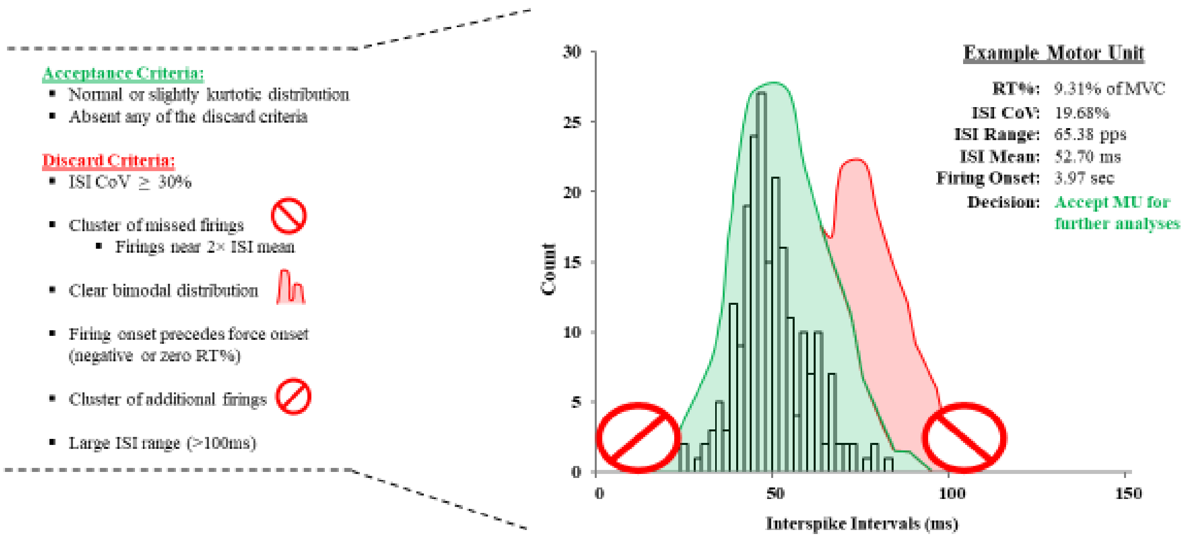

Figure 2 example of acceptance criteria for each MU collected using the 4-ch sensor and evaluated offline using custom build software to validate ISI CoV.

3.3.6 EMG Decomposition: 64-ch Sensor

Similarly, the sEMG signals collected from submaximal contractions were stored and decomposed offline using blind source separation (BSS) and manual inspection methods described by Holobar & Zazula (2007a,2007b)[36,37] which is commonly used to decompose and identify MU firing times across a broad range of forces[32]. These were manually edited to allow for the identification and removal from lower quality spikes that are not suitable for that respective train. Inclusion criteria for MU: display a signal-to-noise ratios ≥ 30 dB, and have no firing instances (relative to contraction types and intensities) separated by more than 2 s. Identification, addition, and removal of firing instances were carefully investigated, and followed standard operating procedures as discussed by Holobar & ZaZula (2007a,2007b)[36,37] and Del Vecchio et al 2020[38]. Those MUs that met inclusion criteria were further evaluated for MU firing behaviors (i.e., MFR, RT%).

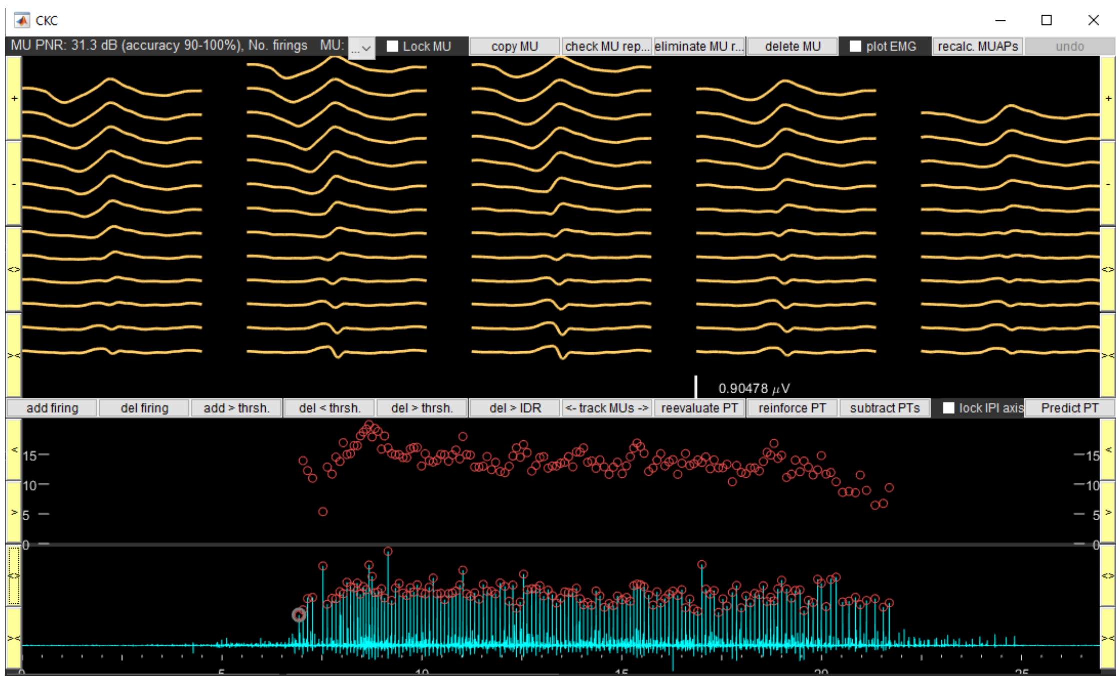

Figure 3. Example of the visual inspection and manual editing techniques used for MUs acquired by the 64-ch sensor and decomposed by BSS.

3.4 Statistical Analysis

Due to the differences in processing and validation techniques used from the two systems and recording devices, a 2-way mixed factorial analysis of variance

(ANOVA)(sensor [4-ch vs 64-ch] × contraction intensity [10% vs 20% vs 50%]) was used to examine mean differences in MU yield during all contraction performed during the study protocol. In the case of a significant interaction or main effect, follow-up analyses included 1-way repeated measures ANOVA to examine differences in MU yield between the two systems and Bonferroni corrected independent samples *t* test to examine differences between contraction intensities.

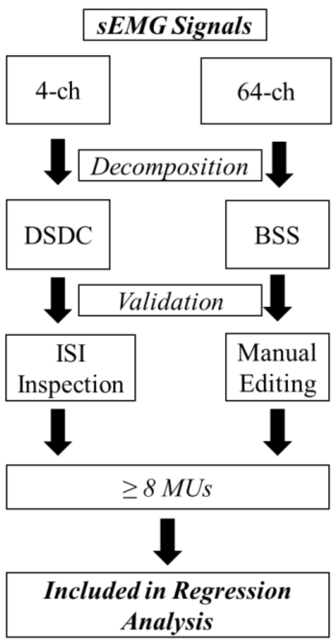

Figure 4 flow chart for MU validation and yield for inclusion in statistical analyses.

Separately, for validated MUs, linear regression coefficients were calculated using Excel (Microsoft Inc., Seattle, WA, USA) to determine slopes and y-intercepts of the RT versus MFR relationships during submaximal contractions ($RT/MFR_{10\%}$, $RT/MFR_{20\%}$, and $RT/MFR_{50\%}$). For each contraction intensity, a minimum of 8 MUs were needed to be include in the regression analysis. RT bin widths of 5% (e.g., 0-5, 5-

10%, etc.) were used to condense the data. The average for each bin was used to test the differences between slope coefficients and *y*-intercepts from the 4- and 64-ch sensors during $RT/MFR_{10\%}$, $RT/MFR_{20\%}$, and $RT/MFR_{50\%}$ (as described by Pedhazur 1997b). Due to only one subject meeting the inclusion criteria, regression analysis for $RT/MFR_{10\%}$ was performed on the firing properties of the subjects' single contraction at 10%. Paired samples *t* tests were used to compare RT between sensors at each intensity. All statistical analysis were performed using SPSS Statistics 24 (International Business Machines Corp., Armonk, NY, USA) and a priori alpha level of 0.05 and 0.10 was used to determine significance in ANOVA and linear regression slope and *y*-intercept comparisons, respectively.

CHAPTER IV

RESULTS

4.1 Number of Decomposed MUs

Following validation procedures from both recording systems, 925 out of 1480 and 698 out of 2553 MUs were kept from the recordings using the 4-ch and 64-ch sensors, respectively (Table 1). These were then used for further analysis in MU yield and subsequent firing behaviors. Figure 4 displays a flow chart that describes the procedures used for MU validation.

Table 1. Number of MUs seperately decomposed and validated at each contraction intensity

	4-ch			64-ch		
Intensity	Decomposed - PD III	ISI Validated	% Kept	Decomposed - BSS	Manually Edited	% Kept
10%MVC	333	90	27	682	259	38
20%MVC	508	331	65	779	250	32
50%MVC	639	504	79	1092	189	17
Total	1480	925	63	2553	698	27

The results of the 2-way mixed factorial ANOVA revealed a significant interaction between sensor and intensity for MU yield ($p < 0.05$), as well as a main effect for intensity ($p < 0.05$). Follow up 1-way ANOVA for the 4-ch sensor indicated that the MU yield at $50\%_{MVC} > 20\%_{MVC}$ and $10\%_{MVC}$ (12.6 ± 0.74 vs 8.28 ± 0.86 and 2.25 ± 0.61, respectively). Conversely, for the 64-ch sensor, $50\%_{MVC} < 20\%_{MVC}$ and $10\%_{MVC}$ (4.73 ± 0.89 vs 6.25 ± 1.20 and 6.48 ± 0.95, respectively). Bonferonni corrected independent samples *t* tests showed significant differences in MU yield between the two sensors at $10\%_{MVC}$ and $50\%_{MVC}$ ($p < 0.05$), but not during $20\%_{MVC}$ ($p = 0.18$).

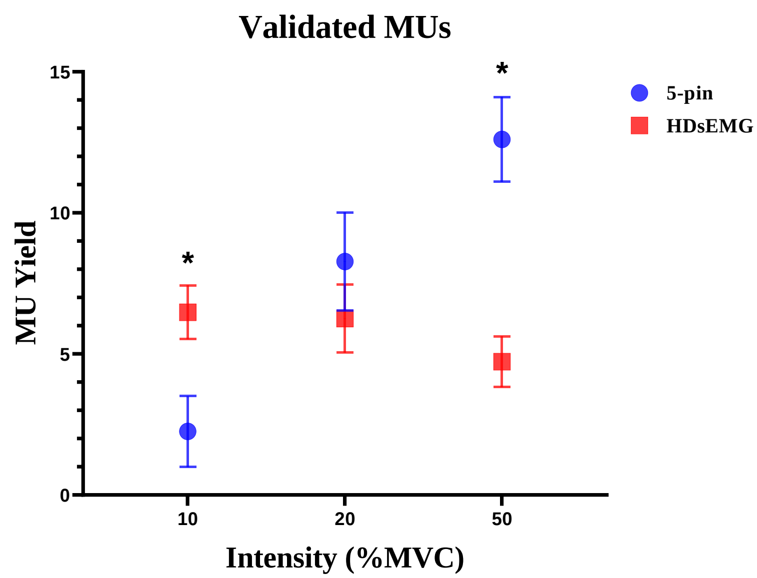

Figure 5 validated MUs from the 4-ch and 64-ch sensors during $10\%_{MVC}$, $20\%_{MVC}$, and $50\%_{MVC}$. Values presented as mean ± SE. * Significant difference between sensors

4.2 Recruitment Threshold and Mean Firing Rate

Table 2. Individual and grouped linear regression coefficents from recruitment threshold versus mean firing rates at each contraction intensity for 4-ch and 64-ch sensors.

		4-ch			64-ch		
		#MU	slope	*y*-intercepts	#MU	slope	*y*-intercepts
10%MVC	S008	16	-0.778	17.014	21	-0.365	11.538
20%MVC	S008	25	-0.529	18.155	28	-0.362	14.054
	S009	15	-0.305	13.952	12	-0.237	13.044
	Grouped	40	-0.262	15.058	40	-0.314	13.646
50%MVC	S002	16	-0.488	25.611	15	-0.145	14.225
	S006	18	-0.453	27.886	7	-0.173	14.072
	S008	17	-0.501	22.559	21	-0.285	20.708
	S009	14	-0.440	24.426	8	-0.104	15.425
	Grouped	65	-0.337	22.030	51	-0.196	16.855

Individual slope and y-intercept values are displayed in Table 2. A total of 7 contractions ($10\%_{MVC} = 1$, $20\%_{MVC} = 2$, $50\%_{MVC} = 4$) passed validation and MU yield inclusion criteria for linear regression analysis. Displayed in Figure 6a, for S008 $10\%_{MVC}$, there was a significant difference ($p = 0.015$) between the slopes of $RT/MFR_{10\%}$ recorded from the 4-ch and 64-ch sensors. However, for the grouped (i.e., bin) slopes during $20\%_{MVC}$, there were no significant difference in slope coefficients, but there were differences in y-intercepts ($p = 0.002$) of $RT/MFR_{20\%}$ (Figure 6b). For $RT/MFR_{50\%,}$ Figure 6c shows a significant difference between the slopes ($p = 0.008$) of firing relationships collected between the two sensors. Additionally, individual RT/MFR slopes ± 95% confidence intervals are displayed under $RT/MFR_{20\%}$ and $RT/MFR_{50\%}$.

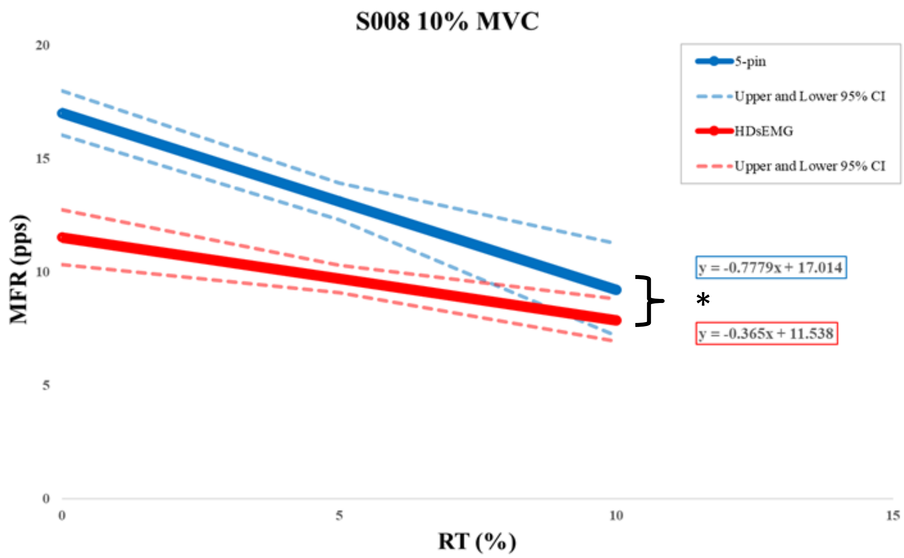

Figure 6a comparison of linear regression lines and upper and lower confidence intervals (CI) for RT/$MFR_{10\%}$ for $S008_{10\%}$ validated contraction. Data is presented from the calculated RT and MFR of the validated MUs, due to this being the only contraction that met inclusion criteria. Legend in **6a** is consistent throughout **6b** and **6c.** Regression coefficients (slope and *y*-intercepts) for each all validated contractions are displayed in *Table 2***. *** indicates significant difference between slopes ($p < 0.10$).

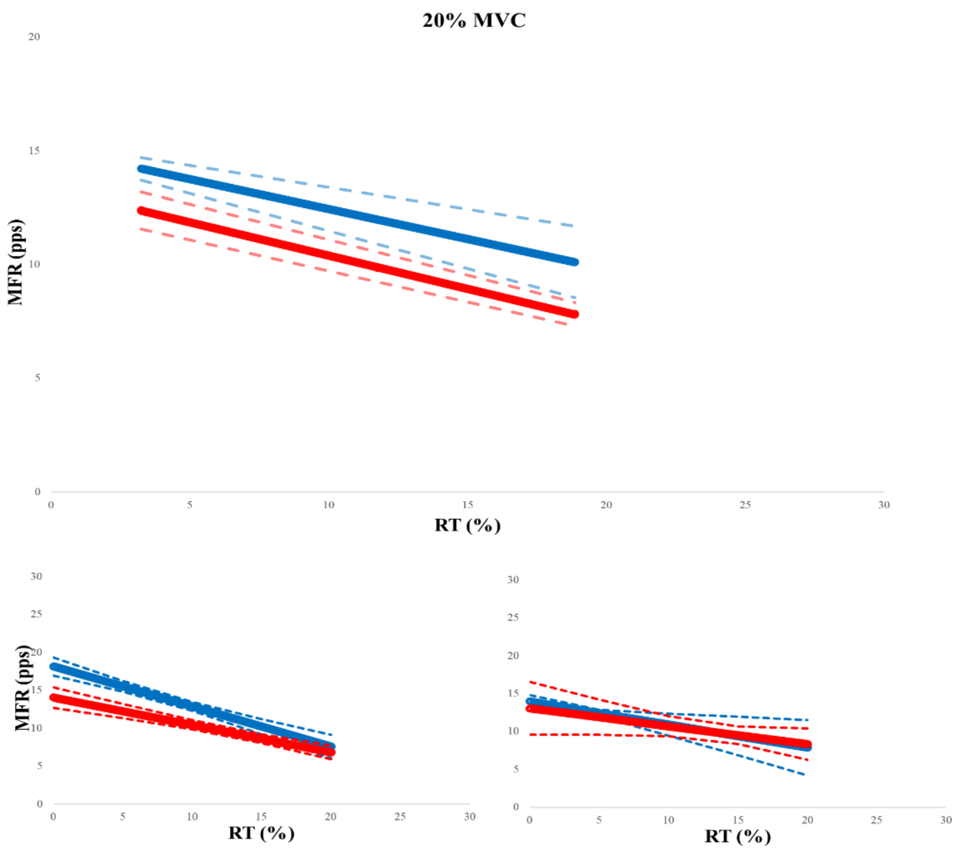

Figure 6b grouped (top) and individual (bottom) mean slope ± 95% CIs for comparisons of linear regression lines for the RT vs MFR relationships at 20%$_{MVC}$. ***** indicates significant difference between slopes ($p < 0.10$).

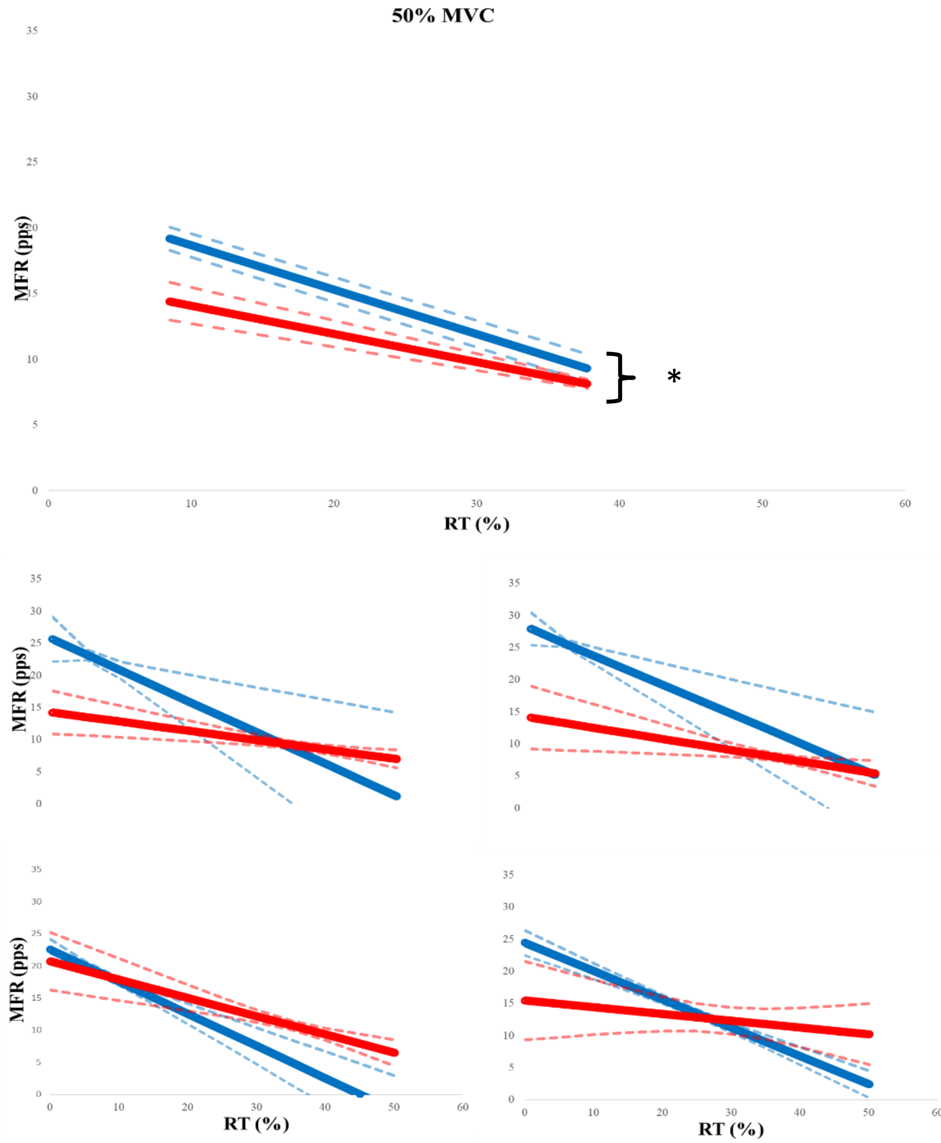

Figure 6c grouped (top) and individual (bottom) mean slope ± 95% CIs for comparisons of linear regression lines for the RT vs MFR relationships at 20%$_{MVC}$. ***** indicates significant difference between grouped mean slopes ($p < 0.10$).

During each intensity, the RT of validated MUs included in regression analysis, was significantly different ($p < 0.001$) between records from the 4-ch and 64-ch sensors.

Figure 7 shows grouped (20%$_{MVC}$ & 50%$_{MVC}$) and individual (10%$_{MVC}$) instantaneous RTs comparisons at 10, 20, and 50%$_{MVC}$.

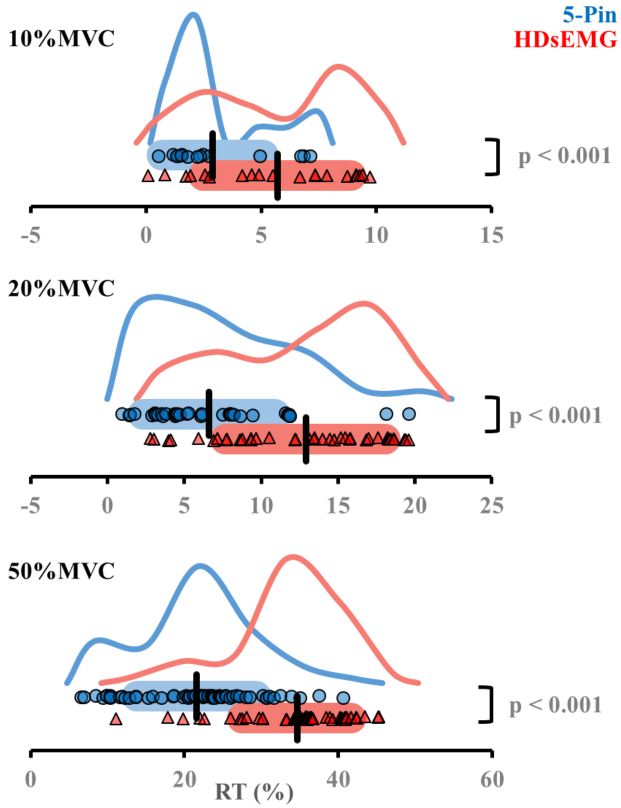

Figure 7 comparison of individual (S008 10%$_{MVC}$) and grouped (20%$_{MVC}$ and 50%$_{MVC}$) RT for validated MUs at each intensity level from the 4-ch and 64-ch sensors.

CHAPTER V

DISCUSSION

5.1 Implications and Significance

The purpose of this study was to concurrently record muscle activation from two different sEMG decomposition devices, and separately investigate respective signals for validated MU yield and firing behaviors for comparison. Accommodating for various constraints involved with sEMG decomposition, the researchers were able to offer interpretation of discrepancies regarding the analyses and editing techniques between the two recording devices. Several of these are influenced by a number of variables subject to inaccuracies and are greatly considered in the findings of this study.

5.2 MU Yield

Following collection of the sEMG, automated decomposition using the DSDC and BSS are markedly dependent on the quality of the acquired signal[17,33]. Thus, sEMG signals were visually inspected during contractions, and in the present study, subjects performed four separate contractions at each intensity to increase the probability of acquiring reliable signals. However, even with appropriate precaution, the amplitude and quality of the sEMG is subjective to muscle characteristics and the recording capabilities of the sensors used (i.e., “pick-up area). For example, prior to validation, the number of identified MUs from the 4-ch and 64-ch sensors were greater at each intensity and overall total (*Table 1).* The sEMG amplitude from each of these sensors would have likely influence the amount of MU action potentials identified since MU activity is uniquely represented by the surface action potential where it is recorded[17]. Given that the 64-ch array is a larger sensor, the number of electrodes would influence the amount of pick up area from contracting muscle, thus yielding a greater amount of MUs. In an investigation by Farina et al. (2008), the authors investigate the difference in identified MUs yielded from multichannel sEMG recordings similar to those of the present. In this study, the authors conclude that the relatively few MUs can be distinguished from the sEMG signal when fewer channels are utilized. This may have initially been applicable considering the greater amount of MUs identified from BSS, however, following manual editing only 27% of the decomposed MUs were kept compared to the 63% following ISI validation from DSDC.

Validation procedures from both devises extensively eliminated MUs at each intensity level. As shown in Figure 5, there were significant differences in MU yield between the 4-ch and 64-ch sensors at $10\%_{MVC}$ and $50\%_{MVC}$, and were likely due to ISI validation and manual inspection procedures, respectively (*Table 1*). Following ISI inspection at $10\%_{MVC}$, only 27% of the MUs were able to pass validation requirements. Of those eliminated, many were identified at a RT prior to the onset of force (i.e., IFR) or did not meet the ISI CoV (CoV < 30) inclusion criteria that removes the potential errors produced by DSDC[10,39]. Although a relatively similar amount of MUs decomposed by

BSS at 10%$_{MVC}$ were also eliminated, the manual editing techniques proved customized procedures that allows the investigator to postulate elements that are beneficial to acquiring precise information from the sEMG signal[38]. For example, the number of decomposition “runs”, or number of iterations to build upon each MU spike train, can be selected based on the estimated number of MU to be identified in the contraction[36,37]. The length of time in which the sEMG signal, and its initial offset, can be segmented into durations that facilitate the decomposition. Specifically, portions prior to the onset of the contraction may be contaminated with noise artifact from various sources and can be eliminated. These however are not possible methods that are available with decomposition techniques of the 4-ch sensor.

Conversely, for 50%$_{MVC}$, the number of validated MUs following respective decomposition were significantly greater for the 4-ch sensor compared to the 64-ch array. When first proposed, the ability of the 4-ch sensor to distinguish MUs of low- and high-thresholds during high intensity contractions was emphasized as a technological advancement in the capabilities of the PD III algorithm[12]. Although these capabilities are viewed as somewhat of a “black box”, the number of MUs that passed validation procedures yielded a significantly greater amount versus 64-ch sensor. The 64-ch sensor has shown to accurately identify high threshold MUs during contractions at higher intensity levels in muscle of the lower limb[12]. However, to our knowledge these have not been performed on the vastus lateralis using simultaneous sEMG decomposition techniques. The number of validated MUs from 64-ch sensor at this intensity level may have affected further investigation of firing behaviors. Discussed hereafter, the extraction and limitation of MUs during $50\%_{MVC}$ were not comprised in a manner that would allow for a qualitative spread in RT.

Following BSS decomposition, manual editing and inspection of the identified MUs are then performed on each of the trains and respective delta pulses. As depicted in Figure 5 of Del Vechio et al 2020[38], the investigator inspects each of the MUs in a series of three panels and can edit delta pulses (denoting the discharge times/firing times) allowing identifiable inter-discharge intervals to exhibit consistent behavior, or choose to eliminate the MU all together[36,37]. The regularity of the discharges (pps) during the contraction time (s) are clearly visible in an accurately identified MUs, and are typically unaffected by base-line noise (Figure 3). As mention previously, the base-line noise and movement artifact may be a limitation of the automated techniques of the DSDC decomposition, successively eliminating MUs of lower-thresholds that are labeled as pre-activated.

In the case that a train needs appropriate editing, MU discharges can be added or deleted from the MU spike train. However, applying these methods may be subjective to the user. In a recent tutorial, Del Vecchio and colleagues (2020) discuss the primary components of analyzing MU discharge characteristics recorded from 64-ch sensor. Of significance, the authors explain the subjectivity of the manual editing techniques, which provide a re-calculation of the MU spike train in order to optimize the accuracy in which the filtering of adding or removing pulses. These may have affected the outcome of the number of validated MU following decomposition.

5.3 Firing Behaviors- RT/MFR

The low amount of validated MUs were a direct limitation of subsequent analysis of linear regression coefficients from RT vs MFR. The 7 out of 120 contractions that did meet criteria from both validations were compared at each intensity. Caution must be taken when interpreting these results, nevertheless, these recording were from the same contraction and may offer some insight into the comparisons of validation techniques and regression coefficient comparisons.

Only one subject met the inclusion criteria for comparisons of slope and *y*-intercept in $RT/MFR_{10\%}$ (Figure 6a), showing a significant difference between the slopes of the two sensors. These differences (along with those illustrated in Figures 7b and 7c for RT/ $MFR_{20\%}$ and $RT/MFR_{50\%)}$ are to be expected due to the differences in RT for the validated MUs (Figure 7). As previously mentioned, the extensive validation of both systems may have eliminated MUs that would have offered a more diverse spread of RTs throughout the contractions. For many of the contractions that did not meet validation inclusion criteria, low-threshold MU recruited earlier on by the 4-ch sensor at $10\%_{MVC}$ were eliminated during ISI inspection due to early detection of initial discharges prior to force onset (potential baseline noise and movement artifact). However, in S008 $10\%_{MVC}$, these MU showed a lower RT with higher MFRs as depicted by the greater slopes and decline in the inverse relationship between RT/MFR. Although not as identifiable (Figure 6b), similar differences in RT of MUs during $20\%_{MVC}$ likely contributed to the differences in *y*-intercepts for $RT/MFR_{20\%}$. Of concern, the differences in RT between validated MUs at $50\%_{MVC}$ are well represented in the differences between the slopes of $RT/MFR_{50\%}$ (Figure 6c). Many of the aforementioned recording, decomposition, and manually editing techniques required by the 64-ch sensor can hinder the ability of analyzing a robust and meaningful population-based analyses of firing behaviors. The number of MUs identified at a higher RT% displayed a lower, less steep slope because of the lower MFRs associated with higher threshold MUs[2].

5.4 Conclusion

The purpose of this investigation was to determine potential differences in MU yield and subsequent firing behaviors between two commonly used sEMG decompositions recording devices. To the best of the authors' knowledge, this is the first investigation to examine sEMG recordings simultaneously recorded from the vastus lateralis muscle using the 4-ch and 64-ch sensors. The findings from the MU yield from separate decomposition and validation procedures support previous findings regarding the capabilities and limitations involved with sEMG. Unfortunately, these did hinder further application into subsequent firing behaviors that can be used to investigate useful information regarding MU properties. Yet, the reported differences in MU yield and RT/MFR slopes and y-intercepts affirm the 4-pin sensors ability to distinguish high-threshold MUs at greater intensity levels, and collectively follow the inverse relationships on a consistent basis[20]. Furthermore, this investigation is not limited to the direct conclusion of utilizing one technique or validation approach. However, the current findings can provide insight into advantages and disadvantages of decomposition and validation procedures of both. Although more MUs were validated from the 4-ch sensor, the low amount of MUs validated from the 64-ch sensor at higher contraction intensities did limit further investigation into the additional firing behaviors. These concerns follow the many discrepancies reported with investigator using the 64-ch sensor and the apprehension of successful decomposition of sEMG at higher intensities. Although not reported, concurrent two-source methods were used to further identify and validate MU firing properties (i.e., fine wire EMG). Additionally, inclusion of inter and intra-rater reliability may help to eliminate the subjectivity of MU validation and manual editing techniques. Therefore, further investigation and validation procedures would greatly benefit these preliminary findings.

REFERENCES

1. LeFever, R. S., & De Luca, C. J. (1982). A procedure for decomposing the myoelectric signal into its constituent action potentials-part I: technique, theory, and implementation. IEEE transactions on biomedical engineering, (3), 149-157.
2. De Luca, C. J., & Hostage, E. C. (2010). Relationship between firing rate and recruitment threshold of motoneurons in voluntary isometric contractions. Journal of neurophysiology, 104(2), 1034-1046.
3. De Luca, C. J., & Nawab, S. H. (2011). Reply to Farina and Enoka: the reconstruct-and-test approach is the most appropriate validation for surface EMG signal decomposition to date. Journal of neurophysiology, 105(2), 983-984.
4. Stegeman, D. F., Kleine, B. U., Lapatki, B. G., & Van Dijk, J. P. (2012). High-density surface EMG: Techniques and applications at a motor unit level. Biocybernetics and biomedical engineering, 32(3), 3-27.
5. Farina, D., Merletti, R., & Enoka, R. M. (2014). The extraction of neural strategies from the surface EMG: an update. Journal of Applied Physiology, 117(11), 1215-1230.
6. Farina, D., Negro, F., Gazzoni, M., & Enoka, R. M. (2008). Detecting the unique representation of motor-unit action potentials in the surface electromyogram. Journal of neurophysiology, 100(3), 1223-1233.
7. Farina, D., Negro, F., Muceli, S., & Enoka, R. M. (2016). Principles of motor unit physiology evolve with advances in technology. Physiology, 31(2), 83-94.
8. Farina, D., Merletti, R., & Enoka, R. M. (2004). The extraction of neural strategies from the surface EMG. *Journal of applied physiology*, *96*(4), 1486-1495.
9. Holobar, A., Farina, D., Gazzoni, M., Merletti, R., & Zazula, D. (2009). Estimating motor unit discharge patterns from high-density surface electromyogram. Clinical Neurophysiology, 120(3), 551-562.
10. Holobar, A., Minetto, M. A., Botter, A., Negro, F., & Farina, D. (2010). Experimental analysis of accuracy in the identification of motor unit spike trains from high-density surface EMG. IEEE Transactions on Neural Systems and Rehabilitation Engineering, 18(3), 221-229.

11. B Holobar, A., Minetto, M. A., & Farina, D. (2014). Accurate identification of motor unit discharge patterns from high-density surface EMG and validation with a novel signal-based performance metric. Journal of neural engineering, 11(1), 016008.
12. De Luca, C. J., Adam, A., Wotiz, R., Gilmore, L. D., & Nawab, S. H. (2006). Decomposition of surface EMG signals. Journal of neurophysiology, 96(3), 1646-1657.
13. Enoka, R. M. (2019). Physiological validation of the decomposition of surface EMG signals. Journal of Electromyography and Kinesiology, 46, 70-83.
14. DeFreitas, J. M. (2019). In regards to motor unit decomposition, are we caring about the right information?. Journal of electromyography and kinesiology: official journal of the International Society of Electrophysiological Kinesiology, 47, 121-122.
15. Farina, D., & Enoka, R. M. (2011). Surface EMG decomposition requires an appropriate validation. Journal of neurophysiology, 105(2), 981-982.
16. Del Vecchio, A., Casolo, A., Negro, F., Scorcelletti, M., Bazzucchi, I., Enoka, R., ... & Farina, D. (2019). The increase in muscle force after 4 weeks of strength training is mediated by adaptations in motor unit recruitment and rate coding. The Journal of physiology, 597(7), 1873-1887.
17. Martinez-Valdes, E., Laine, C. M., Falla, D., Mayer, F., & Farina, D. (2016). High-density surface electromyography provides reliable estimates of motor unit behavior. Clinical Neurophysiology, 127(6), 2534-2541.
18. Farina, D., & Negro, F. (2015). Common synaptic input to motor neurons, motor unit synchronization, and force control. *Exercise and sport sciences reviews*, *43*(1), 23-33.
19. Farina, D., Merletti, R., & Enoka, R. M. (2015). Reply to De Luca, Nawab, and Kline: The proposed method to validate surface EMG signal decomposition remains problematic. Journal of Applied Physiology, 118(8), 1085-1085.
20. Nawab, S. H., Chang, S. S., & De Luca, C. J. (2010). High-yield decomposition of surface EMG signals. *Clinical neurophysiology*, *121*(10), 1602-1615.
21. Mambrito, B., & De Luca, C. J. (1984). A technique for the detection, decomposition and analysis of the EMG signal. Electroencephalography and clinical neurophysiology, 58(2), 175-188.
22. Hu, X., Rymer, W. Z., & Suresh, N. L. (2013). Motor unit pool organization examined via spike-triggered averaging of the surface electromyogram. *Journal of neurophysiology*, *110*(5), 1205-1220.
23. Hu, X., Rymer, W. Z., & Suresh, N. L. (2013). Reliability of spike triggered averaging of the surface electromyogram for motor unit action potential estimation. Muscle & nerve, 48(4), 557-570.
24. McManus, L., Hu, X., Rymer, W. Z., Suresh, N. L., & Lowery, M. M. (2016). Muscle fatigue increases beta-band coherence between the firing times of simultaneously active motor units in the first dorsal interosseous muscle. Journal of neurophysiology, 115(6), 2830-2839.

25. McManus, L., Hu, X., Rymer, W. Z., Suresh, N. L., & Lowery, M. M. (2017). Motor unit activity during fatiguing isometric muscle contraction in hemispheric stroke survivors. Frontiers in human neuroscience, 11, 569.
26. Hu, X., Suresh, N. L., Jeon, B., Shin, H., & Rymer, W. Z. (2014, August). Statistics of inter-spike intervals as a routine measure of accuracy in automatic decomposition of surface electromyogram. In 2014 36th Annual International Conference of the IEEE Engineering in Medicine and Biology Society (pp. 3541-3544). IEEE.
27. Hu, X., Rymer, W. Z., & Suresh, N. L. (2013). Assessment of validity of a high-yield surface electromyogram decomposition. Journal of neuroengineering and rehabilitation, 10(1), 99.
28. De Luca, C. J., Nawab, S. H., & Kline, J. C. (2015). Clarification of methods used to validate surface EMG decomposition algorithms as described by Farina et al.(2014). Journal of Applied Physiology, 118(8), 1084-1084.
29. Fuglevand, A. J., Lester, R. A., & Johns, R. K. (2015). Distinguishing intrinsic from extrinsic factors underlying firing rate saturation in human motor units. Journal of neurophysiology, 113(5), 1310-1322.
30. Haun, C. T., Mumford, P. W., Roberson, P. A., Romero, M. A., Mobley, C. B., Kephart, W. C., ... & Mackey, C. S. (2017). Molecular, neuromuscular, and recovery responses to light versus heavy resistance exercise in young men. Physiological reports, 5(18), e13457.
31. Pope, Z. K., Hester, G. M., Benik, F. M., & DeFreitas, J. M. (2016). Action potential amplitude as a noninvasive indicator of motor unit-specific hypertrophy. Journal of neurophysiology, 115(5), 2608-2614.
32. Del Vecchio, A., Ubeda, A., Sartori, M., Azorin, J. M., Felici, F., & Farina, D. (2018). Central nervous system modulates the neuromechanical delay in a broad range for the control of muscle force. Journal of applied physiology, 125(5), 1404-1410.
33. Negro, F., Muceli, S., Castronovo, A. M., Holobar, A., & Farina, D. (2016). Multi-channel intramuscular and surface EMG decomposition by convolutive blind source separation. Journal of neural engineering, 13(2), 026027.
34. Mrówczyński, W., Celichowski, J., Raikova, R., & Krutki, P. (2015). Physiological consequences of doublet discharges on motoneuronal firing and motor unit force. Frontiers in Cellular Neuroscience, 9, 81.
35. Defreitas, J. M., Beck, T. W., Ye, X., & Stock, M. S. (2014). Synchronization of low-and high-threshold motor units. Muscle & nerve, 49(4), 575-583.
36. Holobar, A., & Zazula, D. (2007, September). Gradient convolution kernel compensation applied to surface electromyograms. In International Conference on Independent Component Analysis and Signal Separation (pp. 617-624). Springer, Berlin, Heidelberg.
37. Holobar, A., & Zazula, D. (2007). Multichannel blind source separation using convolution kernel compensation. IEEE Transactions on Signal Processing, 55(9), 4487-4496.

38. Del Vecchio, A., Holobar, A., Falla, D., Felici, F., Enoka, R. M., & Farina, D. (2020). Tutorial: Analysis of motor unit discharge characteristics from high-density surface EMG signals. Journal of Electromyography and Kinesiology, 102426.
39. Hernandez-Sarabia J, Luera M, Barrera-Curiel A, Estrada C, Defreitas J. Does strict criteria for individual motor units alter population based models for the motor unit pool? Journal of Neural Engineering. 2020 In Review
40. Piervirgili, G., Petracca, F., & Merletti, R. (2014). A new method to assess skin treatments for lowering the impedance and noise of individual gelled Ag–AgCl electrodes. Physiological measurement, 35(10), 2101.

APPENDICES

School of Kinesiology, Applied Health, and Recreation

INFORMED CONSENT FOR PARTICPATION IN THE STUDY

Comparison of Techniques in Decomposition Electromyography

Background Information

You are invited to be in a research study investigating various techniques in muscle activation and analysis of the leg muscles during the knee extension exercise. You were selected as a possible participant because you meet the requirements for inclusion, being recreationally active, lower-body resistance trained, and having no current or previous neuromuscular disorders. We ask that you read this form and ask any questions you may have before agreeing to be in the study. Your participation is entirely voluntary.

This study is being conducted by: Mike Luera, under the direction of Dr. Jason DeFreitas, School of Kinesiology, Applied Health, and Recreation, Oklahoma State University

Procedures

If you agree to be in this study, we would ask you to do the following things: Participants will visit the Applied Neuromuscular Physiology Laboratory on 5 separate occasions, each lasting approximately 45-90 minutes. Prior to any data collection, subjects will be required to sign an informed consent document, a health history questionnaire, and a physical activity readiness questionnaire (PAR-Q). The health history questionnaire (HHQ) will provide us with detailed information regarding subjects' current health and physical activity status from which we can determine their eligibility to participate in this study. The familiarization visit (1st visit) will consist of collecting age, height, weight, and a few brief, maximal contractions of the legs. Each subject will be de-identified and given a participation number. All descriptive information will be transposed onto an electronic document and stored on a laboratory/department computer. All documents will be locked away in a file cabinet of the PI's laboratory office.

On Visits 2-5, subjects will undergo standardized warm-up. After warm-up, sensors will be placed on the surface of the skin of the thigh to non-invasively measure the electrical activity of a muscle. During 2 of the visits, recordings from fine-wire EMG will be collected. Prior to sensor placement, the skin will be shaved at the aforementioned areas with a razor, lightly abraded, and cleaned with isopropyl alcohol. All P.I.'s have successfully completed blood born pathogen training through the Environmental Health & Safety Office. Additionally, each investigator will be thoroughly trained on all procedures and necessary safety measures involved with fine-wire EMG.

A total of 18 contractions will be performed during each visit with 1-2 min. to rest in between each one. In addition, ultrasound will be used to examine the muscle before contractions to allow for proper placement of each sensor.

During all testing, participants will be strongly encouraged to verbally express to the PI and or co-PI's at any time if they are experiencing any lightheadedness, pain, or any other physical problems. In addition, participants will be encouraged to contact the PI's if they experience pain, injury or other complication as a result from the study.

Ultrasound:

Ultrasound (or sonography) is a test that uses high-frequency sound waves to show what is inside your body. You will lie on a cushioned table and gel will be applied to your skin; the gel acts as a conductor. A transducer, a hand-held device that sends and receives ultrasound signals, is moved over the area of the body being imaged. Images instantly are seen on a television-like monitor and sent to film or videotape for a specialist to review and interpret.

Fine-wire EMG:

Briefly, fine-wire EMG involves inserting a thin gauge needle (29 gauge or 0.34 mm diameter) with extremely thin wire, thinner than the thickness of a hair, through the skin and into the muscle. The needle itself is then removed leaving the very thin wire inserted into the muscle. The residual wire coming out from the skin surface will be connected to a recording device that will record the electrical activity of the muscle contraction.

Low-Frequency Nerve Stimulation
A low-frequency impulse will be sent through femoral nerve and elicit a small impulse that will cause a short muscle action following each contraction.

Participation in the study involves the following time commitment: Each of the 5 visit will last approximately 45-90 minutes depending on the order of randomized testing conditions. Specifically, the preparation for fine-wire may add time due to the procedures involved.

Risks and Benefits of being in the Study

The study involves the following foreseeable risks: Possible risks that could occur from the specific protocols include delayed onset muscle soreness and temporary blood pressure/heart rate elevation due to resistance-training movements.

Fine-wire EMG:
Possible bleeding, inflammation, or bruising following insertion and extraction of fine wire electrodes. Minimal risks involved during the protocol, however, subject may experience some discomfort for the initial insertion of the fine wire electrodes and/or during the muscle contractions required throughout the testing protocol. Examples may include inflammation of the insertion site, hematoma or slight bleeding of the area, or potential vaso-vagal episode (fainting in response to procedures). Proper precaution will be taken to remove biohazardous material and to inform the participant of any of these potential occurrences. There is minimal risk of infection at the insertion site because sterile procedures will be followed. The electrodes and the needles will be fully autoclaved and sterilized before use on an individual and electrodes and needles used on an individual will not be used on other persons. Insertion needles will be disposed of in a biomedical waste container immediately following the use. Only individuals that have undergone training will perform the techniques. The likelihood of lightheadedness or fainting is moderate to minimal. Medical records will only be used during the screening process. In case of injury or illness resulting from this study, emergency medical treatment will be available (CPR certified investigators and 911). No funds have been set aside by Oklahoma State University to compensate you in the event of illness or injury. It is important to note that you are free to withdraw from the study at any time without prejudice or penalty.

Ultrasound:
The gel may be sticky, but the test should not cause any pain or discomfort.

Low-Frequency Nerve Stimulation
The stimulation to the femoral nerve may cause slight discomfort to the applied area and/or the contracting leg muscles.

The benefits to participation are: The benefits which may reasonably be expected to result from this study are, an understanding of muscle activation patterns during the leg extension exercise.

Compensation

You will receive no payment for participating in this study.

Confidentiality

The information that you give in the study will be handled confidentially. Your information will be assigned a code number/pseudonym. The list connecting your name to this code will be kept in a locked file. When the study is completed and the data have been analyzed, this list will be destroyed. Your name will not be used in any report

We will collect your information through the testing procedures mentioned above. All data and information will be stored on a password locked computer that is not accessible to anyone other than the primary investigator. Research records will be stored securely and only researchers and individuals responsible for research oversight staff will have access to the records. All data forms will contain a unique code given to you, and will be secured separately from the health history questionnaire and consent form. It is possible that the consent process and data collection will be observed by research oversight staff responsible for safeguarding the rights and well-being of people who participate in research. Any written results will discuss group findings and will not include information that will identify you. Following the conclusion of the de-identified data, for research presentations and peer-review manuscript submissions, the keys to all coded data will be properly discarded and unable to be accessed thereafter.

This is expected to occur no later than May 2020. This informed consent form will be kept for a required minimum of 3 years after the study is complete, and then it will be destroyed. Your data collected as part of this research project, may be used or distributed for future research studies.

It is unlikely, but possible, that others responsible for research oversight may require us to share the information you give us from the study to ensure that the research was conducted safely and appropriately. We will only share your information if law or policy requires us to do so.

Contacts and Questions

The Institutional Review Board (IRB) for the protection of human research participants at Oklahoma State University has reviewed and approved this study. If you have questions about the research study itself, please contact the Principal Investigator at 405, [E-mail address]. If you have questions about your rights as a research volunteer or would simply like to speak with someone other than the research team about concerns regarding this study, please contact the IRB at (405) 744-3377 or irb@okstate.edu. All reports or correspondence will be kept confidential.

You will be given a copy of this information to keep for your records.

Statement of Consent

I have read the above information. I have had the opportunity to ask questions and have my questions answered. I consent to participate in the study.

Indicate Yes or No:

I give consent for my data to be used in future research studies:
___Yes ___No

I give consent to be contacted for follow-up in this study or future similar studies:
___Yes ___No

Signature:____ Date: _________

Signature of Investigator:________________________________ Date: _________

VITA

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Doctor of Philosophy

Dissertation: A COMPARISON OF TECHNIQUES FOR DECOMPOSING SURFACE EMG SIGNALS INTO MOTOR UNIT ACTION POTENTIAL TRAINS

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Research:

- **Luera MJ,** Dowling B, Muddle TWD, Jenkins NDM. Differences in rotational kinetics and kinematics for professional baseball pitchers with higher- versus lower-pitch velocities. *Journal of Applied Biomechanics*. (Accepted, December 2019).
- **Luera MJ**, Dowling B, Magrini MA, Muddle TWD, Colquhoun RJ, Jenkins NDM. Rotational kinematics may play an important role in minimizing elbow varus torques for professional versus high school pitchers. *The Orthopaedic Journal of Sports Medicine*. (2018).
- **Luera, M.J.**, Stock, M.S., and A.D.W. Chappell. Electromyographic amplitude versus concentric and eccentric squat force relationships for monoarticular and biarticular thigh muscles. *Journal of Strength and Conditioning Research*, 28(2):328–338, 2014