Title:

A novel methodological framework for the assessment of the neural control of the shoulder
using high-density surface electromyography.

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ABSTRACT

The complex function of the shoulder relies on the coordinated activation of small and large muscles, including the deltoid, pectoralis major, trapezius, and latissimus dorsi. However, detailed knowledge of their neuromuscular control remains limited. This study aimed to develop a methodological framework to investigate the neural control of the larger superficial shoulder muscles by combining a six-degree-of-freedom load cell attached to a robotic arm with high-density surface electromyograms (HDsEMG). Six healthy participants performed isometric contractions (abduction, adduction, flexion, and extension) at 30% of maximal voluntary contraction with the shoulder positioned at 30° and 65° of lateral abduction. HDsEMGs were recorded from the four muscles and analysed at the global activation, spatial distribution of activation and motor unit levels. Global activation was quantified using averaged normalized root-mean-square (RMS) amplitude and spatial distribution using coefficient of variation of the topographic maps. Moreover, HDsEMGs were decomposed into individual motor unit spike trains using convolutive blind source separation, and motor unit behaviour was characterized by mean discharge rate and spatial distribution of motor unit action potentials (MUAPs). RMS maps revealed action-specific activation within and between muscles, with the upper trapezius active across all tasks, while the anterior, middle, and posterior deltoid, clavicular pectoralis major, and latissimus dorsi were predominantly activated during abduction, flexion, and extension. Motor unit discharge rate also showed task-dependent activity. MUAP spatial distributions further showed distinct motor unit territories within arrays, suggesting region-specific recruitment strategies across actions. In conclusion, this framework demonstrates that individual motor unit activity can be reliably measured non-invasively in the superficial shoulder muscles. The approach provides a methodological basis for novel incorporation of neural control information into biomechanical models of shoulder function.

64 **Keywords**: motor unit discharge rate, HDsEMG spatial distribution, neural control, shoulder

65 biomechanics.

1.0 INTRODUCTION

Actions of the humerus require the complex activation of the muscles surrounding the glenohumeral joint to contract and, ultimately, cause the desired action. During these actions, the activation of large muscles such as the *trapezius, pectoralis major, latissimus dorsi,* and *deltoid* must be precisely coordinated with several smaller muscles, in particular the rotator cuff elements, to generate torque across multiple joints and adapt to changes in both task demands and musculoskeletal alterations, while also maintain glenohumeral stability in a delicate balance (Veeger & van der Helm, 2007). According to Hughes and colleagues (1999), the muscles of the glenohumeral joint (hereafter referred to as the shoulder) and their strength are important components of functional capacity. Thus, understanding the neuromuscular behaviour of these muscles is essential for a complete understanding of the activation and function of this muscle group, which can inform the prescription of appropriate training and rehabilitation strategies. Biomechanical modelling of the shoulder relies on simple mechanical analyses of body segments and the internal and external forces acting on the joint to predict shoulder muscle control (Hughes & An, 1997; Chang *et al.*, 2000; Dickerson *et al.*, 2008; Seth *et al.*, 2019). However, to date, there are restrictions on modelling advances in the shoulder, particularly in including neural control of active muscles and, specifically, their partitional behaviour. Simplifying assumptions include single on/off activation of entire muscles or semi-arbitrarily defined mechanical elements. These limitations stem from the historical methodological approach to bipolar surface electromyogram recordings for inferring the intricate behaviour of the individual motor units involved in muscle activation. In fact, to date, the inclusion of motoneuron behaviour from the shoulder muscles in neuromechanical models remains elusive (Hughes & An, 1997). Therefore, shoulder models must rely on indirect measures of neural control, using metrics that only partially capture the complexity of muscle activation strategies (Farina *et al.*, 2010; Lulic-Kuryllo *et al.*, 2021*a*).

Classically, the approach to assessing muscular neural control has been global bipolar

surface electromyography (EMG), using amplitude measures to infer neuromuscular

adaptations to specific actions with applications in biomechanics (Lawrence & De Luca, 1983;

Woods & Bigland-Ritchie, 1983; Hermans *et al.*, 1999; Day & Hulliger, 2001; Karlsson &

Gerdle, 2001; Keenan *et al.*, 2005; Farina *et al.*, 2010). However, the specificity of EMG

amplitude in inferring neural drive to the muscle remains debated (Christie *et al.*, 2009; Farina

et al., 2010). Amplitude cancellation of surface motor unit action potentials is a key factor

affecting the relationship between EMG amplitude and neural drive (Yao *et al.*, 2000; Day &

Hulliger, 2001; Dimitrova & Dimitrov, 2003; Keenan *et al.*, 2005), because as additional motor

units may be recruited during the gradation of force, the superposition of positive and negative

phases of action potentials could result in a non-proportional increase in EMG amplitude (Day

& Hulliger, 2001; Keenan *et al.*, 2005). Additionally, bipolar EMG provides only muscle

activity in the recording volume of the electrodes (i.e., local information). In the case of the

shoulder, where large muscles produce complex actions across multiple degrees of freedom,

not all muscle portions may be either monitored or active during all actions (i.e., within-muscle

localized activation) (Lulic-Kuryllo *et al.*, 2021*a*) via standard bipolar surface EMG. For

instance, previous evidence suggested that localized regions of the deltoid, pectoralis major,

and latissimus dorsi can be activated based on the specific shoulder action direction (Wickham

& Brown, 1998, 2012; Brown *et al.*, 2007; Lulic-Kuryllo *et al.*, 2022). Finally, when recording

myoelectric signals with global bipolar EMG, the risk of crosstalk must be considered. Signals

generated by secondary muscles may lead to the erroneous interpretation of myoelectric signals

from the primary muscle under investigation (De Luca & Merletti, 1988). Therefore, caution

must be taken when assessing muscle activity using bipolar global signals, given the possibility

of cancellation or crosstalk in the myoelectric recordings.

Advancements in myoelectric recordings in the early 1980's have led to high-density

surface electromyograms (HDsEMG) (Masuda *et al.*, 1983, 1985; Reucher *et al.*, 1987;

Merletti *et al.*, 1999). The benefit of recording with the evolving HDsEMG technology is its

non-invasive nature and the ability to provide the spatial distribution of activation within and

across muscles during complex actions. Additionally, when combined with motor unit

decomposition algorithms (Holobar & Zazula, 2007; Ning *et al.*, 2015; Chen & Zhou, 2016;

Negro *et al.*, 2016), the approach has the potential to record a large number of concurrently

discharging motoneurons during voluntary contractions (Inglis *et al.*, 2025). As the alpha

motoneuron is the final common pathway of the neuromuscular system, integrating synaptic

inputs from spinal and supraspinal pathways (Liddell & Sherrington, 1925; Sherrington, 1925),

the analysis of motor unit discharge behaviour provides an effective way to assemble

information on the generation and control of actions. A recent systematic review and meta-

analysis revealed that most motor unit behaviour investigations have focused on the lower

rather than the upper-body, leading to a paucity of data on the upper limb (Inglis *et al.*, 2025).

Additionally, it was revealed that of the upper limb muscles, specifically major movers of the

shoulder, there are few motor unit behaviour studies investigating the *upper trapezius*

(Westgaard & De Luca, 2001; Falla & Farina, 2008*a*, 2008*b*; Gerdle *et al.*, 2008; Holobar *et*

al., 2009; Dideriksen *et al.*, 2016; Kirk *et al.*, 2019) and *pectoralis major* (Lulic-Kuryllo *et al.*,

2021*b*) using invasive or non-invasive techniques*,* none that examined the *latissimus dorsi*

(Padovan *et al.*, 2024) and only three examining the *deltoid* (De Luca *et al.*, 1982; Doherty &

Stashuk, 2003; Baslo *et al.*, 2024), which all used intramuscular electrodes (Joseph *et al.*,

2019).

Therefore, the aim of this paper was to develop a novel methodological framework to

investigate four isometric shoulder actions (abduction, adduction, flexion and extension) at two

shoulder elevation angles to fully assess and compare the neural control of the superficial

shoulder muscles (deltoid, clavicular pectoralis major, upper trapezius and latissimus dorsi) at the global activation level (average HDsEMG amplitude), the spatial distribution of activation level (coefficient of variation of the HDsEMG amplitude maps) and the motor unit level (discharge patterns). For this, we combined a six-degree-of-freedom load cell with a robotic arm with the evolving HDsEMG technology. Additionally, this paper provides several examples of the type of extractable neural shoulder control information within the proposed framework. The novel framework proposed aims to establish guidelines for future investigations of the shoulder complex's large muscles and to advance shoulder musculoskeletal modelling.

2.0 METHODS

2.1 Participants

Six healthy individuals free of any musculoskeletal or neuromuscular abnormalities (1 female; mean ± SD: age 28 ± 4 years; height 180 ± 7 cm; mass 79 ± 9 kg) voluntarily participated. All participants provided written informed consent before starting the experiments that conformed to the standards set by the latest version of the Declaration of Helsinki, which was approved by the ethics committee of the Università degli Studi di Brescia (code NP5890).

2.2 Experimental protocol

2.2.1 Experimental setup

Each participant visited the lab on a single day where, in a randomized order, they performed multiple degree-of-freedom isometric contractions in a 2-hour session. Participants were seated in a rigid testing chair with their hip and knee joints at 90° of flexion (0° indicates full extension) and a broad Velcro® strap placed around the midsection to minimize lateral torso movements. The participant's arm was extended and placed slightly (<20°) in the scapular plane to avoid discomfort when performing the selected actions. The participants'

upper arm was placed into a custom-built testing jig manufactured from rectangular metal

tubing to ensure a rigid transfer of forces produced to the load cell (**Figure 1b**). The testing jig

was attached to a 6-degrees of freedom load cell (© 2025 Advanced Mechanical Technology

Inc., Watertown MA, USA, model #MC3A) to measure force output (**Figure 1c**). The load cell

has a capacity of F_x, F_y of 225 N and F_z of 445 N, and a sensitivity of F_x, F_y of 5.4 µV/V-N and

F_z of 1.35 µV/V-N. The load cell was attached to a robotic arm (Kawasaki Robotics Inc.,

Wixom MI, USA; **Figure 1a**) with a custom-machined aluminium coupling (**Figure 1d**). The

importance of implementing the robotic arm was to position the testing jig and load cell through

fine adjustments to accommodate individual anatomical landmarks, ensuring participants

performed each action in identical relative positions and enabling precise recording of the

forces produced during each action. Once positioned, the robotic arm was switched off and

disconnected from all electrical sources to minimize electrical interference in the EMG signal.

Figure 1. Mechanical components designed and manufactured for the robotic setup *(a)*
Complete robot assembly showing the robot with all components mounted. (b) 3D model and
2D technical drawings of the jig. (c) 3D model and 2D technical drawings of the six degree-
of-freedom load cell. (d) 3D model and 2D technical drawings of the coupling between the
robotic arm and the load cell. Measurements are reported in millimetres (mm).

2.2.2 Experimental tasks

Each participant performed four isometric contractions at 30° and 65° of lateral

shoulder abduction. The actions were: abduction, adduction, flexion and extension. **Figure 2**

illustrates the position of the participant during the four actions across two shoulder angles. In

Figure 2, the direction of the force vector is depicted with arrows for each action. Each action

was performed at two shoulder angles to alter the deltoid muscle length (30° long; 65° short)

and optimize the angles where the deltoid can initiate action during abduction (Peterson &

Rayan, 2011).

All experimental actions were repeated twice for each action and shoulder angle,

resulting in a total of 16 trials (4 actions × 2 shoulder angles × 2 trials). Prior to each condition

(angle/action), participants performed three voluntary maximum isometric contractions

(MVCs) for 3-5 s, with a 3-min rest between each bout. The greatest value across the three

MVCs was the maximal isometric shoulder force output for the individual condition and was

used as a reference to set the target for the submaximal contractions.

Subsequently, participants underwent a short familiarization period, during which they

were instructed to exert isometric shoulder forces at 10% MVC for approximately 10 s. This

familiarization task was repeated up to a maximum of five times to ensure that participants

could maintain the force output as steadily as possible at a target level, which was visually

assessed by a researcher (JGI). Following the familiarization period, participants were

instructed to perform two isometric contractions following a trapezoidal profile. The

trapezoidal profile involved a linear increase at a rate of 3% MVC/s, a plateau phase for 30 s,

and a linear decrease at a rate of 3% MVC/s. The target force output was set at 30% MVC (i.e.,

moderate force output) to mimic common outputs during activities of daily living (Lulic-

Kuryllo *et al.*, 2021*a*).

Abduction / Adduction **Flexion / Extension**

Shoulder at 30°

Shoulder at 65°

Figure 2. Experimental Actions. Shoulder abduction/adduction (left, anterior view) and flexion/extension (right, lateral view) performed at 30° (top) and 65° (bottom) of shoulder abduction. The arm was secured to a passive robotic interface, which secured the participant in position for each given action. A broad black Velcro® strap was placed around the midsection and secured to the testing chair to minimize actions of the trunk.

2.2.3 Force feedback

A novel graphical user interface (GUI) was developed in MATLAB (version 2024a) to provide visual feedback of the force output and the rate at which the force was to be developed during the trapezoidal profile (**Figure 3e**). Force feedback was provided based on the axis being investigated relative to the three-axis load cell connected to the PC by an A-D board (National Instruments CORP., NI USB-6343). The selected axis was represented by either a horizontal or vertical line, where the Y direction was utilized for abduction and adduction, and the Z direction during flexion and extension (frontal) relative to the load cell. The GUI provided a green target box with the dimensions set to ± 2.5% MVC and a blue circle to represent the changes in force output. Each participant was instructed to maintain the blue circle inside the

square. In cases where the error in force production exceeded 2.5% MVC, the square would

turn red, and the participant would be encouraged to adjust the force output to meet the target

force. Visual feedback was displayed on a monitor placed 1.3 meters in front of the participant

at eye level.

2.3 Data collection

HDsEMG were acquired from the four major humeral movers (anterior, lateral and

posterior deltoid, clavicular pectoralis major, upper trapezius, and latissimus dorsi) during the

submaximal isometric actions using six two-dimensional adhesive arrays (HD08MM1305 and

HD08MM1606; 1 mm diameter; 8 mm inter-electrode distance; OT Bioelettronica, Turin,

Italy). The arrays placed over the deltoid and clavicular pectoralis major (see below) consisted

of 64 electrodes arranged in 13 rows by 5 columns, with one missing channel in the top left

corner. As for the upper trapezius and the latissimus dorsi, the arrays consisted of 96 electrodes

arranged in 16 rows by 6 columns. Prior to array and reference electrode placement, the area

was shaved, mildly abraded (EVERI, Spes Medica, Genova, Italy), cleaned with distilled water,

and dried with paper towel. The electrode-skin contact was ensured by filling the foam cavities

with conductive paste (AC cream, Spes Medica, Genova, Italy). Two reference electrodes were

used; one positioned on the right elbow (electrode strap) at the level of the epicondyles and the

other on the acromion process (snap electrode).

Several arrays and configurations were piloted to achieve the greatest motor unit yield

in the deltoid (see appendix A). For the anterior/lateral deltoid, the electrode was placed

approximately 2 cm distal to the proximal deltoid tendon, starting from the anterior border

horizontally towards the posterior portion perpendicular to the insertion of the lateral fibres to

the humerus (*array 1*, **Figure 3a**). To cover more of the anterior and lateral aspects of the

deltoid, a second array was placed distally and slightly offset (2 array columns) and parallel to

the proximal array (*array 2*, **Figure 3a**). For the posterior deltoid, the electrode array was

positioned in line with the posterior aspect of the deltoid adjacent to the distal deltoid array

(*array 3*, **Figure 3a**). For the clavicular region of the pectoralis major, the electrode array was

placed approximately 2 cm inferior to the clavicle (superior *array* in (Lulic-Kuryllo *et al.*,

2021*a*); *array 4*, **Figure 3b**). For the upper trapezius, the electrode array was positioned to

bisect the 7th and 8th row (*array 5*, **Figure 3c**) along a line drawn between C7 and the lateral

edge of the acromion. This was done because of the use of the 96-electrode array which is

larger (3 extra rows and 1 extra column) than the standard 64 electrode array used for the

deltoid and clavicular pectoralis major above (modified from (Falla & Farina, 2008*b*). For the

latissimus dorsi, a line was drawn approximately 3 cm inferior to and horizontal to the posterior

axillary crease and parallel to the superior aspect of the iliac crest. From these lines, a diagonal

line was drawn to follow the latissimus dorsi muscle fibres, and the array was placed in the

middle of this area, ensuring that the majority of the array covered the bulk of the muscle

(Padovan *et al.*, 2024) (*array 6*, **Figure 3d**).

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Figure 3. Electrode placement, visual feedback and HDsEMG analysis. (a) Electrode
placement for the deltoid. Array 1 was positioned horizontally approximately 2 cm distal from
the insertion of the deltoid tendon. Array 2 was placed distally and slightly offset to cover the
lateral and anterior deltoid. Array 3 was positioned vertically adjacent to the distal deltoid
array. (b) Electrode placement for the clavicular region of the pectoralis major. Array 4 was
positioned horizontally and approximately 2 cm inferior to the clavicle, avoiding the sternum.
(c) Electrode placement of the upper trapezius. Array 5 was positioned to bisect the 7th and 8th
row of the array along the line between C7 and the acromion process. (d) Electrode placement
of the latissimus dorsi. Array 6 was positioned diagonally in an area delimited by two
horizontal lines, drawn approximately 3 cm inferior to the posterior axillary crease and
parallel to the superior aspect of the iliac crest, the array was placed in line with the muscle
fibres. (e) Visual feedback of the force output. Visual feedback was provided from a line, a
square box and a circle representing the profile, the target force and the force output of the
participant, respectively. (f) Schematic representation of the HDsEMG decomposition.
HDsEMGs were visually inspected and, subsequently, decomposed into motor unit innervation
pulse train (IPTs). The 30-s plateau phase was considered for this analysis (light grey
rectangle).

HDsEMG and force outputs were digitized synchronously at a sampling frequency of

2000 Hz using a 16-bit A/D converter (10-500 Hz bandwidth; Novecento+, OT Bioelettronica,

Turin, Italy). HDsEMG signals were recorded in monopolar derivation and amplified (8 V/V)

to maximize signal resolution while avoiding saturation.

2.4 Data analysis

HDsEMG recordings were analysed offline in MATLAB (version 2024a) using

custom-written scripts. As potential applications of the developed novel experimental

framework, analyses were performed at the global activation level (average HDsEMG

amplitude), the spatial distribution of activation level (coefficient of variation of the HDsEMG

amplitude maps) and the motor unit level (discharge patterns) to assess the neural control of

shoulder muscles. For all analyses, the 30-s plateau phase of each contraction was evaluated.

HDsEMG were bandpass filtered between 20 and 900 Hz using a third-order Butterworth filter

and subsequently visually inspected to identify and remove bad channels due to electrode-skin

contact issues or artifacts (mean ± SD: 2 ± 1 channels removed across all actions and shoulder

angles).

2.4.1 HDsEMG root-mean-square amplitude and coefficient of variation

To assess the global activation of the investigated muscles, the root-mean-square

(RMS) amplitude was utilized. Initially, single-differential (bipolar) EMGs were calculated as

the algebraic difference between monopolar signals detected by consecutive rows of electrodes.

Then, the RMS amplitude was calculated for each of the 59 (64-electrodes array) and 80 (96-

electrodes array) single-differential channels over the duration of the trial, generating

topographic maps of activation. The RMS amplitude maps were normalized to the average

RMS value obtained from a 250 ms window, centred around the peak maximum isometric

voluntary contraction, which was identified by a trained researcher (SR). RMS normalization

was repeated for each muscle, action, and shoulder angle. For simplicity, only *array 2* of the

deltoid was analysed. The average normalized RMS amplitude across all channels was then

considered as the global RMS amplitude of the muscle for each action and shoulder angle.

To assess the variability in spatial distribution of the muscle activation, the coefficient

of variation (CoV) of the normalised RMS amplitude maps was calculated (i.e., the standard

deviation of the normalized RMS amplitudes divided by their mean). The CoV was calculated

separately for each muscle, movement, and shoulder angle.

2.4.2 HDsEMG decomposition

HDsEMG signals were completely decomposed into their constituent motor unit spike

trains using a convolutive blind-source separation algorithm (Negro *et al.*, 2016). Briefly, after

extending and whitening the HDsEMG myoelectric signals, a fixed-point algorithm that seeks

sources that maximize a measure of sparsity was applied to identify the motor unit spike trains

(**Figure 3f**). The spikes were separated from the noise using the K-means algorithm and, while

iteratively updating the motor unit separation vectors, the discharge times estimation was

further refined by minimizing the coefficient of variation of the inter-spike intervals. This

decomposition method has been previously validated (Negro *et al.*, 2016) and extensively

applied to assess the activity of single motor units (Negro *et al.*, 2016; Cogliati *et al.*, 2020;

Cabral *et al.*, 2024). After the automatic identification of motor units, any missed or

misidentified motor unit discharges that produced non-physiological discharge rates (< 4 pulses

per second or > 50 pulses per second; Negro *et al.*, 2009) were manually identified and

iteratively edited by an experienced operator (JGI, SR, CC), and the subsequent motor unit

spike trains were re-estimated (Martinez-Valdes *et al.*, 2017; Hassan *et al.*, 2020). This

approach has been shown to be highly reliable across operators (Hug *et al.*, 2021). Only motor

units with a silhouette (SIL) value, which is a quality control measure of the decomposition

accuracy, greater than 0.87 were included in the further analysis (Negro *et al.*, 2016).

2.4.3 Motor unit discharge behaviour

The instantaneous discharge rates of each identified motor unit were determined by

computing the inverse of the inter-spike intervals, and subsequently, the mean discharge rate

was calculated and considered for further analysis.

2.4.4 Motor unit action potential amplitude

To assess the spatial distribution of motor unit action potentials (MUAP), templates

were obtained using the spike-triggered averaging technique (Stein *et al.*, 1972). Specifically,

single-differential EMGs were segmented into 100-ms windows centred on the discharge times

of individual motor units and averaged. The peak-to-peak amplitude of each template was then

calculated across channels, generating topographical MUAP amplitude maps. These maps

were used both to validate the decomposed motor units and to exemplify the type of neural

information that can be extracted with the proposed framework. As for the HDsEMG

normalized RMS amplitude analysis, only *array 2* of the deltoid was analysed.

2.5 Statistical analysis

All statistical analyses were performed in R (version 4.2.2), using the RStudio

environment. Linear mixed-effect models (LMM) were applied for all statistical analyses, as

they account for the non-independence of data points within each participant, which is

particularly relevant for motor unit data. To compare global normalized RMS amplitude, CoV

of RMS amplitude maps and motor unit mean discharge rate, random intercept models were

applied with action (abduction, adduction, flexion and extension) and muscle (deltoid,

clavicular pectoralis major, upper trapezius and latissimus dorsi) as the fixed effects and

participant as the random effect (e.g., *mean discharge rate ~ 1 + action*muscle + (1 |*

participant)). LMMs were implemented using the package *lmerTest* (Kuznetsova *et al.*, 2017)

with the Kenward-Roger method to approximate the degrees of freedom and estimate the *p*-

values. The *emmeans* package was used, when necessary, for multiple comparisons and to

determine estimated marginal means with 95% confidence intervals (Lenth *et al.*, 2019). All

individual data of motor unit discharge times are available at

https://doi.org/10.6084/m9.figshare.30999811.

3.0 RESULTS

3.1 HDsEMG root-mean-square amplitude and coefficient of variation

Figure 4a depicts the topographical maps of the RMS amplitude at the 30° shoulder

angle across muscle and actions for a selected participant. During abduction, the deltoid was

the most active muscle, particularly in the lateral-anterior portion of *array 2* (see **Figure 3a**),

followed by the caudal region of the upper trapezius. As for the clavicular pectoralis major and

latissimus dorsi, during abduction, there was no visual evidence of significant activation.

During adduction, most of the activity was in the latissimus dorsi and covered much of the

array, followed by a small amount of activity in the medial-caudal portion of the clavicular

pectoralis major array. However, there was no evident activity in the deltoid and upper

trapezius during the adduction action. During flexion, the activity was predominantly in the

anterior deltoid, followed by the clavicular pectoralis major. As for extension, the deltoid was

again the most active, with the activity primarily towards the posterior aspect of the array.

There was also significant activity in the caudal region of the array in the latissimus dorsi and

upper trapezius, with no evidence of activity in the clavicular pectoralis major. In **Figure 4b,**

the topographical RMS amplitude maps for the same selected participant are depicted, but at a

shoulder angle of 65°. The activation patterns during abduction, adduction, and flexion closely

resembled those reported above for the 30° condition. However, during extension, the deltoid

was significantly active across *array 2*. Additionally, as seen for the 30° condition, a significant

amount of activity was observed in the caudal region of the upper trapezius, but with little

activity in the latissimus dorsi.

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380   ***Figure 4. Muscle activation patterns across shoulder actions at 30° and 65°.*** *RMS amplitude*
381   *topographical maps for the deltoid, clavicular pectoralis major, upper trapezius, and*

latissimus dorsi during abduction, adduction, flexion and extension. These actions were performed with the shoulder positioned at 30° (a) and 65° (b) of lateral abduction. The x- and y-axes represent the rows and columns of the array, with colour intensity (from dark blue to dark red) indicating the level of muscle excitation. Panels (c) and (d) show the group results of the global normalized RMS amplitude (%MVC) for each action and muscle. Panels (e) and (f) show the group result of the coefficient of variation (CoV) of the RMS amplitude maps. Horizontal traces, boxes, and whiskers represent the median value, interquartile interval, and distribution range, respectively.

Figures 4c and **4d** show the group results for the global normalized RMS amplitude at 30° and 65° shoulder angles. For both 30° and 65° there were statistically significant interactions between muscle and action (30°: LMM, $F=20.030$, $p<0.001$; 65°: LMM, $F=28.213$, $p<0.001$). The following pairwise comparisons were common for both angles. For abduction, the deltoid and the upper trapezius had a significantly greater global normalized RMS amplitude compared to clavicular pectoralis major and latissimus dorsi ($p<0.001$ for all comparisons). During flexion, the normalized RMS amplitude of the clavicular pectoralis major was significantly greater compared to upper trapezius and latissimus dorsi ($p<0.001$ for all comparisons). Additionally, at the 30° shoulder angle, during flexion the normalized RMS amplitude of the clavicular pectoralis was significantly greater than the deltoid ($p<0.001$). During extension, both the deltoid ($p=0.022$) and the upper trapezius ($p<0.001$) had a greater normalized RMS amplitude compared to the clavicular pectoralis major, with no significant differences compared to the latissimus. When comparing the same muscle across actions at the 30° and 65° shoulder angle, the deltoid normalized RMS amplitude was significantly greater in abduction, flexion, and extension compared to adduction ($p<0.01$ for all comparisons), as well as during abduction compared to flexion and extension ($p<0.01$ for both comparisons). During flexion the clavicular pectoralis major exhibited the greatest normalized RMS amplitude compared to all other actions ($p<0.001$ for all comparisons). As for the upper trapezius, the normalized RMS amplitude was significantly greater during abduction compared to adduction, flexion, and extension ($p<0.001$ for all comparisons). Finally, the latissimus dorsi was not significantly different across all actions ($p>0.05$ for all comparisons).

The group results for the CoV of the normalized RMS amplitude maps at the 30° and

65° shoulder angle showed a statistically significant interaction between muscle and action

(**Figure 4e and 4f**, 30°: LMM, $F=2.790$, $p=0.007$; 65°: LMM, $F=2.789$, $p<0.001$). The

following pairwise comparisons were common for both angles. For abduction, adduction, and

flexion there were no significant differences observed across muscle ($p>0.05$). During

extension, the CoV of the normalized RMS amplitude maps of the upper trapezius was

significantly greater compared to all other muscles ($p<0.05$ for all comparisons). While

comparing the same muscle across movement, at the 30° shoulder angle, the upper trapezius

had a significantly greater CoV of the normalized RMS amplitude maps during extension

compared to all other movements ($p<0.05$ for all comparisons). While, at the 65° shoulder

angle, the upper trapezius CoV of the normalized RMS amplitude maps was significantly

higher during extension compared to flexion ($p=0.030$).

3.2 Motor unit discharge behaviour

Figures 5 through **8** illustrate, for the different shoulder actions at the 30° shoulder

angle, the neural features at the motor unit level that can be extracted using the proposed novel

framework. For each muscle, representative MUAP amplitude maps and the corresponding

motor unit instantaneous discharge rate are presented. During abduction (**Figure 5**), motor

units could be identified only in the deltoid and upper trapezius for this representative

participant, with 30 and 19 units decomposed, respectively. For the deltoid, the motor units

were distributed across different regions of *array 2*, including anterior (MUAP 1 and MUAP

30), lateral (MUAP 3 and MUAP 5), and posterior regions (MUAP 2 and MUAP 4). In contrast,

most of the motor units identified in the upper trapezius were located cranially within the array.

Additionally, as seen in the right panel of **Figure 5**, most of the motor units identified from

both muscles were tonically active throughout the plateau phase of the action.

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Figure 5: Motor unit action potential amplitude maps and motor unit discharge rate during
shoulder abduction at 30°. *Motor unit action potentials were obtained using the spike-*
triggered averaging technique from array 2. For this representative subject, motor units were
identified in the deltoid (30 units) and upper trapezius (19 units). No motor units were
decomposed from the clavicular pectoralis major or latissimus dorsi (left panel). Abduction
force profile (grey line), and motor unit instantaneous discharge rate are shown in the right
panel. Separately per muscle, each motor unit is represented with a colour, used both for the
MUAP distribution maps and the instantaneous discharge rate.

Figure 6 is the same representative participant during the adduction action at a 30°

shoulder angle. In this condition, motor units were identified in the deltoid (5 units), clavicular

pectoralis major (16 units), upper trapezius (2 units), and the latissimus dorsi (13 units). The

identified motor units in the deltoid were located across all regions of the deltoid. For the upper

trapezius the identified units were located caudally and, in the latissimus, dorsi, as seen in the

deltoid, the units were spread across the entire region. Interestingly, the units identified in the

clavicular pectoralis major covered the entire *array*, however the majority of the activity

following the clavicular pectoralis major line of action. While most of the clavicular pectoralis

major and latissimus dorsi motor units were tonically active throughout the plateau phase, some

deltoid and two motor units identified in the upper trapezius exhibited more phasic activity.

Figure 6: Motor unit action potential amplitude maps and motor unit discharge rate during shoulder adduction at 30°. *Motor unit action potentials were obtained using the spike-triggered averaging technique from array 2. For this representative subject, motor units were identified in the deltoid (5 units), clavicular pectoralis major (16 units), upper trapezius (2 units) and latissimus dorsi (13 units) (left panel). Adduction force profile (grey line), and motor unit instantaneous discharge rate are shown in the right panel. Separately per muscle, each motor unit is represented with a colour, used both for the MUAP distribution maps and the instantaneous discharge rate.*

Figure 7 shows the same representative participant during the flexion action at a 30° shoulder angle. In this condition, motor units were identified in the deltoid (20 units), clavicular pectoralis major (21 units), and upper trapezius (16 units), whereas again no motor units were identified in the latissimus dorsi. The identified motor units in the deltoid were predominantly located in the anterior region of the array (e.g., MUAP 1, MUAP 2, and MUAP 5). For both the upper trapezius and clavicular pectoralis major, motor units were distributed across different regions of their respective arrays. While most deltoid and clavicular pectoralis major motor units were tonically active throughout the plateau phase, some upper trapezius units exhibited more phasic activity.

Figure 7: Motor unit action potential amplitude maps and motor unit discharge rate during shoulder flexion at 30°. Motor unit action potentials were obtained using the spike-triggered averaging technique from array 2. For this representative subject (same as in Figure 5), motor units were decomposed from the deltoid (20 units), clavicular pectoralis major (21 units), and upper trapezius (16 units). No motor units were decomposed from the latissimus dorsi (left panel). The flexion force profile (grey line), and motor unit instantaneous discharge rate are shown in the right panel. Separately per muscle, each motor unit is represented with a colour, used both for the MUAP distribution maps and the instantaneous discharge rate.

Figure 8 illustrates the same representative participant during the extension action at a 30° shoulder angle. In this condition, motor units were identified in the deltoid (9 units), upper trapezius (10 units), and latissimus dorsi (12 units), whereas no motor units were identified in the clavicular pectoralis major. The identified motor units in the deltoid were predominantly located in the posterior region of the array (e.g., MUAP 2, MUAP 3, and MUAP 9). For the upper trapezius, motor unit 1 was dispersed across the entire array, where the other motor units were located more caudally. In the latissimus dorsi, the motor units were distributed across different regions of the array with the majority located more cranially (e.g., MUAP 2, MUAP 4, MUAP 5, and MUAP 12). While most of the deltoid and upper trapezius motor units were tonically active throughout the plateau phase, the latissimus dorsi units exhibited both tonic and phasic activity.

Figure 8: Motor unit action potential amplitude maps and motor unit discharge rate during shoulder extension at 30°. Motor unit action potentials were obtained using the spike-triggered averaging technique from array 2. For this representative subject, motor units were identified in the deltoid (9 units), upper trapezius (10 units) and latissimus dorsi (12 units). No motor units were decomposed from the clavicular pectoralis major (left panel). Extension force profile (grey line), and motor unit instantaneous discharge rate are shown in the right panel. Separately per muscle, each motor unit is represented with a colour, used both for the MUAP distribution maps and the instantaneous discharge rate.

Table 1 summarizes the motor unit yield across all muscles, actions, and shoulder angles. Overall, a greater number of motor units were decomposed at the 30° shoulder angle compared with 65°. Consistent with the representative case, no motor units were identified in the clavicular pectoralis major and latissimus dorsi during abduction, or in the latissimus dorsi during flexion. In addition, no units were decomposed from the clavicular pectoralis major during extension.

Table 1: Average ± standard deviation motor unit yield across all muscles (deltoid, clavicular pectoralis major, upper trapezius and latissimus), actions (abduction, adduction, flexion and extension) and shoulder angles (30° or 65°).

	SHOULDER AT 30 DEGREES				SHOULDER AT 65 DEGREES			
	Abduction	**Adduction**	**Flexion**	**Extension**	**Abduction**	**Adduction**	**Flexion**	**Extension**

Deltoid	12 ± 19	9 ± 15	10 ± 11	12 ± 7	7 ± 7	2 ± 3	10 ± 9	10 ± 11
Pectoralis	0 ± 0	5 ± 3	5 ± 7	0 ± 0	0 ± 0	6 ± 6	5 ± 6	0 ± 0
Trapezius	6 ± 6	5 ± 9	5 ± 5	5 ± 4	5 ± 3	3 ± 1	5 ± 2	4 ± 2
Latissimus	0 ± 0	5 ± 5	0 ± 0	3 ± 3	0 ± 0	3 ± 4	0 ± 0	2 ± 2

Figure 9 illustrates the group results for the mean motor unit discharge rate for the 30°

(**Figure 9a**) and 65° (**Figure 9b**) shoulder angles. For both 30° and 65° there was a statistically

significant interaction between muscle and action (LMM, F=3.190, p=0.008; LMM, F=6.821,

p<0.001). The following pairwise comparison were common for both angles. For abduction,

there were no significant differences between the mean motor unit discharge rate of the deltoid

and upper trapezius (p>0.05). During flexion, at the 30° shoulder angle, the motor units of the

clavicular pectoralis major had significantly higher mean discharge rate compared to the motor

units of both upper trapezius and deltoid (p<0.05 for both comparisons). However, during

flexion, at the 65° shoulder angle, both the deltoid and clavicular pectoralis major had

significantly greater motor unit discharge rates compared to the upper trapezius (p<0.001 for

both comparisons). For extension, both the deltoid and upper trapezius had significantly higher

motor unit discharge rate compared to the latissimus dorsi (p<0.05 for both comparisons).

When comparing the same muscle across actions at the 30° and 65° shoulder angles, the deltoid

had a greater mean motor unit discharge rate during abduction, flexion and extension actions

compared to adduction (p<0.001 for all comparisons). The mean motor unit discharge rate of

the clavicular pectoralis major was higher during flexion than adduction (p<0.001), however,

the upper trapezius mean motor unit discharge rate during abduction was greater compared to

adduction and flexion (p<0.05 for both comparisons).

Figure 9: Motor unit discharge behaviour across actions and shoulder angles. Group results for the mean motor unit discharge rate (a and b) during abduction, adduction, flexion, and extension at both 30° (top) and 65° (bottom) shoulder angles. Horizontal traces, boxes, and whiskers represent the median value, interquartile interval, and distribution range, respectively; individual circles correspond to single motor units.

4.0 DISCUSSION

The aim of this methodological paper was to propose a novel framework based on the investigation of four isometric shoulder actions at two angles to fully assess the neural control of the superficial shoulder muscles (deltoid, clavicular pectoralis major, upper trapezius, and latissimus dorsi). This framework integrates information across multiple levels of analysis, including global HDsEMG amplitude (normalized RMS), spatial distribution of activation (CoV of RMS amplitude maps), and motor unit discharge behaviour (discharge statistics). To our knowledge, this is the first paper to report motor unit behaviour obtained with non-invasive

HDsEMG from the deltoid and latissimus dorsi, and to concurrently assess motor unit activity

across all four superficial shoulder muscles during controlled isometric actions. By combining

a six-degree-of-freedom load cell, a robotic arm, and evolving HDsEMG technology with

motor unit decomposition, the proposed framework demonstrates the feasibility of assessing

neural control of the shoulder musculature beyond traditional bipolar EMG approaches.

Importantly, this framework provides several examples of the type of extractable neural

information that can be obtained when global, spatial, and motor unit analyses are integrated,

thereby offering methodological guidelines for future investigations and contributing to the

advancement of shoulder neuromechanical and musculoskeletal modelling.

554 *4.1 Differences across muscles within the same action*

The application of the proposed framework can be integrated into assessments from

many disciplines, including but not limited to, sport, physiotherapy and assessment of clinical

populations for exercise prescription and rehabilitation. The application of the framework in

clinical population, such as stroke and breast cancer survivors, has the ability to assist in

monitoring the progression of treatment or the potential negative effects of treatment

(Leonardis *et al.*, 2022). The proposed framework can be used in addition to assessments with

standard bipolar recordings, which has been shown to mischaracterize pectoralis major activity

in commonly performed tasks (Lulic-Kuryllo *et al.*, 2021*a*, 2023). Therefore, the additional of

HDsEMG recordings can provide critical information during assessments.

General consensus is that during complex shoulder actions, most of the muscles will

stabilize while others will produce the necessary muscle forces to initiate action (Schenkman

& Rugo de Cartaya, 1987; Kelly *et al.*, 2002). During shoulder abduction, both the deltoid and

upper trapezius exhibited significantly greater global RMS amplitudes compared to the

clavicular pectoralis major and latissimus dorsi, consistently across shoulder angles (**Figures**

4c, 4d). This pattern aligns with the primary mechanical roles of the deltoid in humeral elevation and the upper trapezius in scapular stabilization during force generation (Paine & Voight, 1993; Trudeau *et al.*, 2022). From a global EMG perspective, these two muscles would therefore be interpreted as the dominant contributors to the abduction action. However, the additional spatial and motor unit analyses revealed important nuances not captured by global amplitude alone. Motor units identified in the upper trapezius were predominantly localized in the caudal portion of the array (**Figures 5 through 8**), indicating a more spatially confined activation strategy (Falla & Farina, 2008*b*). In contrast, the deltoid showed region-specific activation, with motor units distributed across anterior, lateral, and posterior regions of the array, reflecting the engagement of multiple functional compartments within the muscle (**Figures 5 through 8**). Despite these distinct spatial recruitment patterns, the mean motor unit discharge rates did not differ between the deltoid and upper trapezius (**Figures 5 through 8**). This combination of findings suggests that, during abduction, differences between these muscles might not be primarily driven by the magnitude of neural drive, but rather by how that drive is spatially distributed within each muscle. These observations illustrate how the proposed framework allows differentiation of muscle-specific control strategies within a single functional action

During shoulder extension, the deltoid and upper trapezius again exhibited the greatest global EMG amplitude, but this was accompanied by a marked increase in spatial heterogeneity of activation in the upper trapezius, as reflected by a higher coefficient of variation of the RMS amplitude maps (**Figure 4**). This region-specific activation of upper trapezius is in line with previous studies showing higher caudal activation during shoulder actions (Falla & Farina, 2008*a*, 2008*b*). While global measures alone would suggest a secondary role of the upper trapezius in this action, the spatial analysis indicates a more complex activation pattern, potentially reflecting action-dependent stabilization or synergistic control demands (Paine &

Voight, 1993; Trudeau *et al.*, 2022). At the motor unit level, both the deltoid and upper

trapezius displayed higher discharge rates than the latissimus dorsi, despite no differences in

normalized global RMS amplitude between them (**Figures 4c, 4d, 9**). This dichotomy between

amplitude and discharge behaviour highlights the added value of motor unit analysis for

interpreting muscle contributions during multi-joint shoulder actions.

Collectively, these findings highlight the limitation of relying solely on RMS amplitude

from bipolar EMG, which, due to waveform cancellation or cross-talk, may not fully capture

underlying neural activity (De Luca & Merletti, 1988; Day & Hulliger, 2001; Dimitrova *et al.*,

2002; Farina *et al.*, 2002, 2010; Keenan *et al.*, 2005). Therefore, the use of global EMG

measures allows for a general understanding of the duration and magnitude a muscle of interest

is activated, but this cannot necessarily indicate the functional role the muscle is contributing

to a specific muscle action (Boettcher *et al.*, 2010). Therefore, the knowledge of motor unit

behaviour during a shoulder action and the region of greatest activation may be essential to

provide the information necessary to understand the functional roles of the involved muscles.

4.2 Differences within the same muscle across actions

The proposed framework allows the assessment of not only the muscles activity across

actions, which we suggest is highly relevant for clinical populations, but also the assessment

of the differing activation of individual muscles across actions. This type of assessment is

highly relevant for basic research questions regarding the neural control of shoulder actions.

To date there is a significant knowledge gap in the neural control of the shoulder, specifically

concerning the deltoid and latissimus dorsi (Inglis *et al.*, 2025). As was highlighted in the

systematic review and meta-analysis by Inglis, Cabral and colleagues (2025), there is much

work still to be done to understand the full complexities of the shoulder. Therefore, this

proposed framework would be utilized to investigate these complexities, specifically in the

deltoid and latissimus dorsi which both have a paucity of research, specifically involving motor

unit behaviour non-invasively. The precision of the proposed framework would also allow the

tracking of minor activation changes across shoulder angles. These activation adjustments may

result from trivial changes in muscle architecture (e.g. pennation angle, muscle length)

(Peterson & Rayan, 2011) and moment arm length (Otis *et al.*, 1994; Ackland *et al.*, 2008) that

have significant implications neuromechanically.

When examining the same muscle across different actions, clear action-dependent

modulation emerged at both the global and motor unit levels. The deltoid demonstrated

significant action-dependent modulation across the investigated shoulder actions (**Figures 4**

through 9). Global RMS amplitude and mean motor unit discharge rate were significantly

greater during abduction, flexion, and extension compared to adduction, confirming its

multifunctional role as a primary mover across several directions (Wickham & Brown, 1998).

Beyond changes in activation magnitude, MUAP distribution maps for a representative

participant revealed a redistribution of activity across the deltoid surface (**Figures 7, 8**), with

anterior regions predominantly active during flexion and posterior regions during extension.

These findings suggest that neural control of the deltoid involves not only scaling of neural

drive but also selective recruitment of different motor unit populations, supporting the notion

of functional compartmentalization within large shoulder muscles.

The upper trapezius also exhibited marked action-dependent modulation. Global RMS

amplitude was greatest during abduction, consistent with its established role in scapular control

(Paine & Voight, 1993; Trudeau *et al.*, 2022). However, the spatial distribution of activation

varied markedly across actions, with extension eliciting a significantly higher coefficient of

variation compared to other actions (**Figures 4e, 4f**). At the motor unit level, discharge rates

were highest during abduction and lower during flexion and adduction, suggesting an action-

dependent adjustment of the neural drive. Together, these results demonstrate that spatial

distribution and motor unit variables provide complementary insights into the neural strategies

underlying the upper trapezius activation that are not evident from global amplitude measures

alone.

For the clavicular pectoralis major, greater global RMS amplitude and motor unit

discharge rates were primarily observed during flexion, whereas little activity was detected

during extension (**Figures 4a, 4b**). Similarly, motor units from the latissimus dorsi were

identified only during adduction and extension (**Table 1**). Together, these findings demonstrate

that muscles contributing to multiple shoulder actions are modulated not only through

adjustments in activation level but also through action-specific recruitment and rate coding

strategies.

Although motor units were not tracked across actions, the differences in sets of active

units between actions strongly suggest action- and angle-dependent recruitment and de-

recruitment, consistent with previous findings in other upper-limb muscles (ter Haar Romeny

et al., 1984). These observations align with previous EMG amplitude evidence showing

localized activation of deltoid muscle regions dependent on action direction (Wickham &

Brown, 1998, 2012; Brown *et al.*, 2007), as well as similar findings in the rotator cuff muscles

(Alenabi *et al.*, 2018; Lulic-Kuryllo *et al.*, 2020). Importantly, the present framework extends

these observations by providing direct evidence at the motor unit level.

4.3 Methodological implications and future applications

The capability of the proposed framework to map both spatial activation patterns

(**Figures 4a, 4b**) and motor unit territories and discharge patterns (**Figures 5** through **8**)

provides a powerful tool for probing the neural strategies underlying complex shoulder control.

The identification of region-specific motor unit recruitment and tonic versus phasic discharge

statistics offers insight into how muscles contribute to both force generation and stabilization

during different actions. This is evident during the extension action where the posterior deltoid

and upper trapezius are tonically active as they are involved in the action and the latissimus

dorsi is both tonically and phasically active as some motor units are recruited to support the

action and others (MUAPs 2-4) likely act as stabilizers (**Figure 6**). This multilevel information

is particularly valuable for large, multifunctional muscles such as the deltoid and upper

trapezius, whose roles cannot be fully understood using global EMG amplitude alone.

Current shoulder musculoskeletal models are based on simplified mechanical analysis

and often treat muscles as single force-generating elements (Hughes & An, 1997). The present

findings suggest that future modelling studies of the deltoid should consider within-muscle

heterogeneity, including compartmentalized segments (compartmentalized within segments:

anterior, lateral, posterior) and how this may change based on muscle architecture, moment

arm length and action (Hughes & An, 1997). Incorporating motor unit-level information into

such models may substantially improve their physiological realism and clinical relevance.

Finally, the differences observed between normalized RMS amplitude and motor unit

discharge behaviour at the group level further highlights the limitations of classic EMG-based

approaches. Discrepancies between global amplitude and discharge rate plausibly reflect

factors such as amplitude cancellation, the limited spatial coverage of HDsEMG arrays, and

potential cross-talk from adjacent muscles (De Luca & Merletti, 1988; Day & Hulliger, 2001;

Dimitrova *et al.*, 2002; Farina *et al.*, 2002; Keenan *et al.*, 2005). By directly assessing motor

neuron output, the proposed framework provides a more precise assessment of neural drive and

regional muscle involvement. This methodological advance has important implications for

both basic research and clinical applications, particularly in populations where subtle

alterations in neural control may underlie functional impairment and where targeted

rehabilitation strategies are required.

5.0 PRACTICAL IMPLICATIONS

• Using the proposed framework, rehabilitation programs can be designed based on the

specific actions that isolate the motor unit activity, not the global muscle activity, to

target specific impairments better

• The proposed framework provides a roadmap for stroke rehabilitation, where the focus

may need to be centred more on motor unit recruitment and rate coding rather than on

general activity within a portion of the muscle.

• The proposed framework provides a detailed description of the equipment used during

the data collection including the testing jig, recording equipment, array locations and

actions at each joint angle to be reproduceable for scientists and clinicians investigating

recruitment of the shoulder musculature.

• The use of the robotic arm in the proposed framework allows for the testing jig and load

cell to be situated so the position of the humerus, glenohumeral joint angle, muscle and

moment arm length do not change across actions, only the jig moves to allow the

recording of the forces produced during the specified action direction.

6.0 LIMITATIONS

Despite the rigor taken to control the specific shoulder actions and position, several

limitations should be acknowledged. First, although participants were secured with straps and

a rigid jig, the inherent flexibility of the shoulder complex may have introduced small

variations in action execution. While this may reduce experimental control, it also increases

ecological validity by better replicating strategies used in daily activities. Second, only two

joint angles within the mid-range were tested, limiting extrapolation to positions closer to $0°$

or $180°$. Third, contractions were restricted to 30% MVC, chosen to reflect moderate force

demands of activities of daily living, but additional work is needed across lower and higher

force levels. Fourth, the sample included only one female participant, precluding sex

comparisons. Finally, only four major superficial shoulder muscles were recorded. The

inability to assess, particularly the scapular stabilizers and rotator cuff, is a key omission given

their likely contribution during extension and adduction actions.

7.0 CONCLUSIONS

The present study demonstrates that it is feasible to record and analyse motor unit

behaviour of major superficial shoulder muscles using HDsEMG during controlled isometric

actions. The framework combines motor unit analyses with precise force measurements to

provide complementary insights into neural control strategies. These methodological advances

are essential for the development of comprehensive biomechanical models of the shoulder that

incorporate not only anatomical and mechanical properties but also neural control strategies.

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