



# Use of muscle synergies and wavelet transforms to identify fatigue during squatting



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## ABSTRACT

The objective of this study was to supplement continuous wavelet transforms with muscle synergies in a fatigue analysis to better describe the combination of decreased firing frequency and altered activation profiles during dynamic muscle contractions. Nine healthy young individuals completed the dynamic tasks before and after they squatted with a standard Olympic bar until complete exhaustion. Electromyography (EMG) profiles were analyzed with a novel concatenated non-negative matrix factorization method that decomposed EMG signals into muscle synergies. Muscle synergy analysis provides the activation pattern of the muscles while continuous wavelet transforms output the temporal frequency content of the EMG signals. Synergy analysis revealed subtle changes in two-legged squatting after fatigue while differences in one-legged squatting were more pronounced and included the shift from a general co-activation of muscles in the pre-fatigue state to a knee extensor dominant weighting post-fatigue. Continuous wavelet transforms showed major frequency content decreases in two-legged squatting after fatigue while very few frequency changes occurred in one-legged squatting. It was observed that the combination of methods is an effective way of describing muscle fatigue and that muscle activation patterns play a very important role in maintaining the overall joint kinetics after fatigue.

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## 1. Introduction

Humans have the ability to perform dynamic movements with little to no conscious effort; however due to the high dimensionality of the neuromuscular system, the underlying mechanisms to perform such tasks are highly complex. Electromyography (EMG) is a commonly used tool for recording and analyzing the role of skeletal muscle during movement. Due to the large number of muscles used in various tasks, it can be difficult to decipher individual muscle activations and their contributions in the larger context of movements such as squatting. This drawback has been recently addressed using non-negative matrix factorization (NNMF) to perform muscle synergy analyses, decomposing large quantities of EMG data into a few, simpler components that can be used to describe muscle roles in a certain task (Tresch et al., 2006).

An increasing amount of studies have used muscle synergies to describe human gait and it has been seen that level walking can be

described by four (Barroso et al., 2014), five (Ivanenko et al., 2004) or six (Allen and Neptune, 2012) synergies. In addition to human gait, muscle synergies have also been used to analyze natural motor behaviours in frogs such as kicking (d'Avella et al., 2003), jumping, swimming and walking (d'Avella and Bizzi, 2005). Here it was observed that three shared, plus a varying number of task-specific synergies, were necessary to decompose the EMG signals of each task indicating that the central nervous system (CNS) may build off of a general template, adding activations for fine-tuning as necessary for specific tasks.

With respect to human exercises, Kristiansen et al. (2015) examined muscle synergies used in bench pressing for trained and untrained individuals. They observed that two synergies were sufficient in describing the movement and that more than 95% of the variance was accounted for (VAF). These two synergies corresponded to movement phases as one characterized the concentric phase, while the other represented the eccentric phase. Synergies have also been used to analyze fatigue during a cyclic, rowing task (Turpin et al., 2011). They observed that three synergies achieved more than 90% VAF in EMG profiles before and after the participants rowed until exhaustion. The authors noted only minor changes in synergy activations, which led the authors to postulate

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that fatigue-induced differences are predominantly seen in muscle firing frequencies rather than changes in the organization of muscle coordination. Unfortunately, these authors neglected to include a frequency analysis and thus, their conclusions are unsupported.

Spectral changes to the EMG signal due to fatigue can be attributed to firing frequency but can also be caused by other central mechanisms such as motor unit synchronization (Hermens et al., 1992) and peripheral mechanisms such as a change in motor neuron conduction velocity (Arendt-Nielsen and Mills, 1985) or a variety of other intramuscular responses (Kirkendall, 1990). Traditional amplitude and frequency analyses have been used to effectively detect these central and peripheral changes to evaluate fatigue in isometric conditions (Viitasalo and Komi, 1977). These methods cannot be effectively applied to dynamic conditions, however, as (1) the EMG signal is not stationary, and (2) there is a relative shift of the electrodes in relation to the origin of action potentials, and the conductivity properties of the soft tissue separating the electrodes from the muscle fibres (Farina, 2006). These limitations of using surface EMG in dynamic contractions can be addressed by conducting short-time Fourier transforms (STFT) in short time epochs to account for the non-stationarity. When using an STFT, the dynamic signal is windowed to small enough periods of time where the contraction can be assumed to be stationary as the traditional Fourier transform has no resolution in the time domain. In certain dynamic tasks with large joint ranges of motions such as squatting, muscle fibres undergo considerable movement in relation to the surface electrodes. Therefore, windowing may not be the most effective technique in analyzing the non-stationary signal as the epoch would have to be quite small in order to negate muscle fibre movement and assume stationarity. Since continuous wavelet transforms (CWT) scale and translate a mother wavelet, they are not limited to the assumption of stationarity and thus, are an attractive alternative. They have been effectively applied to dynamic contractions (Karlsson et al., 2001; Hostens et al., 2004; Cifrek et al., 2009) and have shown better accuracy and precision than other time–frequency methods including STFT (Karlsson et al., 2000).

Even though traditional amplitude and frequency analyses have been effective in quantifying the fatigue-related increases in activation amplitude and decreases in firing frequency during isometric tasks (Viitasalo and Komi, 1977), these techniques are, as noted above, not suitable for dynamic movements (Farina, 2006). Therefore, the primary aim of this study is to supplement CWT with muscle synergy extraction in a fatigue analysis to better describe the combination of decreased firing frequency and altered activation profiles during dynamic muscle contractions. It is hypothesized that similar to isometric contractions, fatigue-related changes during dynamic contractions will be in the form of decreases in muscle firing frequency and altered muscle activation patterns, which will be effectively elucidated through the combined use of CWT and muscle synergies.

## 2. Methods

### 2.1. Participants

Nine healthy young adults ( $23.7 \pm 1.6$  years; 5 male) participated in this study. Exclusion criteria were previous reports of significant lower limb injuries within six months of participation, or any other physical impairment that may influence knee function. All participants read and signed an informed consent form prior to data collection. The study was approved by the university's ethics review board.

### 2.2. Electromyography

Skin preparation included shaving and wiping the muscle belly down with an alcohol swab. Twelve bipolar surface EMG electrodes (electrode dimensions:  $10.0 \times 1.0$  mm; inter-electrode distance: 10 mm; SP-E04, DE 2.1 DelSys Inc., USA) connected to a 16-channel EMG system (DS-B04, Bagnoli-16, DelSys Inc., USA) were placed over the muscle bellies of the gluteus maximus (GLTMAX), gluteus medius (GLTMED), tensor fascia latae (TFL), rectus femoris (RFEM), vastus medius (VMED), vastus lateralis (VLAT), long head of biceps femoris (BFEM), semitendinosus (STEN), tibialis anterior (TA), medial gastrocnemius (MGAS), lateral gastrocnemius (LGAS), and the soleus (SOL). EMG was recorded from the dominant limb, which was determined by which leg would be used to kick a soccer ball a maximal distance. Electrode placements followed the guidelines outlined by SENIAM (Hermens et al., 2000).

MVIC data were collected using an isokinetic dynamometer (850-000, Biodex, USA). Plantar and dorsiflexion maximal efforts were collected with the ankle at  $100^\circ$ , knee flexion and extension maximal efforts were collected with the knee at  $45^\circ$  and hip flexion, extension and adduction were collected with the participant standing with a straight leg. Three MVICs for each seven second ramped contraction were completed with a 45 s rest period between each exertion. In an effort to optimize the myoelectric signal power to low frequency noise ratio for our application (Stegeman and Hermens, 1998; van Boxtel, 2001; Clancy et al., 2002; De Luca et al., 2010), all EMG signals were sampled at 1000 Hz, amplified by a gain of 1000, and band-pass filtered at 20–450 Hz.

### 2.3. Movement trials and fatigue protocol

Motion data were collected using a 15-camera infrared motion capture system (MX-40 cameras; Vicon Nexus, v1.7, UK) at 200 Hz with a Helen Hayes full-body marker set. Three dimensional ground reaction forces were collected using two force platforms (FP4060-08, Bertec, USA).

Participants first completed three successful one-legged squats on the dominant limb. During the one-legged squats, participants were only asked to squat at their own pace, keep their non-dominant leg behind and squat down as low as possible.

To fatigue the participants, a bench was adjusted to the participant's tibia height and the participant began two-legged squatting at a metronome-controlled pace of 35 squats per minute while holding a standard 20.4 kg Olympic bar on their shoulders. Participants continued to squat until complete exhaustion, which corresponded to a maximal value on the Borg Scale of Perceived Exertion (Borg, 1982). The average time to exhaustion was  $8.2 \pm 3.9$  min. Three of the first five recorded squats were used for the non-fatigued state, while three of the last five were used for fatigued state. Once exhausted, participants completed three more successful one-legged squats with all collections occurring within minutes of the fatigue to avoid recovery as much as possible. The selection of successful squats was based on proper balance and form, foot contact occurring completely on the force plates and markers being recognized by the motion capture system.

### 2.4. Data analysis

Trajectories and ground reaction forces were analyzed in Visual 3D (v.4, C-Motion, USA) and were smoothed with a 4th order zero-lag low-pass Butterworth filter with a 10 Hz cut-off. Lower limb joint sagittal plane kinetics and kinematics were computed in Visual 3D with the Helen Hayes model (Kadaba et al., 1990). EMG signals were analyzed in Matlab (2014a, MathWorks, USA) and processed with a 4th order zero-lag high-pass Butterworth

filter with a 30 Hz cut-off based on visual inspection to successfully remove motion artifact and other low frequency noise, then rectified and filtered with a 4th order zero-lag 5 Hz low-pass Butterworth filter. EMG data were then normalized to MVIC and time normalized to the beginning of knee flexion to the end of knee extension. This time normalization was to 101 data points (Matlab *interp1* function).

The high-pass filtered EMG data were subjected to a Matlab Morlet *cwt* function with scales from one to 40 (central frequencies: 20.31–812.5 Hz), which was sufficient in encompassing the entire EMG signal. A scalogram was then used to calculate the percent power of each coefficient over the entire movement. Instantaneous mean frequencies (IMNF) were calculated through Eq. (1) (Karlsson et al., 2001):

$$IMNF(t) = \frac{\int_0^F \omega P(t, \omega) d\omega}{\int_0^F P(t, \omega) d\omega} \quad (1)$$

where  $F$  is the Nyquist Frequency and  $P(t, \omega)$  is the time-dependent power spectral density. IMNF were then averaged over the entire trial to determine mean frequencies (MnF).

Concatenated non-negative matrix factorization (CNMF) concatenates the original EMG data of individual trials (Oliveira et al., 2014) or all trials of all participants (Shourijeh et al., 2016) as in the present study, and while keeping the synergy matrix fixed among those participants/trials. By keeping the synergy fixed among participants, signal variability between the participants is limited to the coefficients and therefore is a more robust approach. Concatenation also circumvents the subjective and unreliable sorting step of classic NMF (Shourijeh et al., 2016). A solver builds the two matrices of coefficients ( $C$ ) and synergies ( $S$ ) so that the multiplication approximates the original data  $A \approx CS$ . The objective function, which is set to be minimized by Matlab's *nnmf* and *als* functions is:

$$J = \sum_{i=1}^N \frac{\|A_i - C_i S\|_F}{\|A_i\|_F} \quad (2)$$

where  $N$  is the total number of trials in the study;  $A$  and  $C$  are the concatenated matrix of EMG data and coefficients, respectively;  $S$  is synergy matrix fixed between trials; and  $\|A_i\|_F$  designates the Frobenius norm of EMG data of the  $i$ th trial. For this application,  $A$ ,  $C$ , and  $S$  will have dimensions of  $2700 \times 12$ ,  $2700 \times N_s$ , and  $N_s \times 12$  ( $N_s$  = number of synergies to be determined; number of data points = 100; number of trials = 27; number of muscles = 12). A value of  $1e-6$  was chosen for function tolerance and search tolerance for the *als*, and the factorization replicates were set to 30 (Shourijeh et al., 2016). Convergence of the optimization is checked by total VAF versus the number of synergies applied. The VAF is defined according to the definition by Torres-Oviedo et al. (2006):

$$VAF = \left( 1 - \frac{\sum_{k=1}^{2700} \sum_{j=1}^{12} \epsilon_{kj}^2}{\sum_{k=1}^{2700} \sum_{j=1}^{12} A_{kj}^2} \right) * 100 \quad (3)$$

where  $\epsilon$  is the error, i.e.,  $A - CS$ , and the indices  $k$  and  $j$  designate the rows and the columns of the quantities  $\epsilon$  and  $A$ .

Required number of synergies was based on (1) providing a  $VAF \geq 90\%$  and (2) adding another synergy will increase the VAF by  $\leq 5\%$ . Since pre and post-fatigue analyses are run separately, the solver may produce similar synergies between fatigue states but in a varied order. Therefore, to correctly match similar synergies, pre and post-fatigue synergies were re-organized to achieve the least amount of error between the two.

To determine if the combination of CWT and muscle synergies is a more effective technique in fatigue analysis to traditional methods, a STFT with 25 ms epochs (the 25 ms window with the greatest mean power) and root mean square (RMS) over the entire

trial were also conducted to quantify mean power frequency (MPF) and EMG amplitude respectively. Squatting velocity was approximately  $60^\circ/s$  and so a 250 ms epoch as previously seen (Potvin and Bent, 1997; Hostens et al., 2004) would include  $15^\circ$  of knee flexion and thus was deemed to not be stationary. Therefore, due to the high squatting rate, a 25 ms epoch for STFT was chosen to reduce the knee flexion to  $1.5^\circ$  per epoch.

## 2.5. Statistical analysis

Coefficients and joint moments were analyzed with Hotelling's paired sample  $T$ -tests (fatigue as independent variable) through statistical parametric mapping (SPM; Pataky, 2010; Penny et al., 2011). Muscle synergies between pre and post-fatigue states were compared with two-tailed Pearson's correlations in SPSS (v 22.0, IBM, USA) and interpreted with Portney and Watkins (2000). Student's paired sample  $T$ -tests were also used to test fatigue-related differences in MnF, MPF, and RMS. Significant differences were determined by  $p$  values  $< 0.05$  while trends were identified as  $0.05 < p < 0.1$ .

## 3. Results

### 3.1. Reconstruction quality

To obtain the 90% VAF minimum with no increases over 5% with an additional synergy, it was found that three synergies were required to reconstruct the observed EMG during two-legged squatting (VAF: 92.8% pre, 90.7% post), while one-legged squatting required four synergies (VAF: 91.1% pre, 94.7% post).

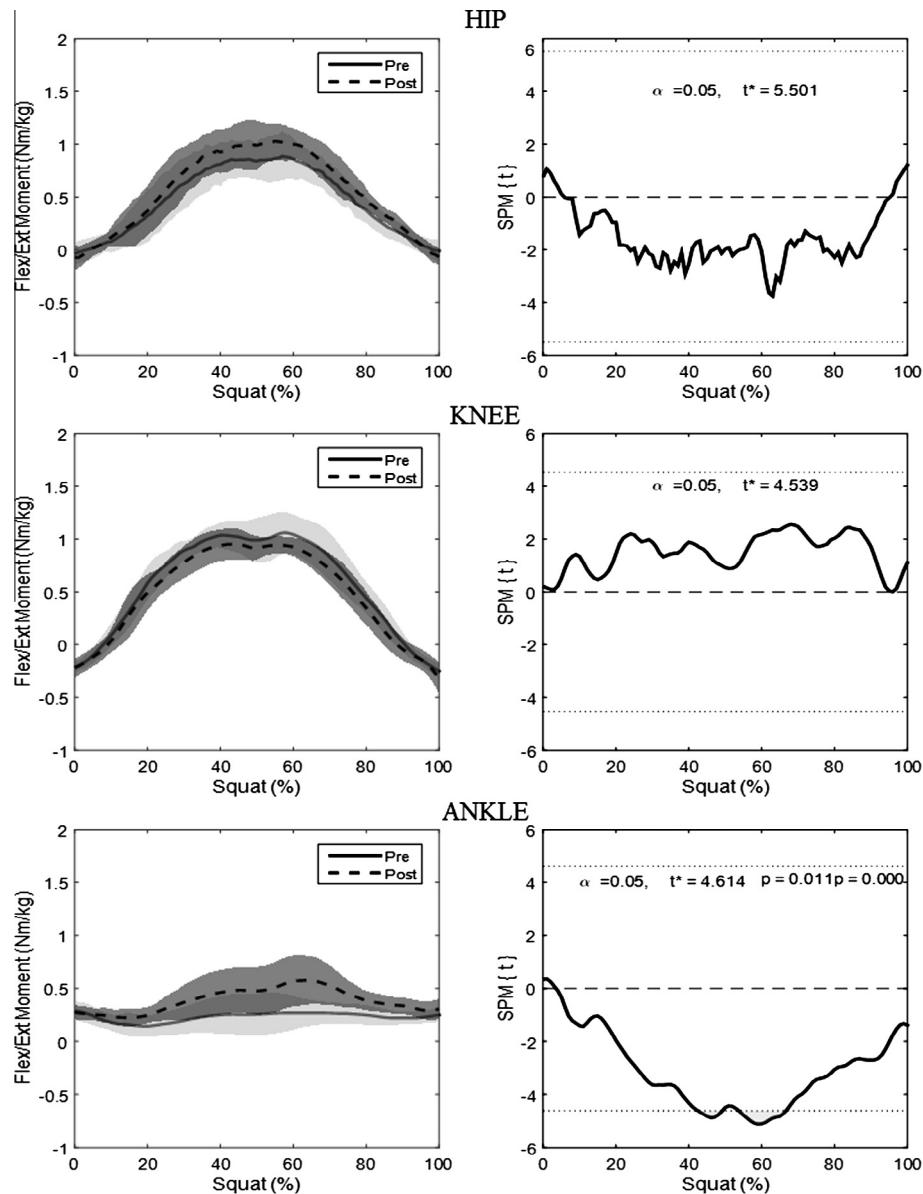
### 3.2. Two-legged squatting

One participant was removed from two-legged squatting analysis due to her placing a considerable amount of her weight on the bench during the post-fatigue trials. Ankle (pre:  $32.1^\circ \pm 2.8^\circ$ , post:  $33.8^\circ \pm 6.2^\circ$ ), knee (pre:  $102.7^\circ \pm 5.4^\circ$ , post:  $106.7^\circ \pm 12.6^\circ$ ), and hip (pre:  $78.1^\circ \pm 7.8^\circ$ , post:  $82.4^\circ \pm 7.2^\circ$ ) sagittal plane range of motions all remained constant between pre and post-fatigue states. Fatigue caused a greater internal extension moment in the ankle throughout the entire movement and significantly ( $p = 0.01$  &  $p < 0.01$ ) between 40% and 70% of the squat (Fig. 1). A decrease in knee and increase in hip internal extension moments were observed after fatigue for the entire squat but failed to reach significance.

Through the use of CWT, mean firing frequency (Fig. 2) significantly decreased after fatigue in the GMAX, GMED, TFL, RFEM, TA, and LGAS. The VMED and MGAS trended towards significant decreases post-fatigue while the SOL trended towards a significant increase post-fatigue. The STFT produced fatigue-related decreases in MPF for the GLTMAX (69 vs 59 Hz,  $p = 0.02$ ), TFL (83 vs 73 Hz,  $p = 0.05$ ), STEN (94 vs 86 Hz,  $p = 0.03$ ) and MGAS (133 vs 117 Hz,  $p = 0.08$ ). Fatigue-related increases in RMS (Table 1) were also observed in the TFL and VLAT while a decrease occurred in the TA.

In two-legged squatting, the percentage of total VAF for synergies one through three prior to fatigue were 41, 18, and 41% respectively, and 42, 19, 40% post-fatigue (Fig. 3). Pre to post-fatigue synergies had very strong correlation for S1 (0.92,  $p < 0.01$ ) good for S2 (0.80,  $p < 0.01$ ) and a moderate correlation for S3 (0.60,  $p = 0.04$ ). No significant differences among coefficients were observed.

S1 was high in vastii activity, reaching its peak at the beginning of ascension. S2, which favoured the VMED, RFEM and TA, was generally used during the descending phase and reached a maximum at approximately the movement's lowest point. S3 remained a general activation of plantar flexors, hamstrings and gluteus muscles



**Fig. 1.** Hip, knee, and ankle flexion/extension moments for two-legged squats during non-fatigued (solid) and fatigued (dash) states. For each joint, the figure on the left displays the mean and standard deviation clouds for pre and post fatigue with positive values indicating internal extension moments. The figure on the right displays the t-statistic continuum determined through statistical parametric mapping.

after fatigue and was weighted the highest in the ascension phase when the body needs to rise against gravity.

### 3.3. One-legged squatting

Ankle (pre:  $31.0^\circ \pm 5.5^\circ$ , post:  $31.3^\circ \pm 5.7^\circ$ ), knee (pre:  $86.4^\circ \pm 13.9^\circ$ , post:  $88.3^\circ \pm 13.8^\circ$ ), and hip (pre:  $75.1^\circ \pm 13.9^\circ$ , post:  $77.6^\circ \pm 16.0^\circ$ ) sagittal plane range of motions all remained constant between fatigue states. After fatigue, the hip internal extension moment was increased throughout the squat and reached significance ( $p < 0.01$ ) between 20% and 40% of the movement (Fig. 4). A systemic decrease in knee internal extension moment was also observed after fatigue and trended towards significance ( $p = 0.08$ ) at 75% of the squat.

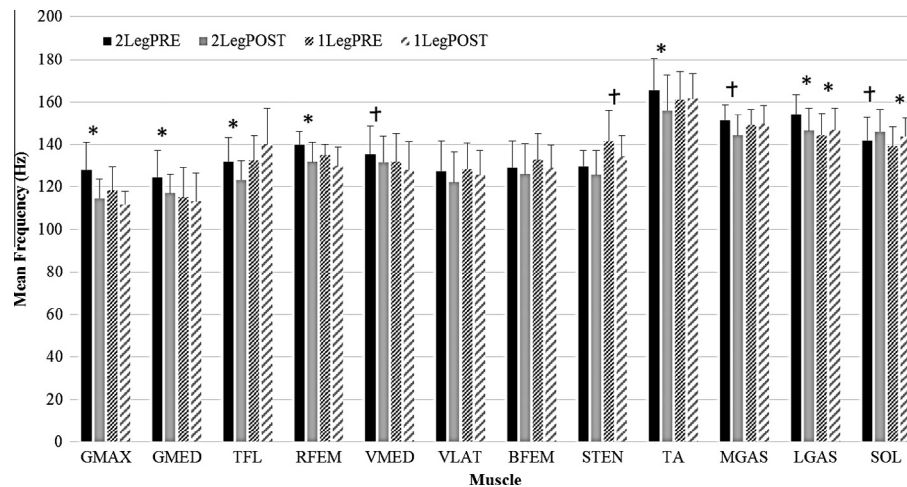
Through CWT, a mean firing frequency (Fig. 2) decrease after fatigue trending towards significance was seen in the STEN while significant increases after fatigue were seen in the LGAS and the SOL. The only difference observed in MPF during one-legged squat-

ting was an increase after fatigue in LGAS from 104 to 115 Hz ( $p = 0.04$ ). Differences in RMS (Table 1) included fatigue-related increases in RFEM and VLAT, while a decrease after fatigue occurred in MGAS.

In one-legged squatting, the percentage of individual VAF for modules one through four pre-fatigue were 18, 23, 31, 28% respectively and 26, 17, 35, 23% post-fatigue respectively. Three of the four synergy pairs had strong correlations between the two fatigue states (S1: 0.78,  $p < 0.01$ ; S2: 0.88,  $p < 0.01$ ; S3: 0.69,  $p = 0.01$ ), while S4 was not correlated between fatigue states (Fig. 5).

S1 corresponded to high knee extensor activation that increased during the descending phase and reached a maximum at approximately the lowest point of the movement. S2 was quite similar to S1 in that it was heavily weighted with knee extensors but there was also a very strong activation of the TA. S3 was high in plantar flexors and gluteus muscles throughout the entirety of the movement, suggesting that these muscles were used for stability during the squat. Pre-fatigue, S4 consisted of general activations through-





**Fig. 2.** Mean Frequencies calculated from continuous wavelet transforms for two and one-legged squats. See text for muscle abbreviations. Asterisks denote differences between fatigue state of  $p < 0.05$  while crosses denote differences of fatigue state of  $0.05 < p < 0.10$ . Error bars represent one standard deviation.

**Table 1**

Root mean square (% MVIC) during one and two-legged squatting before and after fatigue. Greying and bolding indicate significance at  $p < 0.05$  while bolding indicates significance  $0.05 < p < 0.1$ .

MUSCLE	1 LEG PRE	1 LEG POST	2 LEG PRE	2 LEG POST
GLTMAX	17.7 (10.7)	16.0 (6.5)	14.1 (7.1)	16.0 (5.5)
GLTMED	19.1 (03.8)	21.1 (6.1)	12.2 (5.0)	14.5 (4.7)
TFL	13.3 (12.5)	9.0 (7.8)	<b>6.2 (5.2)</b>	<b>8.0 (6.1)</b>
RFEM	<b>23.4 (11.8)</b>	<b>32.0 (8.6)</b>	25.8 (8.8)	31.3 (9.0)
VMED	38.8 (15.6)	47.4 (12.6)	36.7 (9.5)	40.1 (12.9)
VLAT	<b>39.2 (13.4)</b>	<b>52.9 (12.3)</b>	<b>40.5 (11.0)</b>	<b>46.5 (15.1)</b>
BFEM	10.9 (6.70)	12.8 (6.3)	11.3 (9.4)	12.5 (7.5)
STEN	12.4 (7.30)	13.9 (8.4)	9.1 (5.4)	8.8 (5.1)
TA	21.8 (10.3)	22.2 (10.9)	<b>27.0 (14.3)</b>	<b>14.6 (7.6)</b>
MGAS	<b>17.1 (10.2)</b>	<b>9.6 (4.4)</b>	6.5 (2.8)	7.2 (2.5)
LGAS	15.1 (10.4)	10.6 (4.7)	9.9 (10.3)	8.8 (4.0)
SOL	20.6 (11.0)	18.2 (10.7)	11.3 (6.0)	14.9 (11.0)

out the entire movement while post-fatigue, showed a dominance of knee extensors, which increased in the ascension phase. The coefficient of S4 also resembled those of S1 and S2 with its greatest activation at the bottom of the squat. This may indicate that once fatigued, participants could no longer use a general co-activation module but instead needed to adapt a knee extensor specific module based on increases in VMED and RFEM activation in order to compensate for fatigue and to maintain the centre of mass's position against gravity. It appears to be a completely different activation weighting from the pre-fatigue state but due to the large variances as shown by the standard deviation clouds in Fig. 3, the differences between pre and post-fatigue in C4 failed to reach any significance level.

#### 4. Discussion

The objective of this study was to supplement CWTs with muscle synergies in a fatigue analysis to better describe fatigue during dynamic muscle contractions. As hypothesized, the fatigue-related differences manifested in a combination of frequency and activation changes in order to successfully complete the squatting movements.

##### 4.1. Two-legged squatting

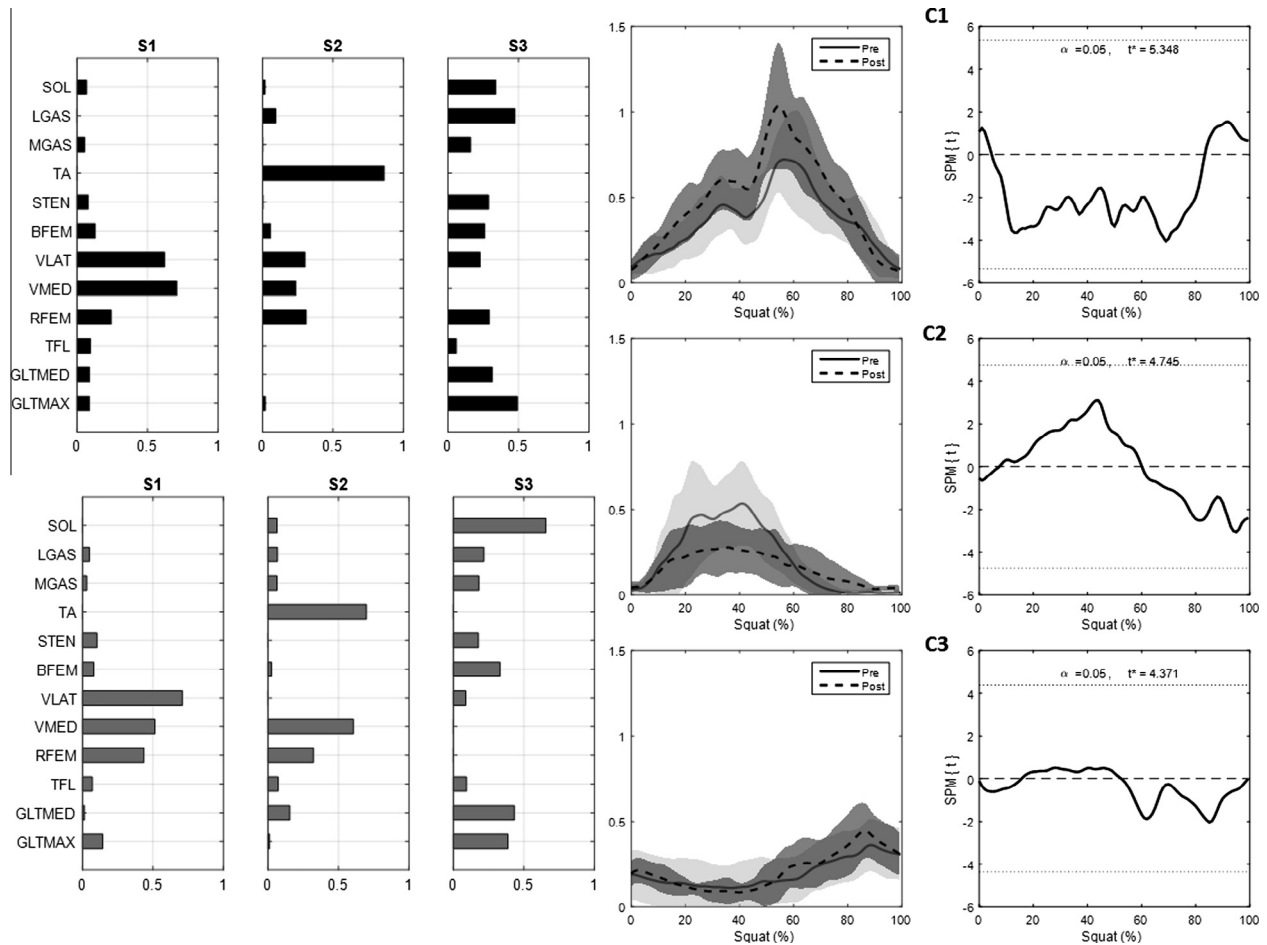
Even though only relatively small changes to muscle activation patterns were observed, many changes to muscle firing frequency

occurred after fatigue. Decreases in MnF were observed in the GMAX, GMED, TFL, RFEM, TA, and LGAS. Despite these many decreases in firing frequency, they appeared to have little effect on the overall joint flexion/extension moments as the only significant fatigue-related difference was observed in the ankle. The observed increase in ankle internal extensor moment can be a result of the increased SOL activation evident in S3 (Fig. 3) as well as its significant increase in firing frequency after fatigue (Fig. 2). Muscle samples from the SOL have shown an increased amount of slow-twitch fibres in this muscle compared to muscles such as the gastrocnemii and VLAT (Gollnick et al., 1974). Therefore, it is plausible that this fibre composition allowed the SOL muscle to function at an efficient level even after the exhausting squatting protocol, which manifested as increased ankle extensor moments.

##### 4.2. One-legged squatting

Few fatigue-related differences were observed in MnF as only the STEN trended towards a significant decrease while the LGAS and SOL had significant increases. The increase in internal hip extension moment was not accounted for by differences in the activations or frequencies of the musculature surrounding the hip. This lead us to believe that the greater extension moment was a result of the faster descending phase after exhaustion (pre: 3.30 s vs post: 2.98 s,  $p = 0.01$ ), despite the metronome timing. This difference in the rate of change of muscle lengths could also affect EMG amplitudes and patterns as has been observed in the lower limb at various contraction velocities (Perry-Rana et al., 2002).

The need for one additional synergy in one-legged squatting is reasonable as the task is more difficult than two-legged squatting. This increase in demand arises as the participants' body weight is shifted from being evenly distributed between two legs to just the test leg in one-legged squatting. Additionally, the participants' base of support is vastly reduced in the one-legged squat and is confined to only the area under the test foot. This difference in demand may also lend support to how fatigue appeared to have manifested dissimilarly between the two tasks. In the two-legged squatting, fatigue-related changes were primarily apparent in frequency content decreases while few alterations were seen in the muscle activation patterns. Contrarily, one-legged squatting had major changes in the activation patterns with very limited differences in the frequency content. Therefore, a more demanding task may require a change in the activation pattern while participants may be able to cope with fatigue in simpler tasks with the same activa-



**Fig. 3.** Muscle synergies (S1–S3) and coefficients (C1–C3) for two-legged squats during non-fatigued (black solid) and fatigued (gray dash) states for each muscle as a function of weighting. For each coefficient, the figure on the left displays the mean weightings and standard deviation clouds for pre and post-fatigue. The figure on the right displays the t-statistic continuum determined through statistical parametric mapping.

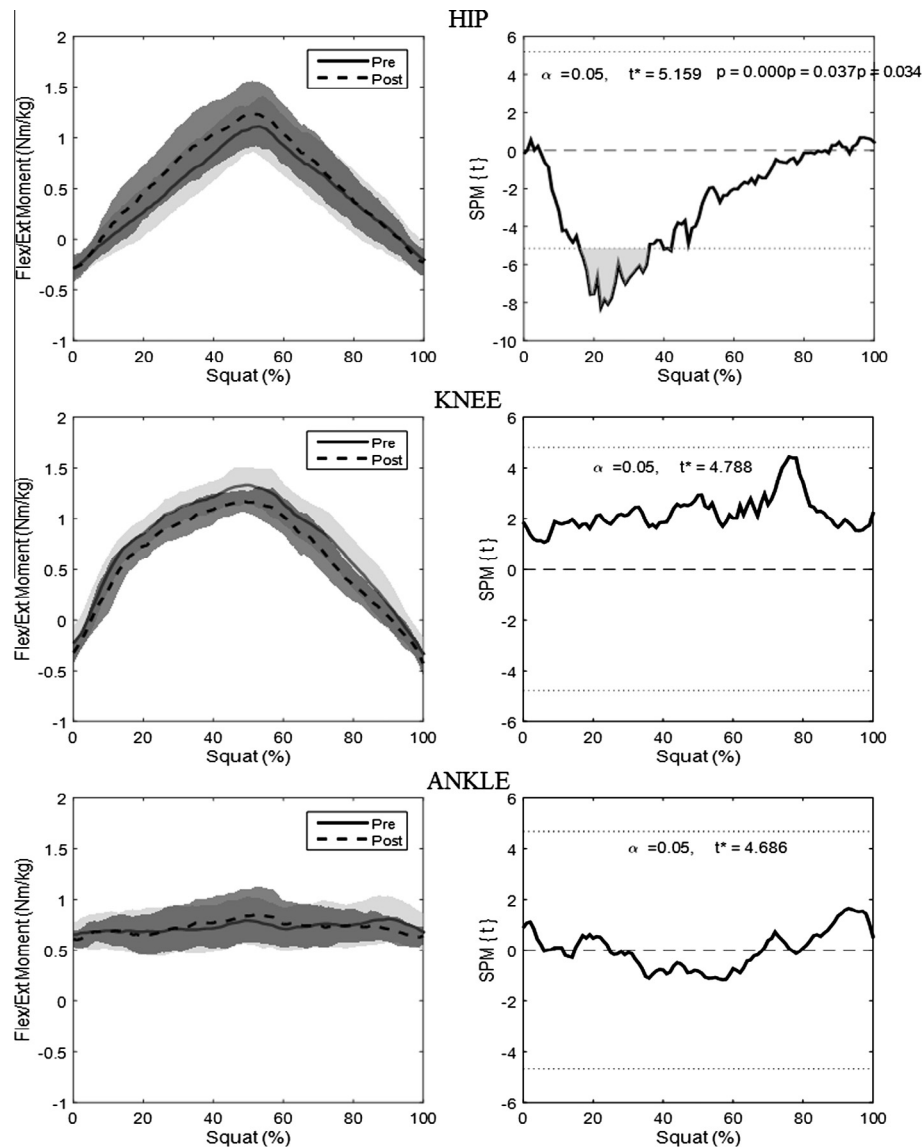
tion patterns. This supports the notion that a combination of methods that investigate both muscle activation patterns and firing frequency content should be implemented when conducting fatigue analyses.

#### 4.3. Suitability to fatigue analysis

The most attractive feature of CWT over STFT is that it has good resolution in both the time and frequency domains. Since the STFT must be done in small enough epochs to assume stationarity, dynamic tasks with large range of motions such as the squatting in the current study need to be windowed into very small intervals. Past studies (Potvin and Bent, 1997; Hostens et al., 2004) who used STFT on dynamic tasks had much smaller range of motions and movement velocities. This enabled them to use a larger epoch (250 ms) than the current study but with the larger time window, they sacrifice resolution in the frequency domain. With the reduction of the window to 25 ms in the current study, the frequency resolution became 40 Hz and therefore the absence of spectral changes may be due to this coarse resolution. The CWT also accounts for the disadvantages as outlined by Farina (2006) in that at any given time during the movement, the electrodes may be positioned differently in relation to the underlying fibres they are recording from. Since the CWT is the process of applying a time-specific scale and translation to a mother wavelet, this method is affected by these electrode shifts to a lesser extent than Fourier transforms, but is still not capable of solving the relative shift with

respect to the origin of action potentials. That said, given that the squat task is cyclical, this artifact is consistent across participants and fatigue states. The current results also suggest that CWT is more sensitive to fatigue-related frequency changes than STFT over short peak activation epochs. When including trends, CWT identified fatigue-related decreases for 8/12 muscles while STFT only identified decreases in 4/12 muscles. Thus, we believe that for dynamic tasks, CWTs are a more effective method of describing muscle firing frequencies and have shown to be sensitive enough to detect fatigue-related changes.

RMS is a technique used to describe changes in EMG amplitude and was shown to be somewhat sensitive to fatigue-related changes in this study (Table 1). Even though it allows for the quantification of muscle activation, RMS provides a single value to represent the entire contraction of interest. The near-infinite number of activation profiles during a movement has been previously described as the muscle redundancy problem (Buchanan and Shreeve, 1996). Therefore, the validity of RMS becomes questionable due to possible changes in activation strategies occurring at different time-points throughout the dynamic task and thus, observed differences cannot be limited to those caused by fatigue. With muscle synergies being a continuous representation of the EMG, we are able to vastly reduce the muscle redundancy problem and limit the observed differences to those attributable to fatigue. Although the CNMF approach is a more robust method, fixing the synergies between subjects could mask inter-subject synergy variability which, in the context of fatigue research, may be of interest.



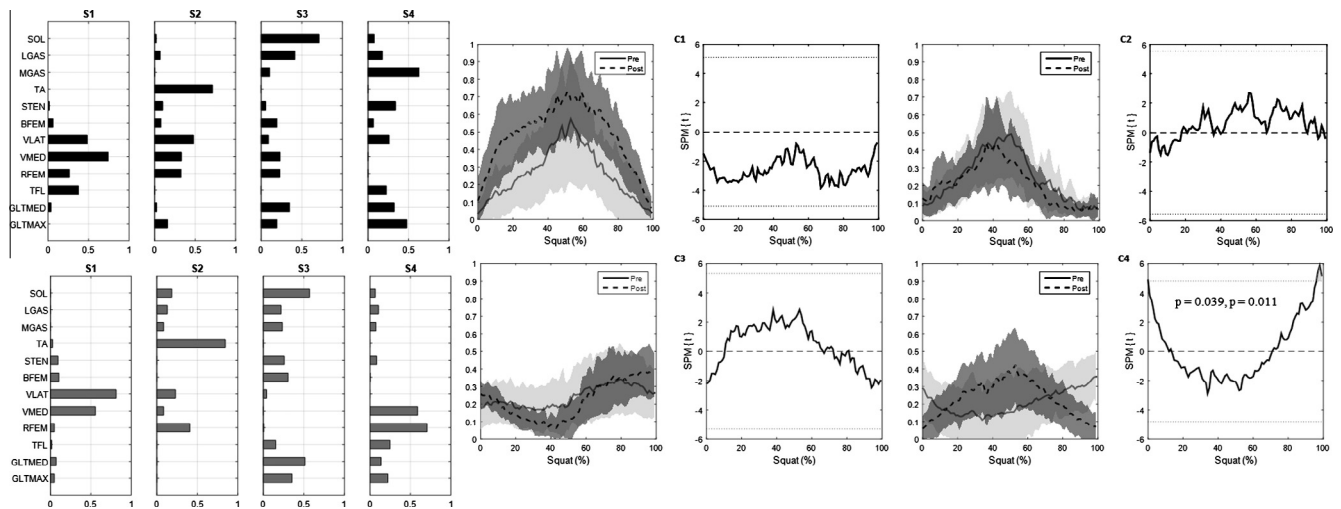
**Fig. 4.** Hip, knee, and ankle flexion/extension moments for one-legged squats during non-fatigued (solid) and fatigued (dash) states. For each joint, the figure on the left displays the mean and standard deviation clouds for pre and post fatigue with positive values indicating internal extension moments. The figure on the right displays the t-statistic continuum determined through statistical parametric mapping.

Nevertheless, using CNMF this inter-subject variability is captured in the coefficients, thus representing a compromise that has greater internal validity. Furthermore, it should also be noted though that the number of muscles included in a synergy analysis has an effect on the resulting VAF (Steele et al., 2013), who compared synergy matches using normalized similarity, which is the correlation coefficient normalized between zero and one where zero indicates similarity due to chance and one is perfect similarity (Tresch et al., 2006). To match the synergies incorporating 30 upper limb muscles to at least 0.8 normalized similarity, at least 11 muscles had to be included in the analysis. The authors also demonstrated that including the muscles that are most dominant in the synergy weightings or largest in size can raise the normalized similarity close to 1.0. Since the 12 muscles included in the present study are the most dominant and largest in the lower limb, the analysis of these muscles should provide a valid representation of the entire lower limb.

A final limitation of this study is that MVICs were used to normalize the EMG signals from a dynamic contraction. During MVICs, the electrodes record from certain muscle fibres while during the

movements, the electrodes are recording from different muscle fibres throughout the range of motion. Other normalization techniques to combat this issue include normalizing to the maximum during the trial or completing isokinetic MVCs, however they also include the uncertainty of when the maximal value occurs during the motions, and thus the contraction velocity and fibres from which the signal was recorded, also leading to uncertainty. In this light, we chose MVICs as they are easier reproduce and were consistent across participants.

It was hypothesized that only subtle fatigue-related changes in muscle synergies would be observed as few were seen in human rowing (Turpin et al., 2011). These authors did not complete any frequency analysis of their EMG pre or post-fatigue so it is within reason to suspect that their lack of activation pattern differences may have been more evident in a decrease in MnF. Furthermore, one and two-legged squatting are arguably more difficult than rowing as the body must maintain balance and remain stable throughout the entire movement and thus explains the more pronounced muscle activation differences observed in the current study compared to those reported in Turpin's work.



**Fig. 5.** Muscle synergies (S1–S4) and coefficients (C1–C4) for one-legged squats during non-fatigued (black solid) and fatigued (gray dash) states for each muscle as a function of weighting. For each coefficient, the figure on the left displays the mean weightings and standard deviation clouds for pre and post fatigue. The figure on the right displays the t-statistic continuum determined through statistical parametric mapping.

In conclusion, it was observed that the combination of methods is an effective way of describing the state of muscle fatigue. The use of muscle synergies and CWTs hold many advantages over RMS and Fourier transforms in the context of fatigue analysis during dynamic tasks and are able to represent fatigue-related differences even though these changes were subtle in joint kinetics.

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