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Patterns of upper limb muscle activation in children with unilateral spastic cerebral palsy: Variability and detection of deviations

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ABSTRACT

Background: The aim of this study was two-fold: (1) to quantify the variability of upper limb electromyographic patterns during elbow movements in typically developing children and children with unilateral spastic cerebral palsy, and to compare different amplitude normalization methods; (2) to develop a method using this variability to detect (a) deviations in the patterns of a child with unilateral spastic cerebral palsy from the average patterns of typically developing children, and (b) changes after treatment to reduce muscle activation.

Methods: Twelve typically developing children ([6.7–15.9yo]; mean 11.0 SD 3.0yo) and six children with unilateral spastic cerebral palsy ([7.9–17.4yo]; mean 12.4 SD 4.0yo) attended two sessions during which they performed elbow extension-flexion and pronation-supination movements. Surface electromyography of the biceps, triceps, brachioradialis, pronator teres, pronator quadratus, and brachialis muscles was recorded. The Likelihood method was used to estimate the inter-trial, inter-session, and inter-subject variability of the electromyography patterns for each time point in the movement cycle. Deviations in muscle patterns from the patterns of typically developing children and changes following treatment were evaluated in a case study of a child with cerebral palsy.

Findings: Normalization of electromyographic amplitude by the mean peak yielded the lowest variability. The variability data were then used in the case study. This method detected higher levels of activation in specific muscles compared with typically developing children, and a reduction in muscle activation after botulinum toxin A injections.

Interpretation: Upper limb surface electromyography pattern analysis can be used for clinical applications in children with cerebral palsy.

1. Introduction

Children with Unilateral Spastic Cerebral Palsy (USCP) frequently have impaired motor control of the upper limb (Steenbergen and Gordon, 2006). In particular, elbow extension (Steenbergen et al., 2000) and supination (Kreulen et al., 2007) may be limited because of spastic contractions of the flexor and pronator muscles respectively (de Bruin et al., 2013; Sarcher et al., 2017, 2015). A lack of active elbow

extension and/or supination restricts autonomy in activities of daily living. A common treatment to reduce excessive muscle activation caused by spasticity or involuntary contractions (Levitt, 2010; Novak et al., 2013) and improve upper limb function is intramuscular botulinum toxin A injections (BTI). To improve supination, both pronator muscles (pronator teres and pronator quadratus) are often treated because, until now, no method existed to clinically determine which of the two muscles is excessively activated during voluntary supination.

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Analysis of patterns of muscle activation during upper limb movements using surface electromyography (EMG) can help to identify muscles that are excessively activated in patients with motor control disorders, and then determine appropriate treatments. The dynamic EMG patterns of an individual with a neurological disorder can be compared to the average patterns of a healthy reference group to determine if they are within normal limits or not. In patients with neurological disorders, the timing (Buurke et al., 2004; Detrembleur et al., 1997; Knuppe et al., 2013; Prosser et al., 2010; Rose et al., 1999, 1994; Sarcher et al., 2015) and magnitude (Meyns et al., 2016; Prosser et al., 2010; Sarcher et al., 2015; Unnithan et al., 1996) of muscle activation are often altered. Co-activations (concomitant activation of the agonist and antagonist muscle) are frequent in individuals with spastic cerebral palsy (Gracies, 2005), and can restrict active movement. Pathological extra-segmental activations (also called synkinesis, mass movements, or overflow) (Gracies, 2005) produce involuntary movements. Analysis of dynamic EMG patterns can help to distinguish between these different types of pathological activation. It can also be useful to evaluate the effect of treatment (Buurke et al., 2004; Grunt et al., 2010).

Several sources of variability may affect dynamic EMG patterns and should be considered during their interpretation. These sources include 1) the natural variability that occurs between consecutive cycles of a movement (inter-trial); 2) the variability that occurs between measurement sessions (inter-session) due to the extrinsic variability of the measurement process (e.g., electrode placement, skin condition) and the natural variability of the disorder over time or with the time of day; 3) variability between subjects (inter-subject) to achieve the same movement: this variability must be considered when using the EMG patterns of several subjects that are averaged to provide a reference, and 4) variability between assessors (inter-assessor variability), which is not explored in this study.

It is not possible to evaluate all the sources of variability for individual patients since this would imply carrying out several measurement sessions. One solution, therefore, is to determine the average variability of the dynamic EMG patterns of a sample of typically developing (TD) children and a sample of children with USCP and to use this data as a reference for the interpretation of dynamic EMG patterns in children with USCP (Schwartz et al., 2004). This has already been done for lower limb muscles during gait in children (Granata et al., 2005; Tirosh et al., 2013), however the different sources of variability for upper limb movements have not yet been quantified in TD children or children with USCP.

This study had two aims:

- (1) To quantify the different sources of variability of upper limb surface electromyographic (EMG) patterns during elbow movements in TD children and children with USCP. In addition to providing the data necessary for the comparison of EMG patterns, we also wished to clarify two issues: a) whether inter-session variability is larger than inter-trial variability, and b) whether upper limb EMG pattern variability differs between children with USCP and TD children. We also compared three methods of EMG amplitude normalization to determine which was less variable with regards to our aims.
- (2) To determine whether differences in EMG patterns can be detected a) between the EMG patterns of a child with USCP and the average EMG patterns of a group of TD children and b) between EMG patterns of a child with USCP before and after a treatment to reduce muscle activation (botulinum toxin A injections). This part of the study was based on a case study of one child with USCP.

Between group comparisons of upper limb EMG patterns were not carried out in this study since other studies have already addressed this subject: differences in the amplitude and timing of upper limb EMG patterns between populations with and without USCP have previously been demonstrated (de Bruin et al., 2013; Sarcher et al., 2017, 2015).

2. Methods

2.1. Experimental protocol

Convenience samples of 12 TD children (aged from 6.7 to 15.9 years, mean 11.0 years, SD 3.0) and 6 children with USCP (aged from 7.9 to 17.4 years, mean 12.4 years, SD 4.0) were included. The upper limb Physician Rating Scale scores of the children with USCP ranged from 11/24 to 22/24. Care was taken to include a wide range of ages in both groups and different levels of upper limb disability in the group with USCP. Participants were volunteers recruited from the patient database of the motion analysis laboratory. Each child participated in two measurement sessions between one day and one month apart, all conducted by the same clinical scientist who is highly experienced in the acquisition of EMG data in children. The fact that only one assessor collected the data ensured there was no inter-assessor error in this study; however inter-assessor error must be considered if several assessors are involved in data collection.

A case study to evaluate the pertinence of individual analysis of EMG patterns was also carried out on one of the children with USCP (Child 1; 13 years old; upper limb Physician Rating Scale 12/24) who was treated with BTI.

Our local ethics committee Comité de Protection des Personnes Ouest IV (France) approved the study (IdRCB No 2016-A01314-47). Written informed consent was obtained from both parents of each child, and informed assent was obtained from all children. Data were processed in accordance with the requirements of the French National Commission for Data Protection and Liberties (CNIL).

During each session, the children performed five cycles of elbow extension/flexion (EF) and five cycles of elbow pronation/supination (PS) at 0.5 Hz. They were asked to carry out maximal elbow extension and flexion, or pronation and supination movements, in synchrony with the beeps of a metronome. A fixed movement frequency was chosen because movement speed has previously been found to have an effect on EMG variability in children during gait (Shuman et al., 2016; Tirosh et al., 2013). Children with USCP performed the movements with their affected arms and the arm used by the TD children was determined by a randomization process. Surface EMG of maximum isometric voluntary contractions (MIVC) against manual resistance for elbow flexion, extension, pronation and supination were also recorded. Surface EMG signals of six muscles: biceps brachii, triceps brachii, brachioradialis, pronator teres, pronator quadratus, and brachialis were recorded using disposable bipolar electrodes (DUO F3010 – rectangular shape 21 * 41 mm; recording diameter 10 mm; Ag-AgCl, lithium chloride gel; unit distance 22 mm; LTT FIAB Vicchio, Firenze, Italy) and sampled at 1000 Hz with a Cometa ZeroWire system (Milan, Italy). The skin was prepared according to established guidelines (Hermens et al., 2000). To determine the placement of the electrodes, each muscle was palpated (Basmajian, 1982) during isometric voluntary contractions against manual resistance of:

- elbow flexion with the elbow in supination for the biceps brachii (Hermens et al., 2000)
- elbow flexion with the elbow in neutral pronation-supination for the brachioradialis (Staudenmann and Taube, 2015)
- elbow flexion with the elbow in pronation for the brachialis on the lateral side of the arm (Staudenmann and Taube, 2015)
- elbow extension for the triceps brachii (Hermens et al., 2000)
- elbow pronation for the pronator teres (Malanga and Campagnolo, 1994; Remaley et al., 2015).

Surface EMG electrodes were placed on the most prominent bulge of each muscle belly (Fig. 1). For the pronator quadratus, surface EMG electrodes were placed on the anterior side of the wrist, perpendicular to the forearm (Basmajian, 1982; Malanga and Campagnolo, 1994) (Fig. 1).

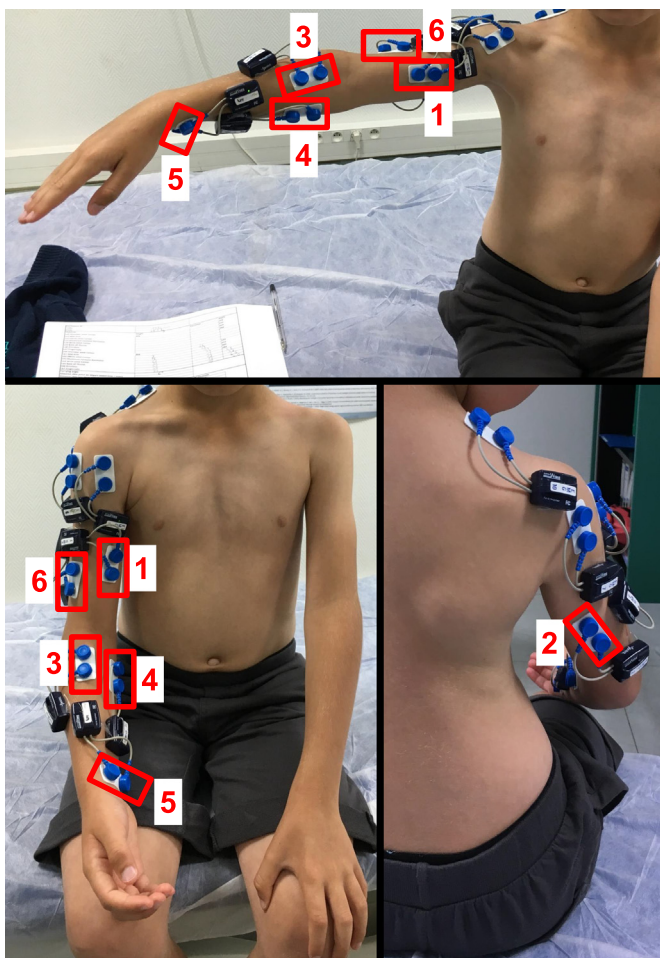


Fig. 1. A typically developing child equipped with surface electromyographic electrodes placed on the biceps brachii (1), the triceps brachii (2), the brachioradialis (3), the pronator teres (4), the pronator quadratus (5), and the brachialis (6).

Post processing of the EMG was performed using custom MATLAB (Mathworks, Natick (MA), USA) routines and the open-source Biomechanical ToolKit library (Barre and Armand, 2014).

The EMG signals were band-pass filtered (10–450 Hz, Butterworth zero-lag 4th order), full-wave rectified, and smoothed with a low-pass filter (50 Hz, Butterworth zero-lag 2nd order) according to previously-published protocols (Sarcher et al., 2017, 2015). EMG patterns were interpolated linearly to 100 points, which represented a whole movement cycle, i.e., for EF, extension occurred from 0 to 50% of the cycle and flexion from 51 to 100%. For PS, pronation occurred from 0 to 50% and supination from 51 to 100%.

It is normal procedure to normalize the EMG signal amplitude so that datasets acquired with different hardware can be compared, however no optimal normalization method has yet been determined (Burden and Bartlett, 1999; Hug, 2011). This is an important issue since the method used affects the subsequent calculation of inter-subject variability. We chose to use the averaged EMG patterns of the TD children as the reference to determine deviations in the patterns of the children with USCP, defined as pathological muscle activation. For this purpose, therefore, differences between the TD children were not of interest, and it was important for inter-subject variability within the TD group to be as small as possible. We chose to study three different methods of EMG amplitude normalization in order to identify the method that resulted in the lowest total inter-subject variability in the TD group. The three different methods applied to each muscle were: 1) normalization by the peak value measured over the averaged EF and

the averaged PS movements (average of the 5 EF cycles and average of the 5 PS cycles); 2) normalization by the peak value in all movement trials (5 EF trials and 5 PS trials); and 3) normalization by the peak value using a 200 ms plateau of the MIVC against manual resistance. Non-normalized EMG patterns were also included in the analysis.

2.2. Variability of the EMG patterns

R 3.2.2 software (R Core Team, 2015) was used for the statistical analyses, using the file “EstVcomp.R” provided as supplementary material in (Chia and Sangeux, 2017), and the “nlme” package for the linear mixed models.

The sources of variability of the upper limb surface EMG patterns were quantified using the framework determined by Schwartz et al. (2004) and the statistical analysis of Chia and Sangeux (2017). We assumed the following linear mixed model would represent our data:

$$\Phi_{ijk} = \mu + Sub_i + Sess_{ij} + Trial_{ijk}$$

where μ denotes the overall mean, and the remaining terms denote the respective random subject, session and trial effects, each following an independent normal distribution with zero mean and variance σ_{source}^2 for their respective source. The residual maximum likelihood method estimates all unknown parameters directly by maximizing the likelihood function implied by the linear mixed model (Brown and Prescott, 2014; Chia and Sangeux, 2017; Patterson and Thompson, 1971).

For the EMG patterns, with and without amplitude normalization, Inter-trial $\sigma_{Inter-trial}^{2USCP/TD}$, Inter-session $\sigma_{Inter-session}^{2USCP/TD}$, and Inter-subject $\sigma_{Inter-subject}^{2USCP/TD}$ variance components were estimated for every point in the movement cycle and averaged over the movement cycle. A mixed model was used to analyze the differences between groups for inter-session and inter-trial mean standard deviations, with a random effect ‘muscle X during movement Y’.

The ratio of the inter-session to inter-trial variability, $r_{Session} = \frac{\sigma_{Inter-session}^{USCP/TD}}{\sigma_{Inter-trial}^{USCP/TD}}$, with N the number of cycles, and the ratio of the

inter-subject to inter-trial variability, $r_{Subject} = \frac{\sigma_{Inter-subject}^{USCP/TD}}{\sigma_{Inter-trial}^{USCP/TD}}$, were also re-

ported because they provided valuable information on the main sources of variability in the EMG patterns.

The total relative inter-subject variability in the TD group, $r_{Subject_Total} = \sum_{muscle} \sum_{EF,PS} \frac{\sigma_{Inter-subject}^{TD}}{\sigma_{Inter-trial}^{TD}}$, was calculated for each normalization method. The amplitude normalization method, which provided the smallest relative inter-subject variability in the TD group, was retained in order to compare the EMG patterns.

2.3. Comparison of EMG patterns: case study – child 1

This part of the study was based on a case study of Child 1. Child 1 had a significant impairment of active supination, which was clinically suspected to be due to excessive activation of the pronator muscles. EMG pattern analysis was carried out to determine which of the two pronator muscles (pronator teres and the pronator quadrates) was excessively activated during supination and botulinum toxin injection (BTI) was planned. The initial EMG pattern analysis demonstrated excessive activation of the pronator teres muscle, therefore the treatment involved injection of 50 units of onabotulinum toxin-A (Botox®) in two sites of this muscle under electrical stimulation guidance. A second EMG pattern analysis was carried out one month after the BTI.

The aim of this study was to determine whether upper limb surface EMG pattern analysis could detect a) differences in muscle activation between Child 1 and a group of TD children, and b) differences in the EMG pattern before and one month after BTI in Child 1. Evaluation of the functional outcome of the BTI was not the aim of this case study,

therefore is not described in detail. However, it is important to note that the BTI was considered to be effective since one-month later there was no spasticity of the pronator muscles and active supination had increased considerably.

Inter-group differences in EMG patterns were not evaluated as the aim was not to highlight general differences between children with USCP and TD children, but to evaluate the pertinence of individualized EMG pattern analysis for children with USCP, based on this case study.

The variability of the EMG patterns was calculated for each child, muscle and movement, at each time point of the movement cycle as a combination of the inter-trial and inter-session variance of the corre-

$$\text{sponding group: } \sqrt{\frac{\sigma_{USCP/TD}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{USCP/TD}^2 \sigma_{Inter-trial}^2}{5}}{2}}$$

For each muscle and movement, the variability of the EMG patterns for a group (TD or USCP) was calculated at each time point of the movement cycle as a combination of the inter-trial, inter-session, and inter-subject variance of the corresponding group:

$$\sqrt{\frac{\sigma_{USCP/TD}^2 \sigma_{Inter-subject}^2 + \sigma_{USCP/TD}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{USCP/TD}^2 \sigma_{Inter-trial}^2}{5}}{2}} \quad (\text{Chia and Sangeux, 2017}).$$

Standard statistical techniques were used to detect significant differences between EMG patterns (Schwartz et al., 2004).

Case A₁: For the comparison of the EMG patterns during session 1 (S1) (before BTI) of Child 1 and the EMG patterns of the TD group, the test statistic used was:

$$z_{A_1}(t) = \frac{\Phi^{USCP1_{S1}}(t) - \Phi^{TDgroup}(t)}{\sqrt{\frac{\sigma_{USCP}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{USCP}^2 \sigma_{Inter-trial}^2}{5}}{2} + \frac{\sigma_{TD}^2 \sigma_{Inter-subject}^2 + \sigma_{TD}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{TD}^2 \sigma_{Inter-trial}^2}{5}}{2}}}$$

Case A₂: For the comparison of the EMG patterns during session 2 (S2) (post BTI) of Child 1 and the EMG patterns of the TD group, the test statistic used was:

$$z_{A_2}(t) = \frac{\Phi^{USCP1_{S2}}(t) - \Phi^{TDgroup}(t)}{\sqrt{\frac{\sigma_{USCP}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{USCP}^2 \sigma_{Inter-trial}^2}{5}}{2} + \frac{\sigma_{TD}^2 \sigma_{Inter-subject}^2 + \sigma_{TD}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{TD}^2 \sigma_{Inter-trial}^2}{5}}{2}}}$$

Case B: For the comparison of the EMG patterns during sessions 1 and 2 (pre-post BTI) of Child 1, the test statistic used was:

$$z_B(t) = \frac{\Phi^{USCP1_{S1}}(t) - \Phi^{USCP1_{S2}}(t)}{\sqrt{\sigma_{USCP}^2 \sigma_{Inter-session}^2 + \frac{\sigma_{USCP}^2 \sigma_{Inter-trial}^2}{5}}}$$

The terms $\Phi^{USCP1_{S1}}(t)$ and $\Phi^{USCP1_{S2}}(t)$ denote the mean EMG pattern for sessions 1 and 2 of Child 1 respectively. The term $\Phi^{TDgroup}(t)$ denotes the mean EMG pattern for the TD children.

The null hypothesis was that the population means were equivalent, so $z(t) = 0$. Similarly to (Schwartz et al., 2004), we used a probability of type I error of 5%, meaning that population means were defined as different if the p-value was less than 0.05. p-Values are reported graphically for each time point of the movement cycle. Type II errors are not reported in this study.

3. Results

3.1. EMG pattern variability

Tables 1A–1D shows the inter-trial, inter-session, and inter-subject mean standard deviations for both groups during EF and PS for: 1) EMG pattern amplitude normalized by the peak value of the averaged movement trials (Table 1A); 2) EMG pattern amplitude normalized by the peak value of all movement trials (Table 1B); 3) EMG pattern amplitude normalized by the peak value maintained for at least 200 ms of

Table 1A

Mean standard deviation (%) (square root of the average inter-session, inter-trial and inter-subject variance components), and ratios $r_{Session}$ of the inter-session to inter-trial variability (no unit), $r_{Subject}$ of the inter-subject to inter-trial variability (no unit), for the normalized post processed EMG waveforms of the typically developing (TD) group and the group with Unilateral Spastic Cerebral Palsy (USCP), with normalization by the peak value measured over the averaged movement trials.

		Flexion/extension		Pronation/supination	
		TD	USCP	TD	USCP
$\sigma_{Inter-session}$	Biceps	10.8	11.1	10.0	8.7
	Triceps	12.8	13.1	3.3	12.2
	Brachioradialis	10.9	8.8	3.9	8.4
	Pronator teres	9.1	6.6	10.8	11.6
	Pronator quadratus	8.1	12.0	8.6	10.8
	Brachialis	12.8	10.8	6.4	7.7
$\frac{\sigma_{Inter-trial}}{\sqrt{N}}$	Biceps	11.7	9.3	8.4	6.4
	Triceps	16.7	13.4	3.0	6.1
	Brachioradialis	12.9	10.9	2.7	4.2
	Pronator teres	12.1	10.6	12.9	8.5
	Pronator quadratus	6.8	11.4	13.8	8.9
	Brachialis	9.7	9.5	4.4	5.6
$\sigma_{Inter-subject}$	Biceps	8.5	8.3	6.4	2.4
	Triceps	11.8	15.2	2.3	4.8
	Brachioradialis	8.1	11.4	1.9	4.6
	Pronator teres	8.7	13.9	10.0	12.8
	Pronator quadratus	3.5	13.4	12.6	16.7
	Brachialis	7.9	7.4	6.2	7.3
$r_{Session} = \frac{\sigma_{Inter-session}}{\frac{\sigma_{Inter-trial}}{\sqrt{N}}}$	Biceps	0.9	1.2	1.2	1.4
	Triceps	0.8	1.0	1.1	2.0
	Brachioradialis	0.8	0.8	1.4	2.0
	Pronator teres	0.8	0.6	0.8	1.4
	Pronator quadratus	1.2	1.1	0.6	1.2
	Brachialis	1.3	1.1	1.4	1.4
$r_{Subject} = \frac{\sigma_{Inter-subject}}{\frac{\sigma_{Inter-trial}}{\sqrt{N}}}$	Biceps	0.7	0.9	0.8	0.4
	Triceps	0.7	1.1	0.8	0.8
	Brachioradialis	0.6	1.0	0.7	1.1
	Pronator teres	0.7	1.3	0.8	1.5
	Pronator quadratus	0.5	1.2	0.9	1.9
	Brachialis	0.8	0.8	1.4	1.3

the MIVC against manual resistance (Table 1C); iv) non-normalized EMG patterns (Table 1D).

- i) For the amplitude normalization by the peak value of the averaged movement trials, no between-group differences were found for either the inter-session ($F = 1.54$; $p = 0.24$) or the inter-trial mean standard deviations ($F = 1.01$; $p = 0.34$). Ratios of the inter-session to inter-trial variability, $r_{Session} = \frac{\sigma_{USCP/TD} \sigma_{Inter-session}}{\frac{\sigma_{USCP/TD} \sigma_{Inter-trial}}{\sqrt{N}}}$, were between 0.6 and 1.4 for the TD children, and between 0.6 and 2 for the children with USCP.
- ii) For the amplitude normalization by the peak value of all movement trials, no between-group differences were found for either the inter-session ($F = 0.34$; $p = 0.57$) or the inter-trial mean standard deviations ($F = 0.06$; $p = 0.81$). Ratios of the inter-session to inter-trial variability $r_{Session} = \frac{\sigma_{USCP/TD} \sigma_{Inter-session}}{\frac{\sigma_{USCP/TD} \sigma_{Inter-trial}}{\sqrt{N}}}$ were between 0.9 and 1.8 for the TD children and between 0.8 and 1.7 for the children with USCP.
- iii) For the amplitude normalization by the peak value maintained for at least 200 ms of the MIVC against manual resistance, no statistical between-group differences were found for either the inter-session ($F = 0.32$; $p = 0.59$) or the inter-trial mean standard deviations ($F = 0.47$; $p = 0.51$). Ratios of the inter-session to inter-trial variability $r_{Session} = \frac{\sigma_{USCP/TD} \sigma_{Inter-session}}{\frac{\sigma_{USCP/TD} \sigma_{Inter-trial}}{\sqrt{N}}}$ were between 0.8 and 1.8 for the TD children and between 0.7 and 1.8 for the children with USCP.

Table 1B

Mean standard deviation (%) (square root of the average inter-session, inter-trial and inter-subject variance components), and ratios r_{Session} of the inter-session to inter-trial variability (no unit), r_{Subject} of the inter-subject to inter-trial variability (no unit), for the normalized post processed EMG waveforms of the typically developing (TD) group and the group with Unilateral Spastic Cerebral Palsy (USCP), with normalization by the peak value measured in all movement trials.

		Flexion/extension		Pronation/supination	
		TD	USCP	TD	USCP
$\sigma_{\text{Inter-session}}$	Biceps	4.4	5.7	2.4	2.9
	Triceps	4.2	4.8	1.0	1.9
	Brachioradialis	4.8	3.5	1.3	2.6
	Pronator teres	5.1	3.2	3.2	3.7
	Pronator quadratus	2.4	4.8	4.6	3.7
$\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}$	Brachialis	5.8	5.6	3.2	2.5
	Biceps	3.8	4.0	2.6	2.4
	Triceps	4.6	3.8	1.1	1.5
	Brachioradialis	3.9	4.0	0.8	1.5
	Pronator teres	3.9	3.9	3.5	2.9
$\sigma_{\text{Inter-subject}}$	Pronator quadratus	2.1	2.8	4.4	3.4
	Brachialis	3.9	4.2	1.8	2.5
	Biceps	3.9	3.7	2.8	0.9
	Triceps	3.8	5.6	1	1.6
	Brachioradialis	2.4	4.8	0.4	1.7
$r_{\text{Session}} = \frac{\sigma_{\text{Inter-session}}}{\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}}$	Pronator teres	3.5	6.1	4.5	3.9
	Pronator quadratus	1.2	2.0	4.6	8.6
	Brachialis	2.8	2.6	1.7	4.1
	Biceps	1.1	1.4	0.9	1.2
	Triceps	0.9	1.3	0.9	1.2
$r_{\text{Subject}} = \frac{\sigma_{\text{Inter-subject}}}{\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}}$	Brachioradialis	1.2	0.9	1.6	1.7
	Pronator teres	1.3	0.8	0.9	1.3
	Pronator quadratus	1.1	1.7	1.0	1.1
	Brachialis	1.5	1.3	1.8	1.0
	Biceps	1.0	0.9	1.1	0.4
	Triceps	0.8	1.5	0.9	1.1
	Brachioradialis	0.6	1.2	0.5	1.1
	Pronator teres	0.9	1.6	1.3	1.4
	Pronator quadratus	0.6	0.7	1.0	2.5
	Brachialis	0.7	0.6	1.0	1.7

iv) For the non-normalized EMG patterns, no between-group differences were found for either the inter-session ($F = 1.75$; $p = 0.21$) or the inter-trial mean standard deviations ($F = 0.47$; $p = 0.51$). Ratios of the inter-session to inter-trial variability, $r_{\text{Session}} = \frac{\sigma_{\text{USCP/TD}}^{\text{Inter-session}}}{\frac{\sigma_{\text{USCP/TD}}^{\text{Inter-trial}}}{\sqrt{N}}}$, were between 0.7 and 1.8 for the TD children and between 0.6 and 3.3 for the children with USCP.

The normalization method with the lowest inter-subject variance over inter-trial variance ratio in the TD group

$(r_{\text{Subject_Total}} = \sum_{\text{muscle}} \sum_{\text{EF,PS}} \frac{\sigma_{\text{Inter-subject}}^{\text{TD}}}{\frac{\sigma_{\text{Inter-trial}}^{\text{TD}}}{\sqrt{N}}})$ was the normalization by the peak value in the averaged movement trials ($r_{\text{Subject_Total}} = 9.4$); followed by normalization by the peak value in all movement trials ($r_{\text{Subject_Total}} = 10.4$); then normalization by the peak value during MIVC ($r_{\text{Subject_Total}} = 11.8$); and finally no normalization ($r_{\text{Subject_Total}} = 13.2$). Therefore, amplitude normalization by the peak value in the averaged movement trials was retained for the comparison of EMG patterns.

3.2. Comparison of EMG patterns

The following EMG patterns were compared:

Case A_1: Point-by-point evaluation of deviations between the activation of the pronator teres (Fig. 2) and the pronator quadratus (Fig. 3) muscles for both EF and PS movements during S1 of Child 1 and the TD group was carried out. During EF, there was no deviation of the

Table 1C

Mean standard deviation (%) (square root of the average inter-session, inter-trial and inter-subject variance components), and ratios r_{Session} of the inter-session to inter-trial variability (no unit), r_{Subject} of the inter-subject to inter-trial variability (no unit), for the normalized post processed EMG waveforms of the typically developing (TD) group and the group with Unilateral Spastic Cerebral Palsy (USCP), with normalization by the peak value maintained for at least 200 ms of the Maximal Isometric Voluntary Contractions against manual resistance.

		Flexion/extension		Pronation/supination	
		TD	USCP	TD	USCP
$\sigma_{\text{Inter-session}}$	Biceps	3.8	3.8	1.0	1.7
	Triceps	2.7	0.8	0.4	0.6
	Brachioradialis	3.9	3.6	0.5	0.5
	Pronator teres	2.7	2.6	1.7	1.9
	Pronator quadratus	1.1	2.6	3.0	1.1
$\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}$	Brachialis	5.1	3.1	1.2	2.5
	Biceps	2.1	2.1	1.3	1.4
	Triceps	2.0	1.0	0.4	0.4
	Brachioradialis	2.5	2.1	0.4	0.6
	Pronator teres	2.1	2.0	1.7	1.6
$\sigma_{\text{Inter-subject}}$	Pronator quadratus	1.1	2.1	2.7	1.6
	Brachialis	2.8	2.0	0.9	1.6
	Biceps	2.1	2.6	1.8	0.9
	Triceps	1.9	2.0	0.4	0.6
	Brachioradialis	2.0	2.2	0.3	1
$r_{\text{Session}} = \frac{\sigma_{\text{Inter-session}}}{\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}}$	Pronator teres	1.7	3.7	1.7	2.6
	Pronator quadratus	0.8	3.2	3.6	2.6
	Brachialis	2.5	1.9	1.1	3.0
	Biceps	1.8	1.8	0.8	1.2
	Triceps	1.3	0.8	1.1	1.5
$r_{\text{Subject}} = \frac{\sigma_{\text{Inter-subject}}}{\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}}$	Brachioradialis	1.5	1.7	1.2	0.8
	Pronator teres	1.3	1.3	1.0	1.2
	Pronator quadratus	1.0	1.2	1.1	0.7
	Brachialis	1.8	1.5	1.3	1.6
	Biceps	1.0	1.2	1.4	0.6
	Triceps	0.9	1.9	1.1	1.5
	Brachioradialis	0.8	1.1	0.7	1.6
	Pronator teres	0.8	1.8	1.0	1.7
	Pronator quadratus	0.7	1.5	1.3	1.7
	Brachialis	0.9	0.9	1.2	1.9

pronator teres activation, and pronator teres activation was significantly higher during supination (70–100% of the PS cycle) (Fig. 2). During PS, activation of the pronator quadratus was significantly lower mid-cycle (55–70% of the PS cycle) and activation of the pronator quadratus was significantly higher during extension (0–50% of the EF cycle) (Fig. 3).

Case B: The point-by-point assessment of deviations between S1 and S2 of Child 1 with USCP was performed for the pronator teres (Fig. 4) and the pronator quadratus muscles (Fig. 5) for both EF and PS movements. There was no difference in the activation of the pronator teres during EF, but during supination, pronator teres activation was significantly reduced (70–100% of the PS cycle) (Fig. 4). No major difference was detected for pronator quadratus activation during EF or PS (Fig. 5).

Case A_2: Point-by-point assessment of deviations between S2 of child 1 with USCP and the TD group was only carried out for the pronator teres for both EF and PS movements (Fig. 6). The results showed that there was no deviation of pronator teres activation during EF and pronator teres activation was significantly lower during pronation (30–60% of the PS cycle) (Fig. 6).

4. Discussion

We quantified the inter-trial, inter-session, and inter-subject variability of upper limb surface EMG patterns during elbow extension/flexion and pronation/supination movements in convenience samples

Table 1D

Mean standard deviation (%) (square root of the average inter-session, inter-trial and inter-subject variance components), and ratios r_{Session} of the inter-session to inter-trial variability (no unit), r_{Subject} of the inter-subject to inter-trial variability (no unit), for non-normalized post processed EMG waveforms of the typically developing (TD) group and the group with Unilateral Spastic Cerebral Palsy (USCP).

		Flexion/extension		Pronation/supination	
		TD	USCP	TD	USCP
$\sigma_{\text{Inter-session}}$	Biceps	3.7	4.8	1.4	1
	Triceps	1.9	0.5	0.3	0.4
	Brachioradialis	4.2	3.1	0.6	1.5
	Pronator teres	1.7	7.3	0.9	3.2
	Pronator quadratus	0.4	1.3	0.8	4.2
	Brachialis	4.1	1.6	1.0	2.2
$\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}$	Biceps	2.9	2.2	1.9	1.3
	Triceps	1.5	0.8	0.3	0.3
	Brachioradialis	3.0	2.0	0.5	0.5
	Pronator teres	1.4	2.2	1.3	1.4
	Pronator quadratus	0.4	1.4	0.9	1.3
	Brachialis	2.3	1.0	0.8	1.1
$\sigma_{\text{Inter-subject}}$	Biceps	3.0	4.3	2.9	2.2
	Triceps	1.3	1.9	0.4	0.6
	Brachioradialis	3.2	3.1	0.4	0.4
	Pronator teres	1.4	2.2	1.5	1.6
	Pronator quadratus	0.2	1.1	1.5	1.3
	Brachialis	2.7	1.2	0.8	2.0
$r_{\text{Session}} = \frac{\sigma_{\text{Inter-session}}}{\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}}$	Biceps	1.3	2.1	0.7	0.8
	Triceps	1.3	0.6	1.1	1.3
	Brachioradialis	1.4	1.5	1.2	2.8
	Pronator teres	1.2	3.3	0.7	2.2
	Pronator quadratus	1.0	0.9	0.9	3.1
	Brachialis	1.8	1.6	1.2	2.0
$r_{\text{Subject}} = \frac{\sigma_{\text{Inter-subject}}}{\frac{\sigma_{\text{Inter-trial}}}{\sqrt{N}}}$	Biceps	1.0	1.9	1.5	1.7
	Triceps	0.9	2.2	1.5	1.9
	Brachioradialis	1.1	1.5	0.8	0.7
	Pronator teres	1.0	1.0	1.1	1.1
	Pronator quadratus	0.5	0.8	1.6	1.0
	Brachialis	1.2	1.2	1.0	1.9

of TD children and children with USCP.

Secondly, using this data, we compared the EMG patterns of a child with USCP before and after BTI with those of the TD group. The method successfully detected deviations of upper limb muscle activation in the child with USCP in comparison with the TD reference patterns, as well as changes following the BTI.

4.1. Variability of the EMG patterns

Interestingly, in both the TD and USCP groups, irrespective of the EMG amplitude normalization method, the ratios of inter-session to inter-trial variability were quite low (maximum of 2), meaning that the

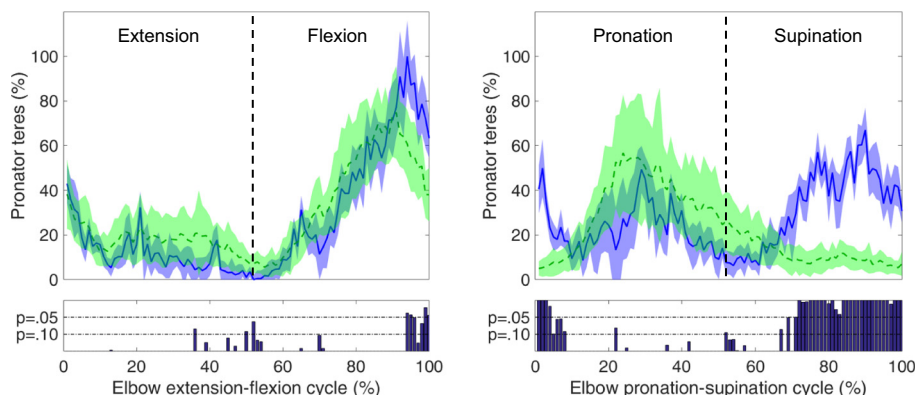


Fig. 2. Mean pronator teres EMG pattern (%) of the TD group (green dotted line) with the total standard deviation calculated using the inter-trial, inter-session, and inter-subject variability (green shaded area), and of session 1 of Child 1 (pre BTI) (solid blue line) with the total standard deviation calculated using the inter-trial and inter-session variability (blue shaded area) during elbow extension-flexion (left), and pronation-supination (right). The statistical significance (p-value) of the deviation is reported for each percent of the movement cycle along the bottom of the graph. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

inter-session variability was essentially equivalent to the inter-trial variability. These results are in accordance with those of Granata et al. (2005), who found similar inter-trial and inter-session variability in the lower limb EMG patterns of TD children during gait. Therefore, for a given subject, the upper limb EMG pattern variability calculated using the inter-trial variability was equivalent to the EMG pattern variability calculated using both the inter-trial and inter-session variabilities since the variability of the EMG patterns for an individual was calculated as:

$\sqrt{\frac{\sigma_{\text{USCP/TD}}^2 \sigma_{\text{Inter-session}}^2 + \frac{\sigma_{\text{USCP/TD}}^2 \sigma_{\text{Inter-trial}}^2}{5}}{2}}$. This demonstrated that the placement of the electrodes was consistent and that the cyclical EF and PS movements synchronized with the metronome were reliable over two separate sessions. When the EMG amplitude was not normalized, the inter-session variability to inter-trial variability ratios were higher. Since this also resulted in higher inter-subject to inter-trial variability ratios in the TD group, we recommend normalizing the amplitudes of the EMG signals.

Irrespective of the method used to normalize the EMG amplitude, variability was not higher in the USCP group than in TD group, even for non-normalized EMG patterns. These results are in agreement with those of Shuman et al. (2016), who found similar inter-trial and inter-session variability of lower limb muscle synergies in TD children and in children with CP during gait.

Normalization of EMG amplitude by the peak values during the averaged movement trials was the most appropriate method for our purposes since it yielded the lowest inter-subject/inter-trial variability ratio in the TD group, and thus had the highest capacity to detect pathological activations in children with USCP compared with the TD reference group.

4.2. Comparison of the EMG patterns

Determination of the most appropriate normalization method to reduce variability allowed the subsequent analysis of the EMG patterns of an individual child with USCP, and comparison with reference to the patterns of TD children as well as before and after BTI.

The analysis showed some similarities and some differences relative to the TD reference group, depending on the muscles and on the movements. Specifically, pronator teres activation was normal during EF, whereas during PS, its activation was higher than that of the TD group during most of the supination movement. This suggested the limitation of active supination was due to co-activation of the pronator teres. In contrast, activation of the pronator quadratus was higher than that of the TD reference group during the extension movement, but normal during PS. The higher activation of the pronator quadratus during the elbow extension movement was likely related to the elbow extension/pronation synergy exhibited during spontaneous movements. Therefore, the EMG pattern analysis identified that the main cause of the lack of active supination was co-activation of the pronator teres muscle, thus appropriate treatment could be determined. This

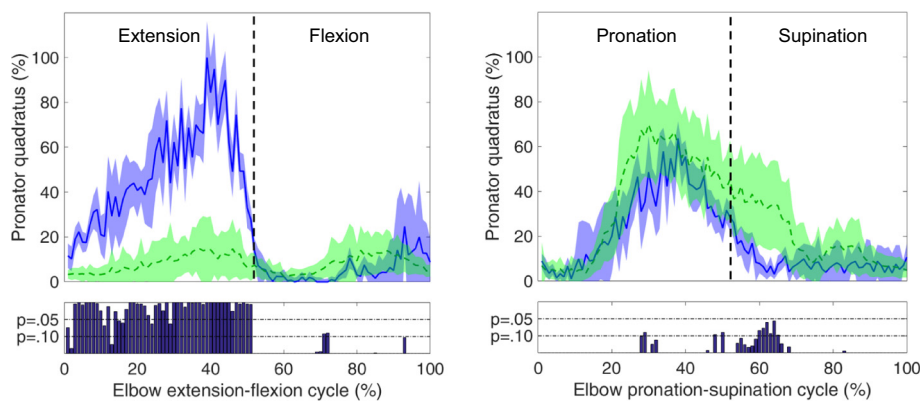


Fig. 3. Mean pronator quadratus EMG pattern (%) of the TD group (green dotted line) with the total standard deviation calculated using the inter-trial, inter-session, and inter-subject variability (green shaded area), and of session 1 of Child 1 (pre BTI) (blue full line) with the total standard deviation calculated using the inter-trial and inter-session variability (blue shaded area) during elbow extension-flexion (left) and pronation-supination (right). The statistical significance (p-value) of the deviation is reported for each percent of the movement cycle along the bottom of the graph. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

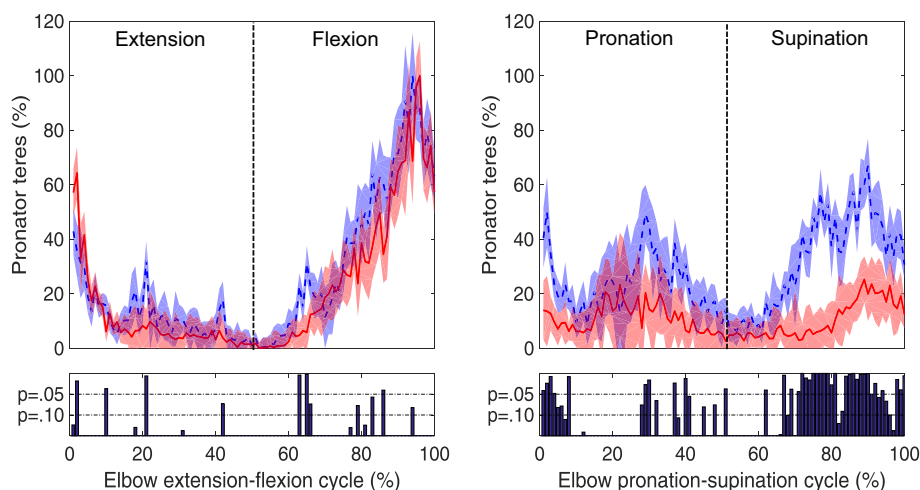


Fig. 4. Mean pronator teres EMG pattern (%) during session 1 of Child 1 (pre BTI) (blue dotted line) with its respective total standard deviation calculated with the inter-trial and inter-session variability (blue shaded area) and during session 2 of Child 1 one month after BTI of the pronator teres muscle (solid red line) with the total standard deviation calculated using the inter-trial and inter-session variability (red shaded area) during elbow extension-flexion (left) and pronation-supination (right). The statistical significance (p-value) of the deviation is reported for each percent of the movement cycle along the bottom of the graph. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

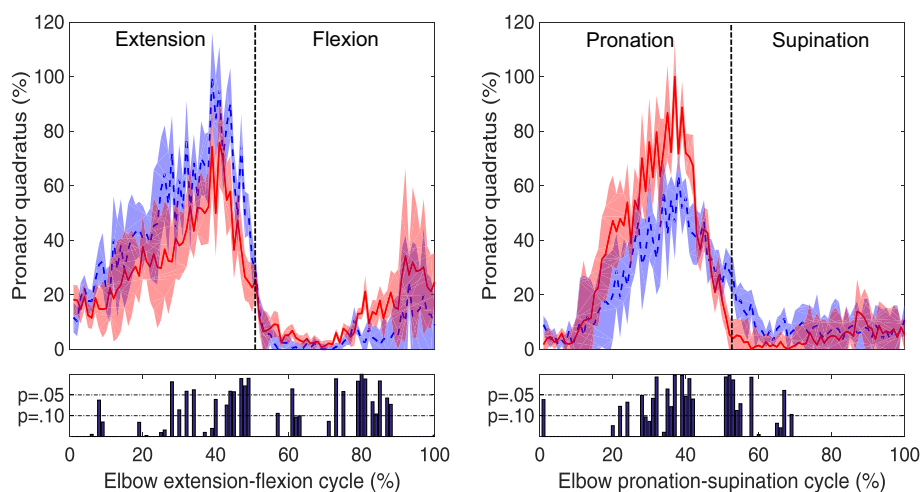


Fig. 5. Mean pronator quadratus EMG pattern (%) during session 1 of Child 1 (blue dotted line) with the total standard deviation calculated using the inter-trial and inter-session variability (blue shaded area) and during session 2 of Child 1 one month after BTI in the pronator teres muscle (solid red line) with its respective total standard deviation calculated with the inter-trial and inter-session variability (red shaded area), during elbow extension-flexion (left), and pronation-supination (right). The statistical significance (p-value) of the deviation is reported for each percent of the movement cycle along the bottom of the graph. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

highlights the benefits of using EMG pattern analysis to plan treatment. Comparison of EMG patterns before and after BTI in Child 1 with USCP showed a considerable reduction in pronator teres activation during supination, indicating that the BTI had been effective.

Therefore, upper limb surface EMG patterns allowed us to identify the differences in muscle activation during the EF and PS movements, despite the various sources of variability.

This study had some limitations. The sample of 12 participants was too small to study the effect of age on variability. Kinematic variability (Sangeux et al., 2016) and lower limb EMG variability (Tirosh et al., 2013) during gait have been shown to decrease with age in TD children,

so it is likely that age could also impact the variability of upper limb EMG patterns. Future studies should therefore evaluate variability in subgroups based on age in order to produce reference data for specific age groups. Another limitation is that the study design did not allow the estimation of either inter-assessor variability or the product of inter-assessor and inter-session variability interaction. Inter-assessor variability should be evaluated before comparing the EMG signals collected by different assessors. Also, the use of surface EMG comports the risk of electromyographic cross-talk between muscles, especially in the small forearms of children. However, the cross-talk of nearby muscles, such as the wrist, hand, and finger muscles, was limited by the choice of one-

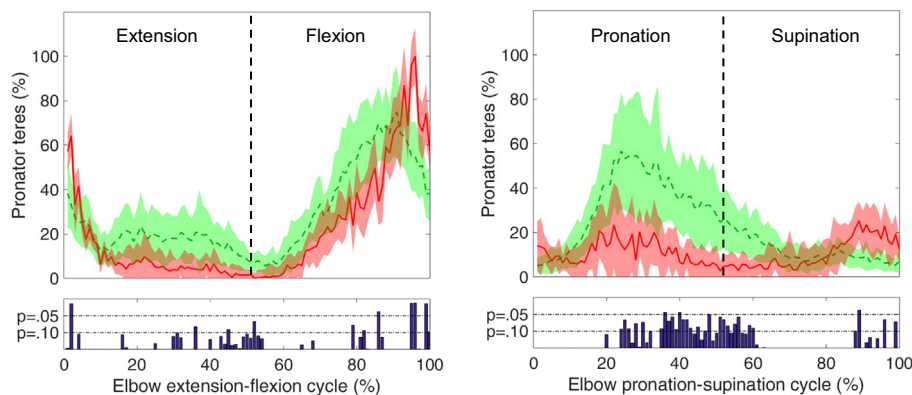


Fig. 6. Mean pronator teres EMG signals (%) of the TD group (green dotted line) with the total standard deviation calculated using the inter-trial, inter-session, and inter-subject variability (green shaded area) and of session 2 of Child 1 one month after BTI (red full line) with the total standard deviation calculated using the inter-trial and inter-session variability (red shaded area) during elbow extension-flexion (left) and pronation-supination (right). The statistical significance (p-value) of the deviation is reported for each percent of the movement cycle along the bottom of the graph. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

degree-of-freedom elbow movements. Moreover, it has been shown that electrode placement strongly impacts surface EMG patterns (Campanini et al., 2007; Hermens et al., 2000; Hug, 2011), therefore the electrodes were placed over the center of the muscle belly as recommended by (Hug, 2011). Care was also taken to ensure that the EMG electrodes did not move during the entire movement since the level of activation is not uniform over a whole muscle (Hug, 2011). Finally, the method presented here represents the inter-trial, inter-session, and inter-subject variability of EMG patterns, along with a statistical method to detect deviations in patterns from a reference group and changes following treatment. We believe that this method should be used in association with an expert reading of the EMG patterns to determine if any deviations are clinically relevant. Future research should focus on determining a rigorous quantitative criterion, for example a minimum duration of the deviation, to detect clinically relevant deviations.

5. Conclusions

Inter-session and inter-trial variability of upper limb EMG patterns during elbow movements were similar between TD children and children with USCP. Normalization of the EMG signal by the peak value during averaged movement trials produced the lowest inter-subject variability in the TD group. This allowed deviations and changes in muscle activation to be detected by the method, as demonstrated by a case study in a child with USCP whose patterns were compared with the TD group, as well as before and after treatment to reduce excessive muscle activation. This study therefore demonstrated that, despite innate variability, upper limb surface EMG pattern analysis could be used to detect clinically-relevant differences in muscle activation.

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Declarations of interest

None.

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