POLITECNICO DI TORINO Repository ISTITUZIONALE

Optimal fluxes, reaction replaceability, and response to enzymopathies in the human red blood cell

Original

Optimal fluxes, reaction replaceability, and response to enzymopathies in the human red blood cell / De Martino, A.; Granata, D.; Marinari, E.; Martelli, C.; Van Kerrebroeck, V.. - In: JOURNAL OF BIOMEDICINE AND BIOTECHNOLOGY. - ISSN 1110-7243. - 2010:(2010), pp. 1-10. [10.1155/2010/415148]

Availability: This version is available at: 11583/2976771 since: 2023-03-10T09:41:26Z

Publisher: Hindawi

Published DOI:10.1155/2010/415148

Terms of use:

This article is made available under terms and conditions as specified in the corresponding bibliographic description in the repository

Publisher copyright

(Article begins on next page)



Article

Variability of Muscular Recruitment in Hemiplegic Walking Assessed by EMG Analysis

Francesco Di Nardo ^{1,*}, Susanna Spinsante ¹, Chiara Pagliuca ¹, Angelica Poli ¹, Annachiara Strazza ¹, Valentina Agostini ², Marco Knaflitz ², and Sandro Fioretti ^{1,*}

- ¹ Department of Information Engineering, Università Politecnica delle Marche, 60100 Ancona, Italy; s.spinsante@staff.univpm.it (S.S.); chiara_pagliuca@yahoo.it (C.P.); angelicapoli93@gmail.com (A.P.); a.strazza@pm.univpm.it (A.S.)
- ² Department of Electronics and Telecommunications, Politecnico di Torino, 10129 Torino, Italy; valentina.agostini@polito.it (V.A.); marco.knaflitz@polito.it (M.K.)
- * Correspondence: f.dinardo@staff.univpm.it (F.D.N.); s.fioretti@staff.univpm.it (S.F.); Tel.: +39-071-220-4838 (F.D.N.)

Received: 26 August 2020; Accepted: 23 September 2020; Published: 25 September 2020



Abstract: Adaptive variability during walking is typical of child motor development. It has been reported that neurological disorders could affect this physiological phenomenon. The present work is designed to assess the adaptive variability of muscular recruitment during hemiplegic walking and to detect possible changes compared to control populations. In the attempt of limiting the complexity of computational procedure, the easy-to-measure coefficient of variation (CV) index is adopted to assess surface electromyography (sEMG) variability. The target population includes 34 Winters' type I and II hemiplegic children (H-group). Two further healthy populations, 34 age-matched children (C-group) and 34 young adults (A-group), are involved as controls. Results show a significant decrease (p < 0.05) of mean CV for gastrocnemius lateralis (GL) in H-group compared to both C-group (15% reduction) and A-group (35% reduction). Reductions of mean CV are detected also for tibialis anterior (TA) in H-group compared to C-group (7% reduction, p > 0.05) and A-group (15% reduction, p < 0.05). Lower CVs indicate a decreased intra-subject variability of ankle-muscle activity compared to controls. Novel contribution of the study is twofold: (1) To propose a CV-based approach for an easy-to-compute assessment of sEMG variability in hemiplegic children, useful in different experimental environments and different clinical purposes; (2) to provide a quantitative assessment of the reduction of intra-subject variability of ankle-muscle activity in mild-hemiplegic children compared to controls (children and adults), suggesting that hemiplegic children present a limited capability of adapting their muscle recruitment to the different stimuli met during walking task. This finding could be very useful in deepening the knowledge of this neurological disorder.

Keywords: surface electromyography; cerebral palsy; hemiplegia; motor disorders; gait variability; coefficient of variation

1. Introduction

Hemiplegia, often observed in children affected by cerebral palsy, is a neurological disease characterized by the fact that only half of the body is affected by the disorder. Modified selective motor control, weakness and spasticity are associated with hemiplegia, conditioning everyday activities including walking [1]. In the late 1980s, Winters et al. introduced a suitable classification of gait in hemiplegia. Based on a kinematics analysis, the authors identified four different gait patterns in the sagittal plane where four categories were discriminated, based on a progressive distal-proximal involvement of the hemiplegic leg [2]. Winters' type I patients present a hypo-activation of dorsi-flexor



muscles of the hemiplegic-leg ankle, causing drop foot during swing. Winters' type II subjects are typified by the persistence of equinism throughout the gait cycle, often related to a hyperextension of the knee during stance phase [2,3]. Winters' type I and II are the forms of hemiplegia most frequently detected in cerebral palsy; thus, many studies have focused on them [3–6].

Surface electromyography (sEMG) is an acknowledged diagnostic technique, typically used to characterize muscular activity by means of a non-invasive approach. Non-invasiveness and easiness of use, associated with the increasing availability of solutions based on sEMG, make this technique particularly valuable for the analysis of those pathologies in which walking is directly affected, as in cerebral palsy. Many studies, including our own, used an sEMG-based approach to identify the gait patterns adopted by hemiplegic children and to compare them with control children [4,6–11]. In particular, two recent studies, performed on numerous strides (hundreds) per patient, reported clear alterations of muscular-recruitment patterns in hemiplegic side: Reduced and less frequent activity during swing and a dearth of activity at loading response of tibialis anterior in type I and II; and a hyper-activation of gastrocnemius around initial contact was identified in type II only [5,6]. Overall, both studies reported significant variability in activation modality of muscles of both hemiplegic leg [5] and contralateral (non-hemiplegic) one [6].

Adaptive variability is typical of human motor development. According to some researchers, the variability in early infant movements is a key aspect of motor development [12]. Moreover, other sEMGbased studies suggest that an initial attempt of adaptation in postural behavior during sitting could be identified in four-month-old infants [13]. Then, all basic motor functions will achieve the first stages of the so-called secondary variability around the age of 18 months. Active trial-and-error experiences, specific to each subject, are typical of this stage. The basic, variable motor skill reached during the phase of primary variability keeps on developing and modifying all through the subject's life, allowing increasingly accurate and organized movements. Consequently, adult subjects master a wide movement repertoire, enabling an efficient motor solution for each specific circumstance [13]. Overall variability of human motion is associated with variability of muscle activity, quantified by EMG signals. In a preliminary study of the present group of researchers [14], sEMG-signal variability was quantified in relation to motor development, comparing adult and children populations by means of a quantitative index, the coefficient of variation (CV), previously tested on different EMG signals [14–16]. That study suggested that CV is an easy-to-measure index able to quantify sEMG variability in different experimental conditions and with different clinical purposes: In adult and pediatric populations and for both intra- and inter-subject studies. sEMG variability has been infrequently assessed in hemiplegic children and only by means of computationally expensive techniques, such as statistical gait analysis [4,6]. To the authors' best knowledge, the CV index has never been applied to quantify the variability of muscular recruitment during hemiplegic walking. Moreover, no attempts were reported in literature to provide a direct and quantitative comparison of sEMG-variability values between hemiplegic children and controls.

Thus, the present work is designed to assess the adaptive variability of muscular recruitment during hemiplegic walking and to detect possible changes in sEMG variability of hemiplegic walking compared to controls. The easy-to-measure CV index is chosen to achieve this goal, in order to propose a novel approach able to limit the complexity of computational procedures. The CV value is computed in 34 school-age hemiplegic children identified as type I and II by Winters' classification and in a large number of cycles per subject (hundreds), resulting in around 30,000 strides in total, to guarantee an adequate number of samples for variability characterization. The same index is used to describe sEMG behavior in two further populations, school-age children (34 subjects) and young adults (34 subjects), to compare and interpret results achieved in the hemiplegic population. The manuscript is organized as follows: Section 2 provides a short summary of the main indices available in the literature and used to quantify and analyze the sEMG signal variability in different scenarios, among which the CV is applied in the present study. Section 3 presents material and methods based on which the research was developed, providing details about sEMG processing, test populations and

parameters computation. Section 4 presents the experimental results that are discussed in Section 5, along with retrospection on the related state of the art. Finally, Section 6 concludes the manuscript and provides insights for future research developments.

2. Indices for sEMG Variability Analysis

The non-invasive recording of muscle electrical activity during dynamic tasks is greatly supported by sEMG, thanks to a huge collection of algorithms and techniques specifically designed to obtain and interpret the muscle activation patterns. The last ones may appear in patients with altered locomotion, and the use of sEMG in clinical gait analysis helps identifying such a condition. Despite the aforementioned advantages, and the market availability of wireless, lightweight and minimally invasive sEMG measurement equipment, such as the Myon [17] or the Freeemg [18] devices, sEMG has not witnessed a pervasive and widespread adoption in clinical assessment or rehabilitation yet. This is motivated by education barrier, i.e., understanding the features and information associated with electrical signals measured on the body may be not easy or straightforward by clinical operators [19,20]. Additional complexity is determined by the possibility to apply a huge variety of parameters, indices and figures, differently defined and computed from the measured sEMG signal samples, according to the specific muscle feature or activation pattern one is interested to observe [21]. For example, root mean square (RMS), median frequency (MF) and mean power frequencies (MPF) based on Fourier Transform [22] have been effectively used in applications dealing with the evaluation of muscular fatigue.

It is well recognized that the human motor system exhibits redundancy, so a single motor task may be performed in several different ways, leading to a similar final result [23]. Redundancy of motor repertoire in human subjects reflects the capability of the nervous system to generate different patterns of muscle activation, for the same given movement. Such a capability motivates either intra- and inter-subject variability of muscle activation, which can be captured by suitably designed indices computed on the measured sEMG signal samples. For example, indices proposed for sEMG analysis focused on aspects pertaining to running are mean, standard deviation (SD) and mean CV, as well as CV calculated over the running cycle [24]. The mean sEMG value at the denominator of the CV definition influences the value of such an index: For sensors located in those body areas where muscle activity is very weak or not present at all, the variability may be overestimated [25]. In order to overcome this limit, other metrics have been introduced, such as the variance ratio (VR) applied in gait analysis [26]. In studying intra-individual variability of sEMG in front crawl swimming, Martens et al. [27] introduced several one- and two-dimension metrics: They included both oneand two-dimension CV, VR and the coefficient of quartile variation (CQV). Corresponding general definitions are reported in Table 1. In particular, the CV of a quantity is defined as the ratio of its standard deviation to its mean, as given in Table 1. Such an index is largely used in many clinical fields, but it is not commonly applied to sEMG signals. In the present work, CV is adopted to quantify the variability of muscle rhythmic activation during walking in three different populations, namely hemiplegic children, healthy school children and young adult. Motivation for choosing the CV is threefold: (i) We aim for applying and testing this index in the evaluation of sEMG variability during walking in hemiplegic children for the first time at our best knowledge; (ii) we aim for checking the suitability of such an easy-to-compute index in reflecting different characteristics between pathological and control children and then between children and young adults, in order to promote the adoption of sEMG in clinical practice: Despite its simplicity, the index is able to satisfactorily discriminate the muscular recruitment during walking exhibited by different populations [14,28]; (iii) CV is a unit-free measure, suitable to compare normally distributed data by directly quantifying the degree of variability relative to the mean of the distributions [28]. The CV index, indeed, is not directly computed on sEMG samples, but it is derived from the standard deviation of the signal, which is by definition a direct measurement of the signal variability. These characteristics seem to make this index more suitable to the aim of the present study, respect to CQV and VR indices. CQV index, indeed, depends on mean and quartiles,

which in turn can be influenced by how they are estimated [28]. VR index, requiring a more articulated computation algorithm, is more indicated for intra-individual variability, being insensitive to mean sEMG amplitude and data smoothing applied to different waveforms [29]. Neither CQV nor VR indices include the standard deviation in their own definition (Table 1).

Index	Definition	Parameters	
One-dimension CV: it permits comparison of the variability of a data set with a larger and a smaller mean and SD	$CV = rac{\sqrt{rac{1}{k}\sum_{i=1}^k \sigma_i^2}}{rac{1}{k}\sum_{i=1}^k \left \overline{X}_i ight }$	$k =$ no. of intervals ^(*) over a cycle; $\overline{X}_i =$ mean of the sEMG values at the <i>i</i> -th interval calculated over all the cycles; $\sigma_i =$ standard deviation of the sEMG values calculated over all the cycles.	
Two-dimension CV	$CV_i = rac{\sigma_i}{\overline{X}_i}$	CV at the <i>i</i> -th interval $(*)$. Note that CV is defined as the mean value of CV _i 's over the number of intervals in a cycle (<i>k</i>).	
Variance Ratio (VR)	$VR = \frac{\sum_{i=1}^{k} \sum_{j=1}^{n} \frac{\left(X_{ij} - \overline{X}_{i}\right)^{2}}{k(n-1)}}{\sum_{i=1}^{k} \sum_{j=1}^{n} \frac{\left(X_{ij} - \overline{X}\right)^{2}}{k(n-1)}}$ where $\overline{\overline{X}} = \frac{1}{k} \sum_{i=1}^{k} \overline{X}_{i}$	$k = \text{no. of intervals}^{(*)}$ over the cycle; $n = \text{no. of cycles}$; $X_{ij} = \text{sEMG}$ value at the <i>i</i> -th interval for the <i>j</i> -th cycle, $\overline{X}_i = \text{mean of sEMG}$ values at the <i>i</i> -th interval over <i>j</i> cycles; $\overline{X} = \text{mean of sEMG}$ values.	
Coefficient of Quartile Variation (CQV)	$CQV = \frac{(Q_3 - Q_1)}{(Q_3 + Q_1)}$	$Q_1 = 25$ th percentile, $Q_3 = 75$ th percentile of the <i>n</i> sEMG values at a given interval ^(*) .	

Table 1. Different indices to quantify intra-individual sEMG signal variability (elaborated from [27]).

^(*) Definition of interval depends on the specific study target (e.g., gait analysis, swimming, walking).

As discussed in [30], sEMG-signal amplitude is typically used as a measure of relative force production and it increases with the number, size and firing rate of active motor units. When collecting sEMG, several aspects may affect the measure of sEMG amplitude and frequency, namely the depth of the active motor units, the thickness of the subcutaneous tissues, proximity to the innervation zone and tendons. As such, electrode placement plays a crucial role in sEMG signal quality. Moreover, it is acknowledged that the thickness of the subcutaneous tissue between the surface electrode and active muscles affect the measurement of electromyographic activity. The amount of excess body fat is considered as an internal noise for EMG because it increases the separation between the active muscle fibers and the detection sites [31]. In this work, sEMG signals have been collected from tibialis anterior (TA) and gastrocnemius lateralis (GL) muscles, based on acknowledged guidelines [32,33] for electrodes positioning to maximize the signal-to-noise-ratio. Moreover, obese subjects have been excluded from the study [34]. So, the potentially limiting factor of a small average sEMG value, associated with CV definition, is avoided.

3. Material and Methods

3.1. Participants

A retrospective study was performed, considering sEMG and foot–floor-contact data from 102 volunteer subjects. Volunteers were split into three different groups. H-group was composed of 34 Winters' type I and II hemiplegic children (18 males and 16 females, 6–13 years, age = 7.9 ± 3.0 years, height = 127 ± 18 cm, mass = 27.4 ± 11.0 kg), originally introduced in [4].

C-group was composed of 34 control children (18 males and 16 females, 6–11 years, age = 9.1 ± 1.1 , height = 134 ± 9 cm, mass = 32.1 ± 6.9 kg), originally introduced in [35]. A-group was composed of 34 healthy adults (18 males and 16 females, 20–30 years, age = 23.9 ± 1.5 years; height = 174 ± 10 cm; mass = 63.1 ± 12.0 kg), picked up from the populations analyzed in the Movement Analysis Laboratory of Università Politecnica delle Marche, Ancona, Italy and previously introduced in [14] and [36]. Obese subjects were not included in the study. The research was undertaken in compliance with ethical principles of Helsinki Declaration and approved by institutional expert committee. Adult participants

signed informed consent prior the beginning of the test. For children, parental consent and child assent were obtained.

3.2. Measurement Chain

Basographic and sEMG signals were acquired and synchronized by means of Step 32 multichannel recording system, (Medical Technology, Turin, Italy, resolution: 12 bit; sampling rate: 2 kHz). Basographich switches (minimum activation force = 3 N), were pasted beneath the heel, the first and the fifth metatarsal heads of each foot, for measuring foot–floor-contact signal. Single differential sEMG probes (Ag/Ag-Cl disk; electrode diameter: 0.4 cm; inter-electrode distance: 0.8 cm; differential amplifier gain: 30 dB; high-pass filter cut-off frequency: 10 Hz; input impedance: 1.5 G Ω ; CMRR > 126 dB; input referred noise: 1 Vrms) were placed bilaterally over TA and GL muscles, following acknowledged guidelines [32,33]. Then, subjects walked barefoot back and forth over the floor at preferred speed and pace for at least 2.5 min. Further details about acquisition procedure could be found in [4,35,36].

3.3. Signal Processing

Single gait cycles and the phases within each cycle were assessed from basographic signals following the procedure reported in [37]. Band-pass filtering (20–450 Hz) was applied to raw sEMG signals to remove the baseline drift associated with movement, perspiration, etc., and any DC offset. Further, sEMG signals x(t) were full-wave rectified and then smoothed computing the following RMS formula:

$$RMS = \sqrt{\frac{1}{T} \int_0^T \left| x(t) \right|^2 dt} \tag{1}$$

over a sliding window of 50 ms (100 samples). The sliding-window approach allows improving the transitory response and guarantees a better temporal resolution. An example of full-wave rectified (panel A) and RMS (panel B) signals in the same stride is reported in Figures 1 and 2, for GL and TA respectively.

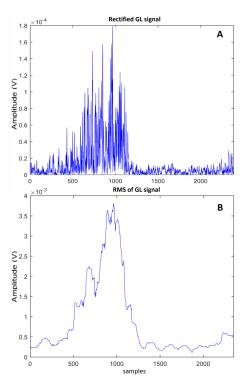


Figure 1. Rectified surface electromyography (sEMG) panel (**A**) and root mean square (RMS) of sEMG signal panel (**B**) for GL in the same representative stride during walking.

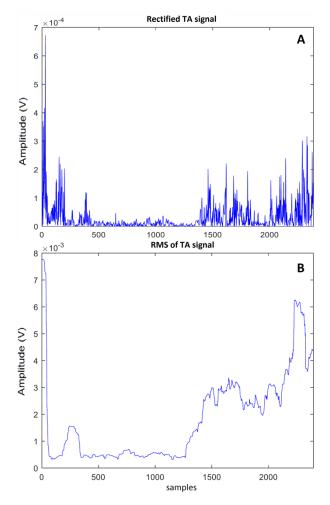


Figure 2. Rectified sEMG panel (**A**) and RMS of sEMG signal panel (**B**) for TA in the same representative stride during walking.

3.4. Variability Index

The CV index is used to measure the variability of muscles rhythmic activation during walking. According to the definition provided in Table 1, the value of this index within a cycle is computed as the ratio of the sEMG signal standard deviation (σ_i) to the mean value (\overline{X}_i) in a single *i*-th interval [15,16]:

$$CV_i = \frac{\sigma_i}{\overline{X}_i}, \ i = 1 \dots k$$
 (2)

As anticipated in Table 1, definition of interval depends on the specific study target (e.g., gait analysis, swimming, walking). The interval considered in the present study is the gait cycle, assessed from the basographic signal. After the evaluation of CV_i index in each single stride, the average over all the *k* strides of a single walking task gives the global CV. High CV values indicate a large range of variability for a muscle, characterized by periods of contraction and periods of relaxation; lower values identify a more uniform and constant muscle activity.

3.5. Statistics

The Shapiro–Wilk test was used to evaluate the hypothesis that each data vector had a normal distribution. Since all the samples resulted normally distributed, the analysis of variance (ANOVA), followed by multiple comparison test, was used to compare the three groups.

4. Results

In the present study, CV values were assessed over 102 subjects, equally split into the three populations, involving 29,042 strides in total. Figure 3 shows, for every subject of H-group, mean CV values (+SD) over all the available strides for both GL (panel A) and TA (panel B). In the same way, Figures 4 and 5 depict mean CV values (+SD) over all the available strides for both GL (panel A) and TA (panel B), for every subject of C-group and A-group, respectively. Considering 6519 strides in total (a mean value of 192 ± 71 per h-subject), mean CV values (+SD) of 0.71 ± 0.16 for GL and 0.72 ± 0.14 for TA were achieved over H-group. In a total of 9923 strides (a mean value of 292 ± 38 per c-subject), mean CV values of 0.83 ± 0.19 for GL and 0.77 ± 0.12 for TA were computed over C-group. Eventually, in a total of 12,600 strides (a mean value of 371 ± 151 per a-subject), mean CV values of 1.10 ± 0.21 for GL and 0.85 ± 0.11 for TA were obtained over A-group.

A direct comparison among average CV values over the three populations is reported in Figures 6 and 7 for GL and TA, respectively. A statistically significant reduction (p < 0.05) of mean CV is detected for GL in H-group compared to both C-group (15% reduction) and A-group (35% reduction). Moreover, the difference observed between C-group and A-group is statistically significant (25% reduction, p < 0.05, Figure 6). A significant reduction (p < 0.05) of mean CV is detected for TA in H-group compared to A-group (15% reduction). The difference observed between C-group and A-group is statistically significant as well (10% reduction, p < 0.05, Figure 7). The 7% reduction of mean CV value detected in H-group compared to C-group is not statistically significant (p > 0.05). A direct comparison between mean CV values computed in GL and in TA within the same group was also performed. In A-group, a significant higher mean CV value for GL than for TA was observed (1.10 \pm 0.21 vs. and 0.85 \pm 0.11, p < 0.05). No further significant differences were detected.

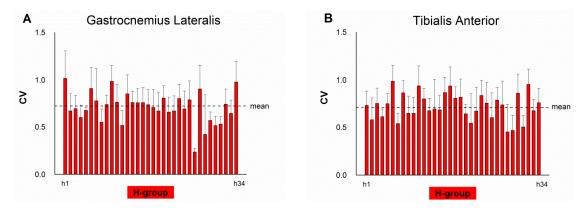


Figure 3. Average coefficient of variation (CV) values (+SD) over all the available strides for GL panel (**A**) and TA panel (**B**) in every subject of H-group. Horizontal dashed line represents the mean value over the H-group.

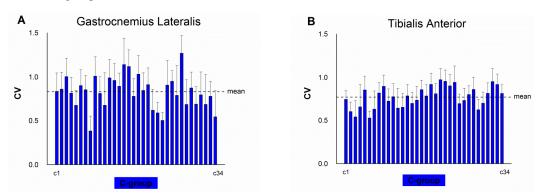


Figure 4. Average CV values (+SD) over all the available strides for GL panel (**A**) and TA panel (**B**) in every subject of C-group. Horizontal dashed line represents the mean value over the C-group.

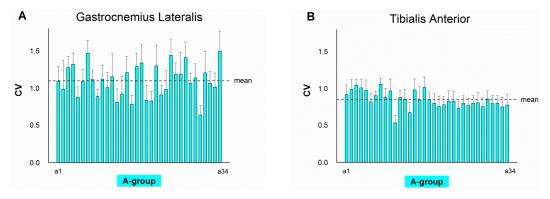


Figure 5. Average CV values (+SD) over all the available strides for GL panel (**A**) and TA panel (**B**) in every subject of A-group. Horizontal dashed line represents the mean value over the A-group.

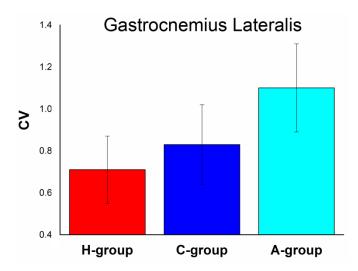


Figure 6. Average CV-GL (\pm SD) values over H-group (red bar), C-group (blue bar) and A-group (cyan bar). Each mean value is significantly different (p < 0.05) from the other two mean values.

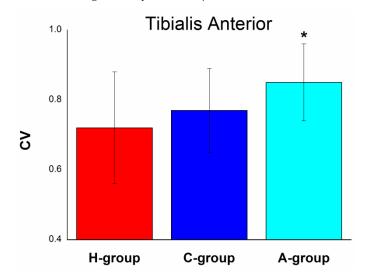


Figure 7. Average CV-TA (\pm SD) values over H-group (red bar), C-group (blue bar) and A-group (cyan bar). * means significantly different (p < 0.05) from the other two mean values.

5. Discussion

Besides the features typically extracted from sEMG signal (RMS, envelope peak, muscle activation timing, median frequency, etc.), some attempts have been recently proposed to consider sEMG-

signal variability as a suitable parameter to deepen the interpretation of muscular recruitment by neuromotor system in pathophysiology [38]. Different approaches have been used to quantify this phenomenon [15,39,40]. Nevertheless, a gold standard has not been identified yet. The CV adopted in this work has been proved to be a suitable and easy-to-measure index to assess in different clinical and experimental environments [14–16]. Thus, the goal of the present study is to assess the variability of the sEMG signal acquired over ankle muscles during hemiplegic-children walking by means of CV-index computation. Tibialis anterior and gastrocnemius lateralis are chosen because a large within-cycle variability of sEMG activity in those muscles is reported during hemiplegic-children walking [4,6]. The size of sEMG variability in hemiplegic children is quantified by a direct comparison with CV-based results achieved in a population of age-matched control children and in a further population of able-bodied young adults.

Differently from able-bodied subjects, hemiplegic children are used to hitting the ground in different ways during the same walking, such as by heel, forefoot and flat foot. It has been reported that each one of these contacts would correspond to a different EMG pattern [4]. This is particularly true for those muscles mainly involved in ankle-joint movements, such as GL and TA. Thus, a certain variability of muscle activity is expected, also in mild forms of hemiplegia, such as Winters' type I and II. Results in the present group of hemiplegic children (H-group), indeed, report high (>0.70) mean CV values (\pm SD) for GL (0.71 \pm 0.16) and for TA (0.72 \pm 0.14), confirming the above-mentioned reports and previsions. This variability may likely be ascribed to the pathophysiological alternation between sub-phases of gait in which muscles are recruited and sub-phases of gait in which muscles are silent. It could be also observed that sEMG variability is comparable in GL vs. TA, since no significant difference (p > 0.05) was detected between mean CV values of the two muscles.

To reliably quantify sEMG variability in hemiplegic children, it is necessary to compare these findings with an age-matched control population, which is represented by C-group in the present study. Alteration of walking in mild hemiplegic children has been widely reported in the literature [2-4,6,8-10]. The classification of hemiplegia proposed by Winters is based only on these differences. Winters' type I children show smaller and less frequent dorsi-flexor recruitment of the ankle in hemiplegic leg, provoking drop foot during swing. This phenomenon is further stressed in Winters' type II, causing a persistence of equinism throughout the gait cycle, often related to a knee hyperextension during stance. This obviously reflects on myoelectric activity of GL and TA. The present study was designed to check if these acknowledged alterations are also associated with a modification of sEMG variability in hemiplegic walking. Results show a decrease of mean CV value in H-group for GL (15%, p < 0.05) and TA (7%, p > 0.05), compared to C-group (Figures 6 and 7). The CV's own definition indicates that higher values of this index correspond to a more elevated variability of the phenomenon observed. Thus, lower CVs indicate a reduction of intra-subject variability of ankle-muscle activity compared to controls, suggesting that hemiplegic children present a limited capability of adapting their muscle recruitment to the different stimuli met during the walking task, also in the mildest forms of the disease (Winters' type I and II). This consideration is supported by the statistical significance only for GL. Decrease of sEMG variability for TA, indeed, is not statistically significant. This leads to reflect on the meaning of the CV index. As mentioned above, the CV index is not computed directly on sEMG samples, but it is derived from the standard deviation of the signal (Table 1), which is by definition a direct measurement of signal variability. Consequently, it is more informative in the assessment of the differences among different populations than the typical approach based on the statistical comparison among mean values. Thus, in our opinion, the information suggested by the present study could be considered reliable, certainly for dorsi-flexor muscles such as GL, but probably also for plantar-flexor ones such as TA. Moreover, these findings pave the way to further studies which will try and figure out if different results achieved on GL and TA are due to the choice of the index or to the statistical analysis or if this is going to stress a real difference in dorsi-flexor vs. plantar-flexion behavior. A further interesting finding is that H-group presents a larger normalized (to the mean value) range of CV values for both GL (0.33–1.43) and TA (0.63–1.36), compared to C-group (0.61–1.53 for GL and 0.68–1.25 for TA), indicating an increased inter-subject variability of sEMG signals during walking. This result is in line with reported studies indicating that the disorder could affect different patients in different ways [2,4], considering also that the present H-group is composed of both Winters' type I and II children.

A previous research pointed out mean CV values higher than 0.86 for GL in an adult population [16], suggesting that older age could increase sEMG signal variability. Thus, a control group of adult subjects (A-group) was also included in the present analysis, to consider the possible influence of age on CV value. In accordance with the observation reported in [16], CV values in the A-group are significantly higher for GL compared to both C-group and H-group $(1.10 \pm 0.21 \text{ vs. } 0.83 \pm 0.19 \text{ and } 0.71 \pm 0.16,$ respectively, p < 0.05, Figure 6). This is true also for TA (0.85 ± 0.11 vs. 0.77 ± 0.12 and 0.72 ± 0.14 , respectively, p < 0.05, Figure 7). Thus, an overall reduction of intra-subject variability is detected in children (hemiplegic and control), suggesting that children are used to adopting a more constant muscular recruitment during walking, with respect to adults. Physiological interpretation of this result may be ascribed to the incomplete maturation of the neuro-motor aspects of walking, acknowledged in school-age children [20]. The CV values reported here in A-group are considerably higher than those shown in [16] for adult people. This is probably due to the difference of gait protocol between the two studies. In the present study, consecutive strides during continuous long-distance gait have been considered. It is reasonable to argue that sEMG patterns may differ and variability could increase, when comparing with signals acquired in single stride during short-distance walking. Moreover, it is acknowledged that a large number of samples are needed to suitably describe the phenomenon of variability of physiological signals [39]. Therefore, the reliability of the present results is strengthened by the numerous strides analyzed here, on average nearly 300 per subject, and 30,000 in total.

The present group of researchers has recently focused its attention on the variability of muscular recruitment in children by means of sEMG analysis, a field where, to our knowledge, only few attempts were carried out. To this aim, different studies were produced, focusing on the assessment of sEMG variability in able-bodied subjects [14,35,36], proposing a new parameter for quantifying sEMG variability [39], looking for novel insights in the maturation of gait [14,36], trying to quantify the asymmetric behavior of muscle recruitment in hemiplegic-children walking [6] and attempting to find a predominant muscle activation pattern able to characterize the different classes of children hemiplegia [8]. However, most of these studies used an advanced signal a processing technique, called statistical gait analysis (SGA), which describes human walking by averaging spatial-temporal and sEMG-based characteristics over numerous strides of the same walking trial. Despite being reliable and robust, SGA is a computationally expensive technique which produces a wide range of results. Thus, the first contribution of the present study is to propose an alternative approach for a suitable assessment of sEMG variability, based on an easy-to-compute and compact index. Table 2 shows the detailed contributions of the present work with respect to each of the abovementioned studies in tabular form. Studies are reported in chronological order. While other studies [5,8,10,11,20,41] investigated muscular recruitment of lower limbs of hemiplegic children during walking, no direct assessment of sEMG variability was reported. Thus, a further contribution of the present study consists in showing the reliability of the CV index in hemiplegic-children walking, in order to also provide information on sEMG variability, besides sEMG amplitude and timing, and all the other typical parameters. A final contribution of the study is the detection of an overall reduction of intra-subject variability of ankle-muscle activity in mild-hemiplegic children compared to controls (children and young adults), suggesting that hemiplegic children present a limited capability of adapting their muscle recruitment to the different stimuli met during walking task. To our knowledge, this information is quantified here for the first time.

Study	Subject/Patient	EMG Processing	Aim	Results	Contributions of the Present Study
Agostini 2010 [35]	100 able-bodied school-age children	Statistical Gait Analysis (SGA)	To assess variability of muscular timing in numerous strides during walking	Variability was quantified by identifying 5 main activation patterns and their occurrence frequency	Quantification of intra- subject sEMG variability in numerous strides not only in control children, but also in hemiplegic children.
Agostini 2014 [42]	30 hemiplegic children—Winters' type I and II and 100 control children	Statistical Gait Analysis (SGA)	Automatic determination of sEMG patterns of hemiplegic children during gait.	Curtailed activity of tibialis anterior during terminal swing and a lack of activity at loading response in both Winters' class. Class II showed abnormal gastrocnemius activity both at initial contact and in terminal swing	Providing an index for asssessing sEMG variability in order to supply concomitant assessment of sEMG activity and variability
Agostini 2015 [4]	38 hemiplegic children—Winters' type I and II and 100 control children	Statistical Gait Analysis (SGA)	Assessment of variability of muscular timing in numerous strides within each Winters' class during walking	Variability was quantified by identifying 4–5 distinct muscle activation patterns. It cannot be defined a predominant muscle activation pattern for characterizing each specific Winters' class.	 (1) Quantification of the decreased intra-subject EMG variability in hemiplegic children compared to both control children and healthy adults (2) Assessment of EMG variability in numerous strides by means of an easy-to-compute index
Di Nardo 2017 [36]	100 able-bodied children and 33 adults	Statistical Gait Analysis (SGA)	Age- and gender-related assessment of EMG variability during walking in control subject to analyze maturation of gait	Increased EMG variability in adult but not in children female, compared to the correspondent male population.	Quantification of the reduced sEMG variability in hemiplegic children compared to both control children and able-bodied adults, providing new insights in maturation of gait and in the effect of hemiplegia on it
Di Nardo2017 [39]	20 able-bodied children and 20 adults	Statistical Gait Analysis (SGA)	To propose the occurrence frequency as a new parameter for assessing sEMG signal variability during walking.	Occurrence frequency is able to provide further information on sEMG variability, besides those supplied by classical temporal sEMG parameters.	Providing an index for asssessing sEMG variability in time domain in order to integrate the information coming from the occurrence frequency

Table 2. Detailed contributions of the present work with respect to the state of the art from the same co-authors.

Study	Subject/Patient	EMG Processing	Aim	Results	Contributions of the Present Study
Spinsante 2019 [14]	30 able-bodied children and 30 adults	CV computation	To measure variability of EMG signal in motor development and test the reliability of CV index to this aim	CV index is shown to be able to effectively discriminate pediatric motor capabilities	Extending the reliability of CV index in assessing EMG variability also to hemiplegic-children population
Di Nardo 2019 [6]	16 hemiplegic children—Winters' type I and 100 control children	Statistical Gait Analysis (SGA)	Assessment of variability of muscular timing and asymmetric behavior of muscle recruitment in hemiplegic-children walking	Increased EMG variability in the hemiplegic side due to a reduced activity in terminal swing and a lack of activity at heel-strike of ankle dorsi-flexors.	Testing the reliability in EMG variability assessment of CV index, in a large population including Winters' type I and type II hemiplegic children. This index could be used for an easy-to-compute assessment of hemiplegic asymmetry

Table 2. Cont.

6. Conclusions and Future Work

Overall, the present findings provide evidence to support the hypothesis of a decreased intra-subject variability of surface electromyography signal of ankle muscle in hemiplegic children during walking, encouraging future studies to deepen the pathophysiological reasons and modalities associate to this phenomenon. This reduction has been detected compared to both control children and able-bodied adults. Thus, it could probably be ascribed to both young age and the specific disease. Concomitantly, an increased inter-subject variability of sEMG signals was detected during hemiplegic walking, confirming that the disorder could affect different patients in different ways. Furthermore, present findings indicate that CV is a reliable index to evaluate the variability of muscle recruitment in different experimental circumstances and with different clinical goals, such as in adult and pediatric populations, in neurological disorders and for both intra- and inter-subject studies. Including the results obtained from the different indices listed in Table 1, on the set of sEMG measurements collected from the three populations, will be an interesting aspect to investigate in a future development of this study.

It has been shown that the first foot–floor contact of each hemiplegic stride could occur in different ways (with heel, forefoot and flat foot) and that each one of these contacts would correspond to a different EMG pattern. Further research developments could be focused on computing and comparing sEMG variability associated with each one of the different foot–floor contacts, trying to identify which one is more involved in the process of variability decrease. Moreover, it is acknowledged that a single gait cycle can be split in two main gait phases, stance and swing: Stance identifies the full time when the foot is on the ground; swing quantifies the period when the same foot is in the air for limb progression. Assessing sEMG variability separately for stance and swing could be one of the future developments of the present study. Since the CV approach seems to succeed in the quantification of sEMG variability in hemiplegia, further studies could involve other populations affected by neuromuscular disorders, such as cerebrovascular accident, Parkinson's disease and multiple sclerosis.

Author Contributions: Conceptualization, F.D.N. and S.S.; methodology F.D.N. and S.S.; software A.S., A.P., C.P. and S.S.; investigation F.D.N. and S.S.; validation, A.S. and C.P.; resources, S.F., V.A. and M.K.; data curation, F.D.N., A.S. and C.P.; writing—original draft preparation, F.D.N. and S.S., writing—review and editing S.F., V.A., A.P. and M.K.; visualization, F.D.N. and S.S. supervision, S.F. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

References

- Ostensjø, S.; Carlberg, E.B.; Vøllestad, N.K. Motor impairments in young children with cerebral palsy: Relationship to gross motor function and everyday activities. *Dev. Med. Child Neurol.* 2004, 46, 580–589. [CrossRef] [PubMed]
- 2. Winters, T.F.; Gage, J.R.; Hicks, R. Gait patterns in spastic hemiplegiain children and young adults. *J. Bone Joint Surg.* **1987**, *69*, 437–441. [CrossRef] [PubMed]
- McDowell, B.C.; Kerr, C.; Kelly, C.; Salazar, J.; Cosgrove, A. The validity of an existing gait classification system when applied to a representative population of children with hemiplegia. *Gait Posture* 2008, 28, 442–447. [CrossRef] [PubMed]
- 4. Agostini, V.; Nascimbeni, A.; Gaffuri, A.; Knaflitz, M. Multiple gait patterns within the same Winters class in children with hemiplegic cerebral palsy. *Clin Biomech.* **2015**, *30*, 908–914. [CrossRef]
- 5. Riad, J.; Haglund-Akerlind, Y.; Miller, F. Power generation in children with spastic hemiplegic cerebral palsy. *Gait Posture* **2008**, *27*, 641–647. [CrossRef]
- Di Nardo, F.; Strazza, A.; Mengarelli, A.; Cardarelli, S.; Tigrini, A.; Verdini, F.; Nascimbeni, A.; Agostini, V.; Knaflitz, M.; Fioretti, S. EMG-Based Characterization of Walking Asymmetry in Children with Mild Hemiplegic Cerebral Palsy. *Biosensors* 2019, *9*, 82. [CrossRef]

- Frigo, C.; Crenna, P. Multichannel SEMG in clinical gait analysis: A review and state-of-the-art. *Clin. Biomech.* 2009, 24, 236–245. [CrossRef]
- 8. Bojanic, D.M.; Petrovacki-Balj, B.D.; Jorgovanovic, N.D.; Ilic, V.R. Quantification of dynamic EMG patterns during gait in children with cerebral palsy. *J. Neurosci. Methods* **2011**, *198*, 325–331. [CrossRef]
- 9. Galli, M.; Cimolin, V.; Rigoldi, C.; Tenore, N.; Albertini, G. Gait patterns in hemiplegic children with cerebral palsy: Comparison of right and left hemiplegia. *Res. Dev. Disabil.* **2010**, *31*, 1340–1345. [CrossRef]
- 10. Patikas, D.; Wolf, S.; Döderlein, L. Electromyographic evaluation of the sound and involved side during gait of spastic hemiplegic children with cerebral palsy. *Eur. J. Neurol.* **2005**, *12*, 691–699. [CrossRef]
- Patikas, D.S.; Wolf, I.; Schuster, W.; Armbrust, P.; Dreher, T.; Döderlein, L. Electromyographic patterns in children with cerebral palsy: Do they change after surgery? *Gait Posture* 2007, *26*, 362–371. [CrossRef] [PubMed]
- 12. Turvey, M.; Fitzpatrick, P. Commentary: Development of Perception-Action Systems and General Principles of Pattern Formation. *Child Dev.* **1993**, *64*, 1175–1190. [CrossRef]
- Hadders-Algra, M. Variation and Variability: Key Words in Human Motor Development. *Phys. Ther.* 2010, 90, 1823–1837. [CrossRef] [PubMed]
- Spinsante, S.; Di Nardo, F.; Strazza, A.; Verdini, F.; Poli, A. A Simple sEMG-Based Measure of Muscular Recruitment Variability during Pediatric Walking. In Proceedings of the 2019 IEEE International Symposium on Medical Measurements and Applications (MeMeA), Istanbul, Turkey, 26–28 June 2019; pp. 1–6.
- 15. Anders, C.; Wagner, H.; Puta, C.; Grassme, R.; Petrovitch, A.; Scholle, H.C. Trunk muscle activation patterns during walking at different speeds. *J. Electromyogr. Kinesiol.* **2007**, *17*, 245–252. [CrossRef] [PubMed]
- Bailey, C.; Corona, F.; Pilloni, G.; Porta, M.; Fastame, M.; Hitchcott, P.; Penna, M.; Pau, M.; Côté, J. Sex-dependent and sex-independent muscle activation patterns in adult gait as a function of age. *Exp. Gerontol.* 2018, 110, 1–8. [CrossRef]
- 17. Available online: https://www.myon.ch/ (accessed on 7 August 2020).
- 18. Available online: https://www.btsbioengineering.com/products/freeemg-surface-emg-semg/ (accessed on 7 August 2020).
- 19. Agostini, V.; Ghislieri, M.; Rosati, S.; Balestra, G.; Knaflitz, M. Surface electromyography applied to gait analysis: How to improve its impact in clinics? *Front Neurol.* **2020**. [CrossRef]
- 20. Campanini, I.; Disselhorst-Klug, C.; Rymer, W.Z.; Merletti, R. Surface EMG in Clinical Assessment and Neurorehabilitation: Barriers limiting its use. *Front Neurol.* **2020**. [CrossRef]
- 21. Donaldson, S.; Donaldson, M.; Snelling, L. SEMG Evaluations: An Overview. *Appl. Psychophysiol Biofeedback* 2003, *28*, 121–127. [CrossRef]
- 22. Hermens, H.; Bruggen, T.; Baten, C.; Rutten, W.; Boom, H. The median frequency of the surface EMG power spectrum in relation to motor unit firing and action potential properties. *J. Electromyogr. Kinesiol.* **1992**, *2*, 15–25. [CrossRef]
- 23. Davies, B.T. A review of "The Co-ordination and Regulation of Movements" By N. Bernstein. *Ergonomics* **1968**, *11*, 95–97. [CrossRef]
- 24. Guidetti, L.; Rivellini, G.; Figura, F. EMG patterns during running: Intra- and inter-individual variability. *J. Electromyogr. Kinesiol.* **1996**, *6*, 37–48. [CrossRef]
- 25. Hug, F.; Drouet, J.M.; Champoux, Y.; Couturier, A.; Dorel, S. Interindividual variability of electromyographic patterns and pedal force profiles in trained cyclists. *Eur. J. Appl. Physiol.* **2008**, *104*, 667–678. [CrossRef] [PubMed]
- 26. Hershler, C.; Milner, M. An optimality criterion for processing electromyographic (EMG) signals relating to human locomotion. *IEEE Trans. Biomed. Eng.* **1978**, *25*, 413–420. [CrossRef] [PubMed]
- 27. Martens, J.; Daly, D.; Deschamps, K.; Fernandes, R.J.P.; Staes, F. Intra-Individual Variability of Surface Electromyography in Front Crawl Swimming. *PLoS ONE* **2015**, *10*, e0144998. [CrossRef]
- Raydonal, O.; Marmolejo-Ramos, F. Performance of Some Estimators of Relative Variability. *Front. Appl. Math. Stat.* 2019, 5, 43. [CrossRef]
- 29. Gabel, R.H.; Brand, R.A. The effects of signal conditioning on the statistical analyses of gait EMG. *Electroencephalogr. Clin. Neurophysiol.* **1994**, *93*, 188–201. [CrossRef]
- Kendell, C.; Lemaire, E.D.; Losier, Y.; Wilson, A.; Chan, A.; Hudgins, B. A novel approach to surface electromyography: An exploratory study of electrode-pair selection based on signal characteristics. *J. NeuroEng. Rehabil.* 2012, 9, 24. [CrossRef]

- 31. Hemingway, M.A.; Biedermann, H.J.; Inglis, J. Electromyographic recordings of paraspinal muscles: Variations related to subcutaneous tissue thickness. *Biofeedback Self Regul.* **1995**, *20*, 39–49. [CrossRef]
- 32. Agostini, V.; Nascimbeni, A.; Gaffuri, A.; Imazio, P.; Benedetti, M.; Knaflitz, M. Normative EMG activation patterns of school-age children during gait. *Gait Posture* **2010**, *32*, 285–289. [CrossRef]
- 33. Di Nardo, F.; Laureati, G.; Strazza, A.; Mengarelli, A.; Burattini, L.; Agostini, V.; Nascimbeni, A.; Knaflitz, M.; Fioretti, S. Is child walking conditioned by gender? surface emg patterns in female and male children. *Gait Posture* **2017**, *53*, 254–259. [CrossRef]
- 34. Available online: https://www.who.int/growthref/who2007_bmi_for_age/en/ (accessed on 17 September 2020).
- 35. Winter, D.A. Biomechanics and Motor Control of Human Movement; John Wiley & Sons: Hoboken, NJ, USA, 2009.
- 36. Hermens, H.J.; Freriks, B.; Merletti, R. European recommendations for surface electromyography, SENIAM. Enschede (NL). *Roessingh Res. Dev.* **1999**, *8*, 13–54.
- Agostini, V.; Balestra, G.; Knaflitz, M. Segmentation and classification of gait cycles. *IEEE Trans. Neural. Syst. Rehabil. Eng.* 2014, 22, 946–952. [CrossRef] [PubMed]
- 38. Lord, S.; Howe, T.; Greenland, J.; Simpson, L.; Rochester, L. Gait variability in older adults: A structured review of testing protocol and clinimetric properties. *Gait Posture* **2011**, *34*, 443–450. [CrossRef] [PubMed]
- Di Nardo, F.; Mengarelli, A.; Strazza, A.; Agostini, V.; Knaflitz, M.; Burattini, L.; Fioretti, S. A new parameter for quantifying the variability of surface electromyographic signals during gait: The occurrence frequency. *J. Electromyogr. Kinesiol.* 2017, *36*, 25–33. [CrossRef] [PubMed]
- 40. Tirosh, O.; Sangeux, M.; Wong, M.; Thomason, P.; Graham, H.K. Walking speed effects on the lower limb electromyographic variability of healthy children aged 7–16 years. *J. Electromyogr. Kinesiol.* **2013**, *23*, 1451–1459. [CrossRef]
- 41. Romkes, J.; Brunner, R. An electromyographic analysis of obligatory (hemiplegic cerebral palsy) and voluntary (normal) unilateral toe-walking. *Gait Posture* **2007**, *26*, 577–586. [CrossRef]
- 42. Agostini, V.; Knaflitz, M.; Nascimberi, A.; Gaffuri, A. Gait measurements in hemiplegic children: An automatic analysis of foot–floor contact sequences and electromyographic patterns. In Proceedings of the 2014 IEEE International Symposium on Medical Measurements and Applications (MeMeA), Lisbon, Portugal, 11–12 June 2014.



© 2020 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).