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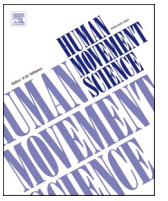

Phasic and tonic muscle synergies are different in number, structure and sparseness

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ABSTRACT

In the last two decades, muscle synergies analysis has been commonly used to assess the neurophysiological mechanisms underlying human motor control. Several synergy models and algorithms have been employed for processing the electromyographic (EMG) signal, and it has been shown that the coordination of motor control is characterized by the presence of phasic (movement-related) and tonic (anti-gravity and related to co-contraction) EMG components. Neural substrates indicate that phasic and tonic components have non-homogeneous origin; however, it is still unclear if these components are generated by the same set of synergies or by distinct synergies. This study aims at testing whether phasic and tonic components are generated by distinct phasic and tonic synergies or by the same set of synergies with phasic and tonic activation coefficients. The study also aims at characterizing the differences between the phasic and the tonic synergies. Using a comprehensive mapping of upper-limb point-to-point movements, synergies were extracted from phasic and tonic EMG signal separately, estimating the tonic components with a linear ramp model. The goodness of reconstruction (R^2) as a function of the number of synergies was compared, and sets of synergies extracted from each dataset at three R^2 threshold levels (0.80, 0.85, 0.90) were retained for further analysis. Then, shared, phasic-specific, and tonic-specific synergies were extracted from the two datasets concatenated. The dimensionality of the synergies shared between the phasic and the tonic datasets was estimated with a bootstrap procedure based on the evaluation of the distribution of principal angles between the subspaces spanned by phasic and tonic synergies due to noise. We found only few shared synergies, indicating that phasic and tonic synergies have in general different structures. To compare consistent differences in synergy composition, shared, phasic-specific, and tonic-specific synergies were clustered separately. Phasic-specific clusters were more numerous than tonic-specific ones, suggesting that they were more differentiated among subjects. The structure of phasic clusters and the higher sparseness indicated that phasic synergies capture specific muscle activation patterns related to the movement while tonic synergies show co-contraction of multiple muscles for joint stabilization and holding postures. These results suggest that in many

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scenarios phasic and tonic synergies should be extracted separately, especially when performing muscle synergy analysis in patients with abnormal tonic activity and for tuning devices with gravity support.

1. Introduction

The control of human movement takes into account the complexity of the kinematics, dynamics and biomechanics of the musculoskeletal system, integrating visual and proprioceptive information about initial and final state of the limb and activating many muscles that act on several joints (D'Avella & Lacquaniti, 2013). Therefore, it has been hypothesized that the central nervous system (CNS) simplifies the planning and production of movement through a combination of a limited number of spatial and/or temporal modules (Bizzi, Cheung, D'Avella, Saltiel, & Tresch, 2008) often referred to as muscle synergies.

Muscle synergies are often used to examine upper limb movements and to explain how the CNS controls multi-joint movements. Previous work showed that muscle patterns are characterized by the presence of phasic and tonic components in the electromyographic (EMG) signals. EMG waveforms generated during reaching movements are related to a combination of dynamic and antigravity torques (Flanders, 1991). The latter contribute to maintaining a postural configuration by counteracting the gravity force and do not scale in magnitude with movement speed, while the former are related to the execution of the movement and they scale with movement speed (Flanders & Herrmann, 1992). Therefore, in the EMG signal, the phasic waveforms are the components related to accelerating and decelerating the joints, while the tonic components are responsible for balancing gravity and stabilizing the movement in presence of perturbations (D'Avella & Lacquaniti, 2013). However, how the CNS controls these two components is not clear and it is still debated.

Some studies hypothesized that phasic and tonic components may be related to different motor control strategies. The phasic activity may be controlled by a predictive scheme while the tonic component may be related to an impedance control mechanism (Yadav & Sainburg, 2011) or to counterbalancing gravity. Other studies hypothesized that also tonic activity is controlled by a predictive scheme (Franklin et al., 2008). Furthermore, Albert et al. (2020) hypothesized that motor commands are generated in the motor cortex and they are integrated in a subcortical postural controller. The separation of the two circuitries may be justified by the fact that the tonic controller involves neural circuitry activated to hold static position or loads - maintaining a constant sensory state, while the phasic controller changes its sensory state to follow the motion of the body (Ivanenko & Gurfinkel, 2018). Phasic and tonic muscle activities are related to the dynamic and postural components of joint torques that are scaled differently in speed and time (Hollerbach & Flash, 1982). Therefore, it is reasonable to hypothesize that different torque components may be generated by different muscle synergies, with different numerosity and specific composition. However, it has never been tested whether phasic and tonic EMG components are generated by modulating in time the same set of synergies or by distinct phasic and tonic synergies. In fact, most of the studies implicitly assume that phasic and tonic synergies can be extracted together without questioning whether phasic and tonic synergies are available to the same neural controller or not. A very common approach in muscle synergies analysis applies the non-negative matrix factorization (NMF) to the whole EMG signal. Such procedure does not guarantee that phasic and tonic synergies can be identified; in general, applying NMF to the whole EMG signals may lead to the extraction of "hybrid" synergies that incorporate a mixture of phasic and tonic components. Given that, a specific procedure that separates phasic and tonic components is needed to verify if and to what extent phasic and tonic EMG components share the same synergies or require specific synergies.

Very few studies on muscle synergies have separated phasic and tonic components of the EMG signal (D'Avella, Portone, Fernandez, & Lacquaniti, 2006; Russo, D'Andola, Portone, Lacquaniti, & D'Avella, 2014; Scano et al., 2019; Zhao, Zhang, Wen, & Scano, 2021). In those studies, the tonic component of the EMG signal is estimated with a linear ramp model. The ramp is computed "connecting" the baseline magnitude of the EMG activity before and after the movement related to a static posture (D'Avella et al., 2006). Then, the ramp is subtracted from the EMG signal to obtain the phasic EMG signal underlying movement. Mira et al. (Mira, Molinari Tosatti, Sacco, & Scano, 2021) showed that, when movements were performed at natural speed, the integral of the phasic activity was lower than the tonic one in almost all the movement directions, and phasic components were clearly detectable in the EMG signal of specific muscles depending on the direction. Moreover, d'Avella et al. (D'Avella, Fernandez, Portone, & Lacquaniti, 2008) found that tonic synergies are modulated in amplitude with direction of the movement, while phasic synergies are modulated in both amplitude and timing with direction and speed. In this view, direction of movements seems to be fundamental in the modulation of both phasic and tonic activities. However, the whole upper limb workspace has been rarely investigated for characterizing the EMG components, including multiple plane such as frontal and upper planes (Mira et al., 2021; Scano et al., 2019).

The aim of this study is to test whether the CNS generates phasic and tonic EMG components by modulating in time the same set of synergies or by combining specific phasic and tonic synergies. To this end the differences between the subspaces spanned by synergies extracted separately from phasic and tonic components have been examined. First, an upper limb dataset containing multi-directional reaching movements in frontal and upward workspaces was used to elicit proximal muscle activations with large variability in both phasic and tonic EMG components of shoulder and elbow muscles. Such movements explored directions typically found in rehabilitation and industrial scenarios. Second, synergies were extracted from phasic EMG signals and tonic EMG signals separately and then synergies were compared at three different levels of reconstruction quality (R^2 threshold of 0.80, 0.85, 0.90). Since the choice of the optimal number of synergies is not univocal and different criteria are used in literature (Ranaldi, De Marchis, Severini, & Conforto, 2021), we provided a multiresolution analysis to guarantee that our results are valid regardless of the criteria chosen to select the number of synergies. Then, we employed a novel approach inspired by the procedure used by Sylos-Labini et al. (2020) to estimate the

dimensionality of the synergies shared between the phasic and the tonic dataset. A bootstrap procedure was used to evaluate the distributions of the principal angles between the subspaces spanned by phasic and tonic synergies due to the noise and to quantify the significative differences between the two subspaces. Then, shared, phasic-specific and tonic-specific synergies were extracted from the combined phasic and tonic datasets. Synergies of the same type (shared, phasic-specific and tonic-specific) were clustered with the k-means clustering algorithm. Clusters of different types were compared based on their number to quantify the modularity. Intra-cluster similarity was used to evaluate synergies variability and the consistency of each cluster. Finally, cluster sparseness identified the differences between synergies structures.

2. Materials & methods

2.1. Dataset description and preprocessing

An overview of the study is shown in Fig. 1. We considered a dataset representing proximal upper-limb coordination in multi-directional reaching movements. The dataset was originally presented in Scano et al. (Scano et al., 2019) and contains EMG signals of 16 healthy participants performing point-to-point movements towards 8 main cardinal directions placed on a circular target (NE, E, SE, S, SW, W, NW, N) and towards the central point O. In Scano et al. (Scano et al., 2019), the set of targets was positioned in five different orientations. In this study, a subset of two orientations was considered for the analysis: target positioned in the frontal plane (frontal) and in the horizontal upwards plane (up) with respect to the subject. These orientations were chosen as they show high variability of both phasic and tonic EMG activity and they represent typical movements in practical applications such as rehabilitation and industrial scenarios. Each group of movements was repeated ten times. EMG signal from sixteen muscles of the right upper limb were recorded according to the SENIAM guidelines (Hermens, Freriks, Disselhorst-Klug, & Rau, 2000): Erector Spinae (ES), Teres

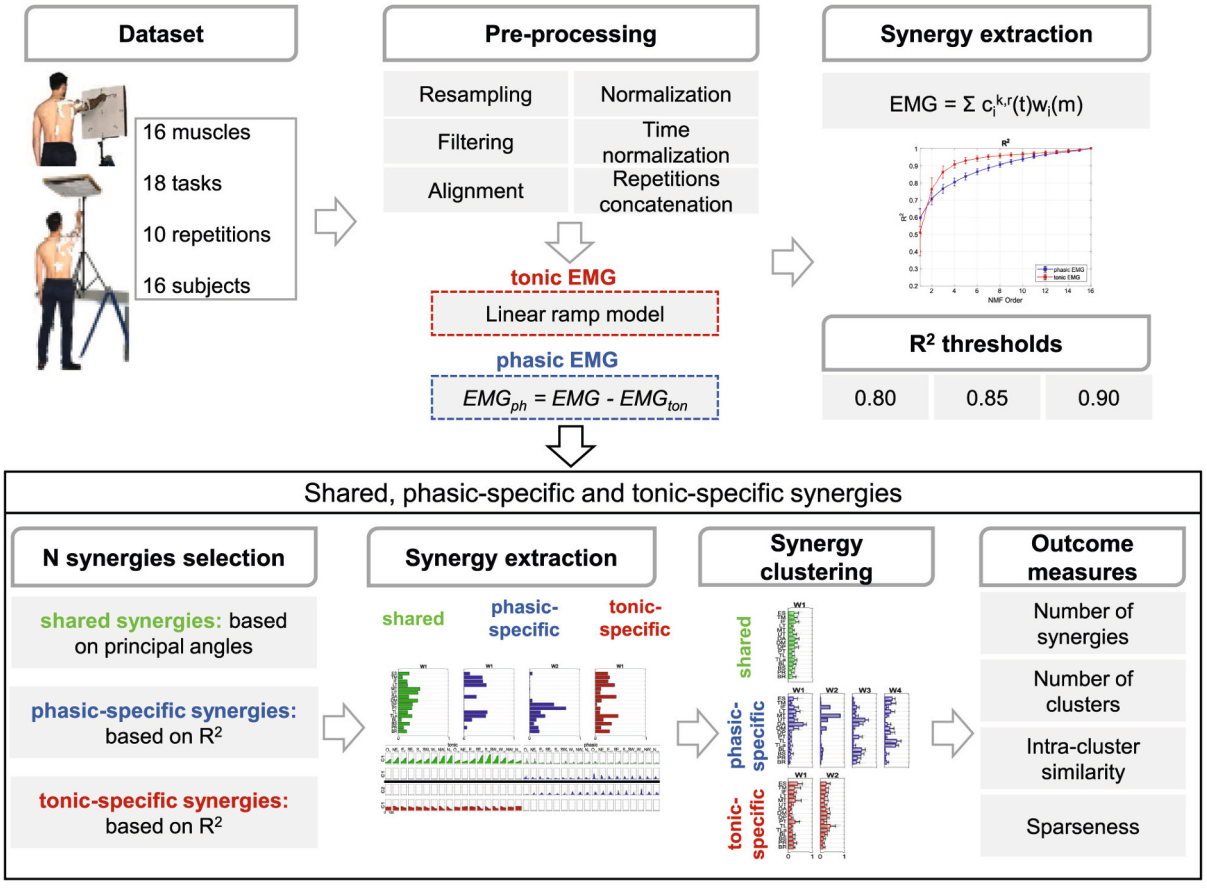

Fig. 1. Schematic of the workflow for the analysis. EMG signals are pre-processed and tonic EMG (modelled with a linear ramp) and phasic EMG components are computed. For both datasets, the reconstruction R^2 is computed and synergies are extracted at three R^2 thresholds. Shared, phasic-specific, and tonic-specific synergies are extracted from the combined phasic and tonic dataset and clustered. Number of synergies, number of clusters, intra-cluster similarity, and sparseness are compared between shared, phasic-specific, and tonic-specific synergies to characterize their structure.

Major (TM), Infraspinatus (IF), Lower Trapezius (LT), Middle Trapezius (MT), Upper Trapezius (UT), Deltoid Anterior (DA), Deltoid Middle (DM), Deltoid Posterior (DP), Pectoralis (PT), Triceps Long Head (TL), Triceps Lateral Head (TLa), Biceps Long Head (BL), Biceps Short Head (BS), Pronator Teres (PR) and Brachioradialis (BR). EMG data were filtered and aligned as described in details in Scano et al. (Scano et al., 2019). In each movement phase, the EMG signal was resampled at 100 time points and normalized by the maximum value of filtered EMG computed over all the tasks for each channel (Scano et al., 2019). Then, the tonic EMG was modelled with a linear ramp model (D'Avella et al., 2006), that considers a constant activation level of the muscle before and after the movement and linear ramp between the two constant activation levels during the movement. The initial and final activity levels of each muscle were estimated by averaging the EMG in the 200 ms before the onset of the movement and in the 200 ms after the end of the movement. The phasic EMG dataset was computed removing the tonic component from the EMG signal as follows:

$$EMG_{phasic} = EMG - EMG_{tonic}$$

and the negative values of the phasic EMG (as they appeared limited in magnitude with respect to positive values) were set to zero as in previous approaches (D'Avella et al., 2006; Scano et al., 2019). Finally, the EMG signals of each repetition were concatenated in both phasic and tonic datasets.

2.2. Extraction of synergies from phasic and tonic datasets separately

First, we extracted synergies from phasic and tonic datasets separately. The synergy extraction methodology decomposes the EMG signals as the product of a time-invariant synergy vectors and time-varying activation coefficients. For each subject, EMG signals were rearranged in two data matrices (phasic and tonic) with M rows and $K \bullet T \bullet R$ columns, where M is the number of muscles and K is the number of tasks sampled with T samples each and R is the number of repetitions. In this study, $K = 18$, $T = 100$, $R = 10$, $M = 16$. Thus, input data was a 16×18000 matrix. The non-negative matrix factorization (NMF) iterative algorithm based on multiplicative updates (Lee & Seung, 1999) was used to extract synergies by decomposing the EMG signals as:

$$EMG(t,k,r,m) = \sum_{i=1}^{S} c_i^{k,r}(t)\, w_i(m)$$

where $\boldsymbol{w_i}$ are the time-invariant (or spatial) synergy vectors and $\boldsymbol{c_i}$ the time-varying scalar activation coefficients for each synergy ($i = 1 \ldots S$), and $EMG(t,k,r,m)$ the activity of muscle m at time t of repetition r in task k. Synergy extraction leads to the identification of S synergies (each a column vector with 16 components) and $S \bullet 180$ time-varying coefficients (100 samples each). Each spatial synergy was normalized by the Euclidean norm of that synergy and the temporal coefficients were normalized by the reciprocal of the norm. The goodness of reconstruction was evaluated with an R^2 value defined as $1 - \frac{SSE}{SST}$ where SSE is the sum of the squared residuals and SST is the sum of the squared differences with the mean EMG vector (D'Avella et al., 2006), computed on the EMG data (containing both phasic and tonic components) in order to allow the comparison between phasic and tonic datasets. Synergies were extracted from order 1 to order 16 (number of muscles) and the algorithm was applied 50 times starting from different random initial conditions in order to avoid local minima. The repetition with the highest R^2 was chosen as the representative of that order. The number of extracted synergies, which may vary across participants, considered for further analyses was selected as the minimum number necessary to achieve a given R^2 value. As such threshold is arbitrary, we considered three R^2 levels – 0.80, 0.85, and 0.90 – commonly adopted in literature to span across multiple realistic conditions (Pale, Atzori, Müller, & Scano, 2020). This multiresolution analysis guarantees that our analysis is valid regardless of a specific choice of the criteria for the selection of the number of synergies.

2.3. Extraction of shared, phasic-specific, and tonic-specific synergies simultaneously

In order to analyze the phasic and tonic synergy structures, we extracted synergies shared between the two datasets and synergies specific of each dataset (phasic-specific and tonic-specific), following a novel procedure that extends those adopted in previous studies (Cheung, D'Avella, Tresch, & Bizzi, 2005; D'Avella & Bizzi, 2005; Sylos-Labini et al., 2020). In the "shared and specific" formulation of the NMF algorithm, information from two datasets is used simultaneously in order to identify synergies common to both datasets (shared) and synergies specific to each dataset. Therefore, synergies were extracted from a data matrix constructed by concatenating the EMG from the tonic and phasic datasets using the NMF algorithm (Lee & Seung, 1999). At the first iteration of the algorithm, the temporal coefficients of phasic-specific synergies were initialized to zero for all the segments of the signal belonging to the tonic dataset, and the temporal coefficients of the tonic-specific were initialized to zero for all the segments of the signal belonging to the phasic dataset. These coefficients were bound to zero throughout all the iterations thanks to the multiplicative update rule used in the algorithm to update W and c. For the extraction of shared synergies, instead, none of the temporal coefficients were initialized to zero, since they are common to both datasets. For each R^2 threshold, the appropriate number of shared, phasic-specific, and tonic-specific were extracted.

2.3.1. Determining the number of shared synergies

The number of shared synergies was determined as the number of shared dimensions of the subspaces spanned by the phasic and the tonic synergies extracted from each dataset separately. Such number was determined with a bootstrap procedure similar to the one employed by Sylos-Labini et al. (Sylos-Labini et al., 2020). The aim of the procedure was to determine how much two synergy sets

extracted from two different datasets (phasic and tonic) would differ because of noise rather than because of structural differences in the datasets. This difference was computed as non-zero principal angles (Golub, 1996) between the subspaces spanned by the two synergy sets. Only the dimensions with principal angles significantly larger than the distribution due to noise were identified as non-shared. To estimate such distribution, we first constructed a bootstrap distribution of principal angles between synergies extracted from random subsets of repetitions of each dataset. Differences in these synergies would represent the effect of noise generating unstructured variations in each repetition. We then constructed combined principal angle distributions between synergy sets extracted from the two datasets. We assumed that each synergy set differed from a common subspace only because of noise, according to the principal angle bootstrap distributions computed for each subset. Specifically, for each subject, and for each phasic and tonic dataset, ten repetitions of each movement phase were randomly divided in two subsets of five repetitions each and concatenated. The same number of synergies obtained with the phasic and tonic dataset were extracted from the two subsets. The principal angles between synergies extracted from the two subsets were computed using singular value decomposition (Golub, 1996). The distributions of principal angles obtained from this bootstrap procedure for phasic and tonic datasets were used to construct randomly two new subspaces (phasic and tonic) by rotating each subspace from a common base according to random samples of the principal angles. Successively, the principal angles between the two new subspaces were computed (combined principal angles). The bootstrap procedure was repeated 100 times (for each condition for each subject) and a distribution of combined principal angles was constructed for each condition and for each subject. The angle representing the 95th percentile (θ_{95}) of the distribution of each principal angle was computed.

The principal angles between phasic and tonic synergies extracted from the real phasic and tonic datasets were computed and each angle was compared to the corresponding θ_{95} of the combined principal angle distribution. The number of shared synergies was defined as the number of principal angles that were lower than the corresponding θ_{95} computed in the bootstrap procedure.

2.3.2. Determining the number of specific synergies

The number of specific synergies was empirically estimated in order to achieve a given R^2 level. Synergies were first extracted from the phasic and tonic datasets concatenated using the NMF based on the multiplicative update algorithm setting the number of shared synergies and no specific synergies. Then, the reconstruction R^2 on the phasic dataset due only to shared synergies was computed. Phasic-specific synergies were iteratively added until the requested R^2 threshold on the phasic dataset was achieved. The same procedure was done for defining the number of tonic-specific synergies. The reconstruction R^2 on the tonic dataset due only to shared synergies was computed. Tonic-specific synergies were iteratively added until the requested R^2 threshold on the tonic dataset was achieved. Finally, shared, phasic-specific and tonic-specific synergies were extracted together from the phasic and tonic datasets concatenated with the number of synergies previously defined.

2.4. Synergy clustering

Since the order of extraction and synergy composition may differ across participants, in order to identify synergistic patterns characterizing all subjects, synergies extracted from all participants were grouped with a cluster analysis. The clustering procedure was done separately for shared, phasic-specific, and tonic-specific synergies. We used the k-means clustering algorithm (Hartigan, 1975) using as inputs to the algorithm the matrix containing the set of muscle synergies extracted from all the participants and the number of clusters. The algorithm gave as output the synergies grouped in each cluster and the centroids (mean synergy for each of the clusters). The clustering algorithm was repeated 200 times with new initial centroids (chosen uniformly at random as in (Arthur & Vassilvitskii, 2007)) with the same number of clusters and we chose the solution that minimized the sum of Euclidean distances of each synergy with respect to the centroids.

The optimal number of clusters was determined based on the cosine similarity within the same cluster. The number of clusters was iteratively increased until the mean similarity was higher than 0.70 in each cluster in order to guarantee a limited number of clusters and a good intra-cluster similarity level (Huang, Chen, Liang, Cheng, & Xiong, 2022; Torres-Oviedo & Ting, 2010).

2.5. Outcome measures and statistics

The goodness of the reconstruction was quantified by a R^2 value for each dataset (phasic and tonic) and a linear mixed-effects model (McLean, Sanders, & Stroup, 1991) was fitted in order to assess the differences between the datasets. First, each dataset was tested for normality using the Kolmogorov-Smirnov test and the R^2 was modelled as follows:

$$R^2 \sim 1 + order^{*}dataset + (1|subject)$$

where dataset (*phasic EMG/tonic EMG*) and NMF order (number of extracted synergies) were fixed effects with interaction, NMF order was a categorical variable, and subjects were included as random effects on intercept. The level of significance (α) was set 0.05.

For each of the three reconstruction R^2 thresholds, synergies were extracted and the mean and the standard deviation of each order of factorization was computed to compare the R^2 achieved with the two datasets. The number of synergies allows to quantify the modularity of the motor control for the phasic and tonic activity separately. The mean orders of factorization of the two datasets were tested for normality and compared with a *t*-test at each reconstruction R^2 threshold, setting $\alpha = 0.05$.

The mean number of shared, phasic-specific and tonic-specific synergies were computed and tested for normality in order to be compared with one-way ANOVA tests at each reconstruction R^2 threshold. A post-hoc test was used to identify the differences between

the number of synergies, with $\alpha = 0.05$. After estimating their numerosity, shared, phasic-specific and tonic-specific synergies were extracted and clustered with the k-means algorithm. Clusters are a synthetic representation of the population, and the number of clusters quantifies their modularity. To evaluate the variability across participants and the consistency of each cluster, the intra-cluster similarity was computed as a cosine angle, comparing all pairs of synergies in a cluster. Finally, the sparseness of the mean synergy of each cluster W was computed as follows (Hoyer, 2004), in order to assess the composition of the mean synergies obtained and to evaluate the distribution of activations across muscles:

$$sparseness(W) = \frac{\sqrt{n} - \frac{\|W\|_1}{\|W\|_2}}{\sqrt{n} - 1}$$

Where n is the dimensionality of W and $\|W\|_1$ is the L_1 norm of W and $\|W\|_2$ is the norm L_2 of W.

3. Results

3.1. Reconstruction R^2 and order of factorization

In Fig. 2, the comparison of the reconstruction R^2 for the phasic and tonic datasets is shown.

The R^2 of the phasic dataset started at 0.60, while the R^2 of the tonic dataset started at 0.51. Between order 1 and 2, the two curves crossed each other and the tonic dataset reached a higher reconstruction quality. The linear mixed-effects model analysis indicated that the 'dataset type' had significant effects on the R^2 from order 1 ($p < 0.001$, $\beta = 0.089$) to order 9 ($p = 0.008$, $\beta = -0.040$). Therefore, the reconstruction quality of the phasic and tonic dataset was different, especially for the orders of extraction that are commonly used in the literature. In Table 1, the mean numbers of extracted synergies across participants for the three R^2 thresholds are reported.

The mean order of factorization was higher for the phasic dataset for all the R^2 levels ($p < 0.001$). This result showed that more synergies are needed for the phasic EMG signals to achieve the same level of reconstruction quality with respect to the tonic dataset.

3.2. Synergy extraction

An example of the combined distributions of principal angles due to noise computed between phasic and tonic subspaces using the bootstrap procedure and the corresponding angles representing the 95th percentile of each distribution is shown in Fig. 3.

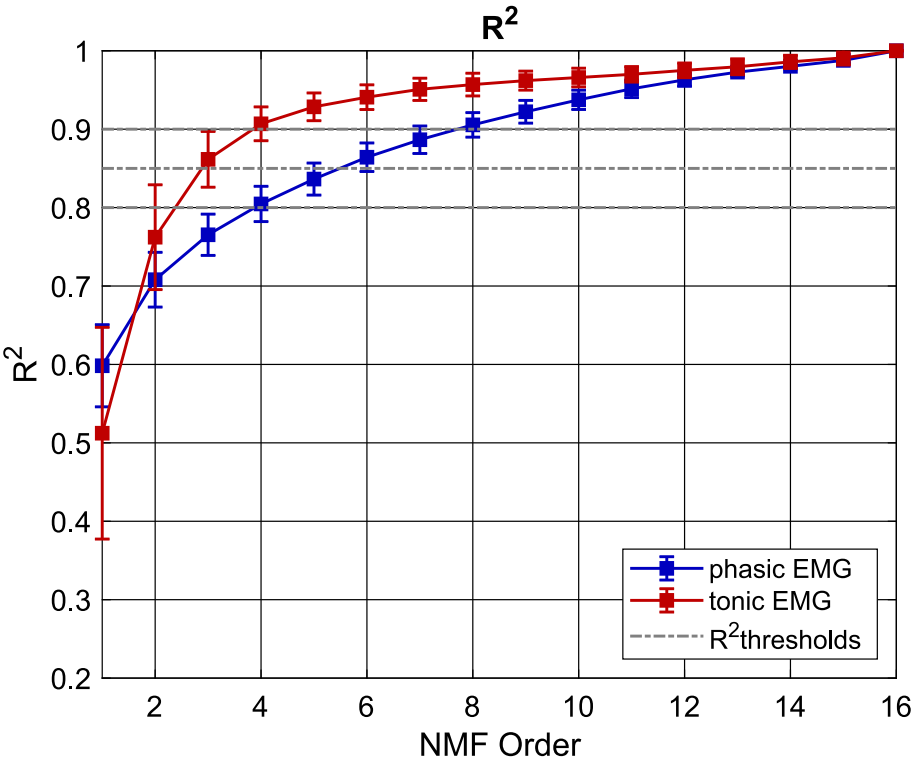

Fig. 2. Reconstruction R^2. Comparison between phasic EMG (blue) and tonic EMG (red). The squares are the mean across participants and the error bars represent the standard deviations. The dashed gray lines are the three R^2 thresholds (0.80, 0.85, 0.90). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 1
Number of extracted synergies. Means and standard deviations across participants for the number of extracted synergies for each R^2 threshold for phasic EMG and tonic EMG datasets. Standard deviations are represented in brackets.

N of extracted synergies			
	$R^2 = 0.80$	$R^2 = 0.85$	$R^2 = 0.90$
Phasic EMG	4.2 (0.8)	6.0 (0.8)	8.2 (1.0)
Tonic EMG	2.8 (0.5)	3.3 (0.7)	4.2 (0.9)

The number of principal angles increased with R^2 level, since the dimensions of the subspaces obtained from phasic and tonic datasets increased. Moreover, higher values of angles corresponded to a higher number of principal angles obtained and, therefore, a higher θ_{95}.

An example of the synergies extracted in a typical subject with one shared, two phasic-specific, and one tonic-specific synergies is shown in Fig. 4.

Shared synergies had non-zero time-varying coefficients in all the movement phases; phasic-specific synergies, instead, showed coefficients equal to zero in correspondence of the tonic dataset and tonic-specific synergies showed coefficients equal to zero in correspondence of the phasic dataset, as set in the input constraints.

From the extraction of all the subjects, the mean number of shared synergies was 0.75 (0.93) at R^2 threshold of 0.80, 1.87 (1.20) at R^2 threshold of 0.85, and 2.56 (1.50) at R^2 threshold of 0.90. The mean number of phasic-specific synergies was 1.62 (0.96) at R^2 threshold of 0.80, 1.87 (1.09) at R^2 threshold of 0.85, and 3.62 (1.45) at R^2 threshold of 0.90. The mean number of tonic-specific synergies was 1.19 (0.91) at R^2 threshold of 0.80, 1.06 (1.29) at R^2 threshold of 0.85, and 1.31 (1.30) at R^2 threshold of 0.90. These data are summarized in Table 2. The number of shared synergies was lower than phasic-specific synergies ($p = 0.03$) only at R^2 threshold of 0.80. The number of tonic-specific synergies were lower than shared ($p = 0.04$) and phasic-specific ($p = 10^{-4}$) synergies at R^2 threshold of 0.90.

3.3. Synergy clustering

Shared, phasic-specific, and tonic specific synergies extracted at each R^2 threshold were clustered separately and the results for the R^2 threshold of 0.80 are shown in Fig. 5.

At R^2 threshold of 0.80, a total of 12 shared, 26 phasic-specific, and 19 tonic specific synergies were extracted from all the participants. One cluster of 12 synergies was obtained for shared synergies, four clusters for phasic-specific synergies, with 8, 1, 13 and 4 synergies in each cluster, and two clusters for tonic-specific synergies, including 9 and 10 synergies each.

Shared, phasic-specific, and tonic specific synergies clusters of the synergies extracted at R^2 threshold of 0.85 are shown in Fig. 6.

At R^2 threshold of 0.85, a total of 30 shared, 30 phasic-specific, and 17 tonic specific synergies were extracted from all the

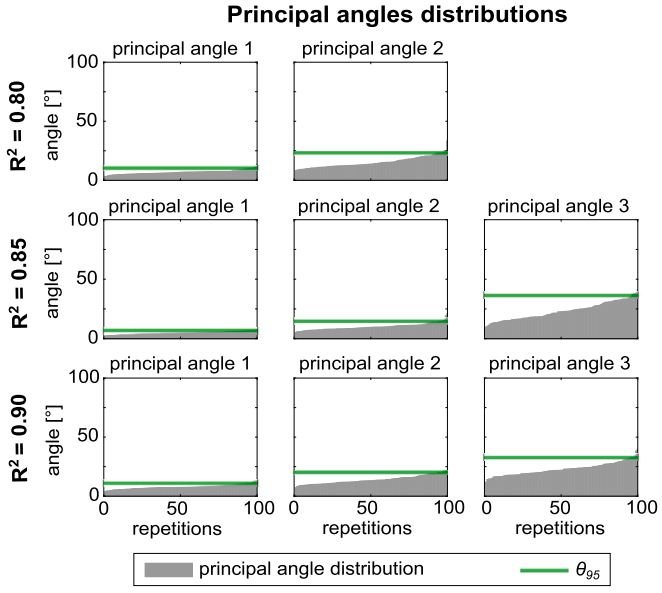

Fig. 3. Combined principal angles distributions. Example of distributions of combined principal angles due to noise computed between the two subspaces (phasic and tonic) obtained with the bootstrap procedure for a typical subject. Principal angles were computed at each R^2 level (in rows). The 95th percentile (θ_{95}) of each distribution was represented in green. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

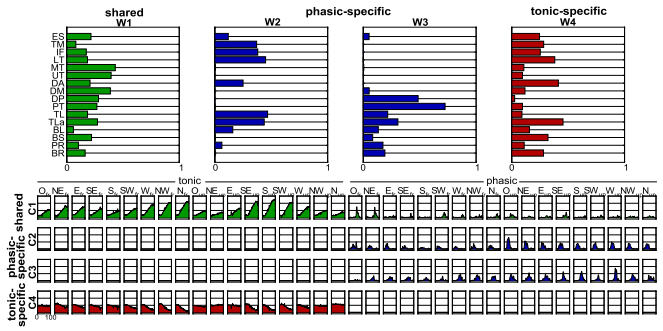

Fig. 4. Synergy extracted. Example of shared, phasic-specific, and tonic-specific synergies extracted from the combined phasic and tonic EMGs of a typical subject. One shared synergy (in green), two phasic-specific synergies (in blue) and one tonic-specific synergy (in red) are extracted with a R^2 threshold of 0.80. Muscle synergies W are shown in the upper panel, identifying the weightings of the 16 muscles in each synergy. The corresponding time-varying coefficients C are shown in the lower panel for each movement condition and dataset. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 2
N synergies, n clusters, similarity, and sparseness. Mean number of extracted synergies, number of clusters, intra-cluster similarity, and sparseness are reported for clusters of shared, phasic-specific, and tonic-specific synergies at three R^2 levels. The number of extracted synergies reports mean and standard deviation (in brackets) of the number extracted synergies between subjects. Intra-cluster similarity and sparseness are reported with mean and standard deviation (in brackets) computed between clusters.

	Shared	Phasic-specific	Tonic-specific
$R^2 = 0.80$			
Mean n of extracted synergies	0.75 (0.93)	1.62 (0.96)	1.19 (0.91)
N of clusters	1	4	2
Mean intra-cluster similarity	0.74 (0.00)	0.77 (0.02)	0.73 (0.03)
Mean sparseness	0.04 (0.00)	0.40 (0.14)	0.09 (0.02)
$R^2 = 0.85$			
Mean n of extracted synergies	1.87 (1.20)	1.87 (1.09)	1.06 (1.29)
N of clusters	6	9	3
Mean intra-cluster similarity	0.77 (0.05)	0.83 (0.04)	0.74 (0.04)
Mean sparseness	0.30 (0.20)	0.51 (0.08)	0.15 (0.08)
$R^2 = 0.90$			
Mean n of extracted synergies	2.56 (1.50)	3.62 (1.45)	1.31 (1.30)
N of clusters	6	11	3
Mean intra-cluster similarity	0.75 (0.04)	0.80 (0.06)	0.73 (0.03)
Mean sparseness	0.28 (0.11)	0.64 (0.10)	0.19 (0.14)

participants. Six clusters were obtained for shared synergies, in which two clusters contained 8 synergies, two clusters only 1 synergy, one cluster 7 synergies and one 5 synergies. Nine clusters were obtained for phasic-specific synergies, in which three clusters contained 2 synergies each, two clusters only 1 synergy each, and the other clusters included 3, 5, 6 and 8 synergies each. Tonic-specific synergies were grouped in only three clusters, including 8, 5 and 4 synergies each.

Shared, phasic-specific, and tonic specific clusters of the synergies extracted at R^2 threshold of 0.90 are shown in Fig. 7.

At R^2 threshold of 0.90, a total of 41 shared, 58 phasic-specific, and 21 tonic specific synergies were extracted from all the participants. Six clusters were obtained for shared synergies, in which two clusters contained 10 synergies, two clusters 4 synergies, one cluster 7 synergies and one 6 synergies. Eleven clusters were obtained for phasic-specific synergies, in which two clusters contained 12 synergies each, two clusters 4 synergies each, two clusters only 1 synergy each and the other clusters included 8, 6, 5, 3, and 2 synergies each. Tonic-specific synergies were grouped in only three clusters, including 10, 9 and 2 synergies each.

Mean number of extracted synergies, number of clusters, intra-cluster similarity, and sparseness are reported for clusters of shared, phasic-specific, and tonic-specific synergies at three R^2 levels in Table 2 and in Fig. 8.

The number of synergies extracted increased with the R^2 threshold and, for R^2 thresholds of 0.80 and 0.90, the number of phasic-specific synergies were higher than shared and tonic-specific synergies. Moreover, the phasic-specific synergies were grouped in more clusters than shared and tonic-specific synergies, even when the mean number of extracted synergies was similar as at R^2 thresholds of 0.80 and 0.85. These results may indicate that phasic-specific synergies are more differentiated and variable between subjects than shared and tonic-specific synergies. The mean intra-cluster similarity was higher for the phasic-specific synergies but this could be due to the higher number of clusters. Interestingly, phasic-specific synergies were sparser than tonic-specific synergies, suggesting that phasic synergies identify specific pattern related to the movement while tonic synergies show the activation of multiple muscles that

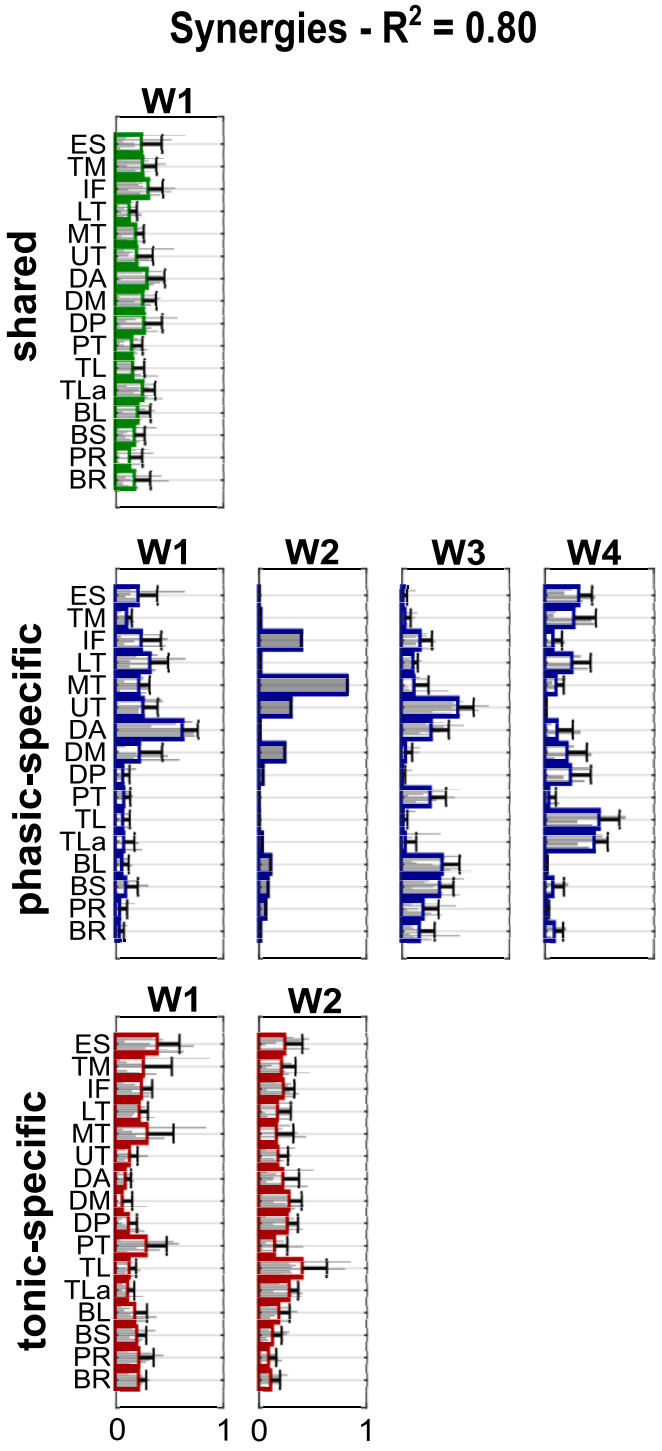

Fig. 5. Synergy clustering $R^2 = 0.80$. Synergy clustering of shared, phasic-specific, and tonic-specific synergies extracted at R^2 threshold of 0.80 from all the subjects.

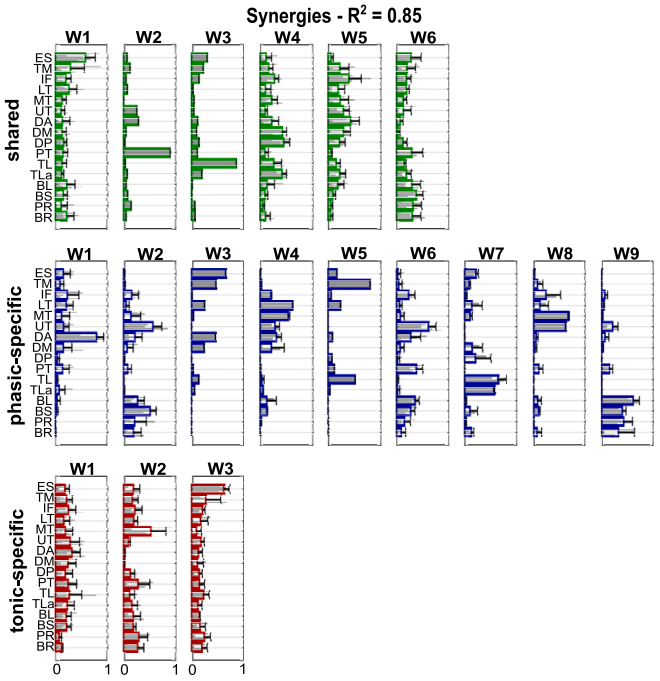

Fig. 6. Synergy clustering $R^2 = 0.85$. Synergy clustering of shared, phasic-specific and tonic-specific synergies extracted at R^2 threshold of 0.85 from all the subjects.

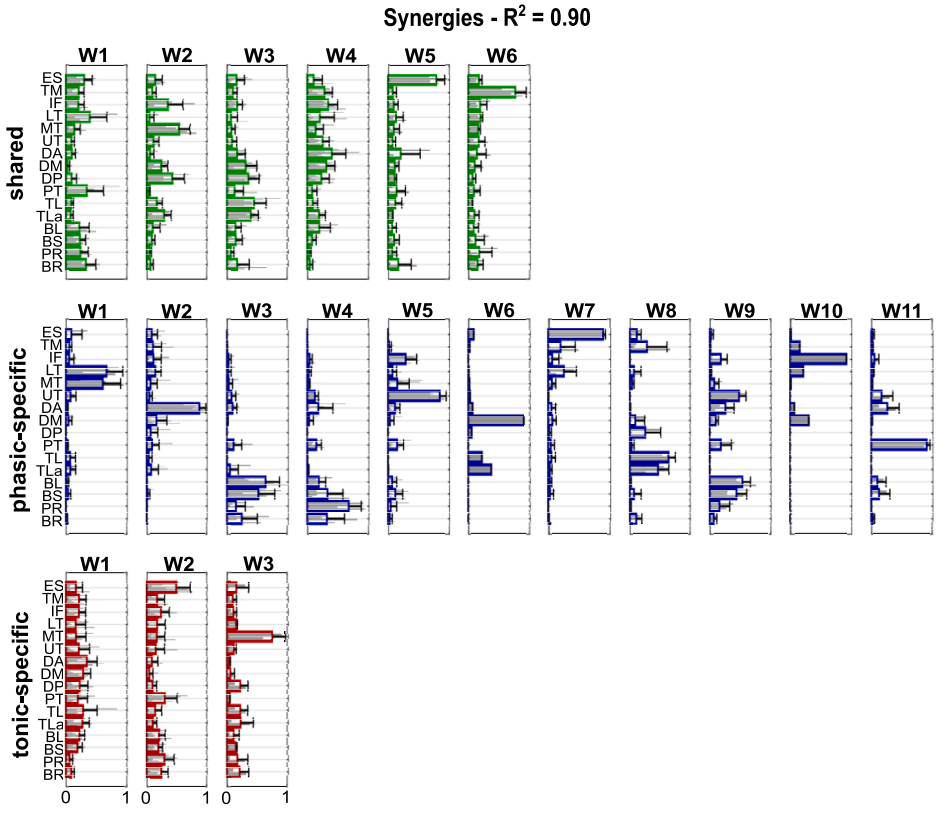

Fig. 7. Synergy clustering $R^2 = 0.90$. Synergy clustering of shared, phasic-specific, and tonic-specific synergies extracted at R^2 threshold of 0.90 from all the subjects.

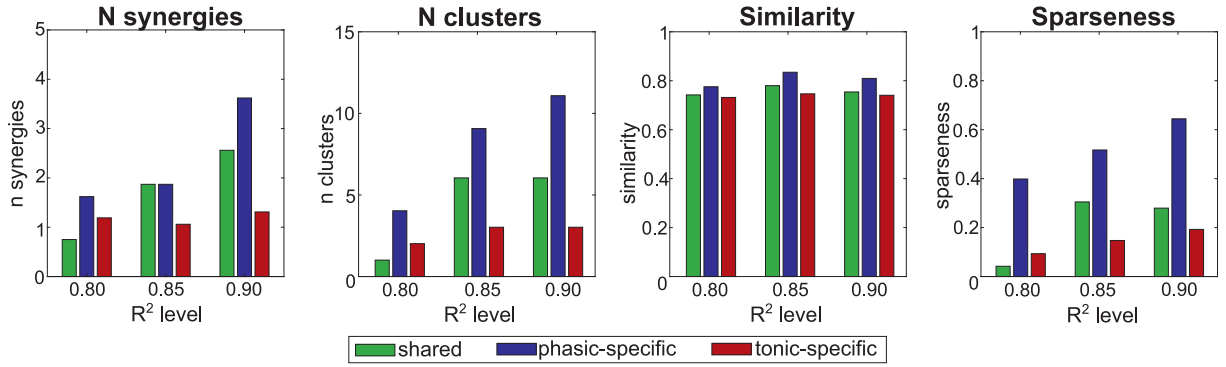

Fig. 8. N synergies, n clusters, similarity, and sparseness. Mean number of extracted synergies, number of clusters, mean intra-cluster similarity, and mean sparseness are compared across shared (in green), phasic-specific (in blue) and tonic-specific (in red) synergies for the three R^2 levels.

co-contract for joint stabilization.

4. Discussion

4.1. Summary of the results

The aim of the study is to provide a detailed characterization of phasic and tonic synergies to assess whether tonic and phasic EMG components share the same synergies (with different coefficients) or they are generated by different synergies. First, synergies were extracted separately from the phasic and the tonic EMG signals recorded during upper limb multi-directional reaching movements. The R^2 curves showed that the tonic EMG signals could be reconstructed with a higher quality than phasic signals according to most of the criteria for the selection of the number of synergies, especially for those that are commonly used in the literature (4–6). Furthermore, shared, phasic-specific and tonic-specific synergies were extracted from combined phasic and tonic EMG signals to directly assess which features are in common between phasic and tonic components and which are different. Then, synergies were clustered based on their similarity and their sparseness was examined. Phasic-specific synergies were clustered in a high number of clusters at all the R^2 levels, indicating that phasic synergies are more differentiated and variable among subjects, differently from the tonic-specific synergies that were grouped in a limited number of clusters. The limited number of shared synergies indicated that phasic and tonic synergies are different and they span separated subspaces. Finally, the sparseness was higher in phasic-specific clusters than in tonic-specific clusters, suggesting that phasic synergies identify specific patterns related to the movement. In contrast, the activation of multiple muscles in tonic synergies suggest they regulate co-contraction for joint stabilization or to counterbalance gravity. In sum, we demonstrated that phasic and tonic EMG components are generated by specific muscle synergies, and that the shared synergy space is limited.

4.2. Few shared synergies suggest that phasic and tonic components should be separated

Phasic and tonic synergies are different in number and structure, indicating that phasic and tonic EMG components are controlled by the CNS with specific synergies. Thus, in general, the CNS selects a subset of phasic synergies from a dictionary of phasic synergies related to the movement to execute and/or a subset of tonic synergies related to the posture to hold. Only few shared synergies can be used for controlling both phasic and tonic activity. In fact, motor control theories suggest that phasic and tonic activities are controlled with different strategies. The phasic activity may be controlled by a predictive scheme for scaling the acceleration needed to reach the target (Yadav & Sainburg, 2011). Phasic synergies activate specific muscles characterized by a peculiar directional tuning, as very selective activations are needed to achieve specific motor output. Thus, phasic synergies are higher in number and show a sparser structure. Since synergies reflect the EMG activity, phasic EMG activities show specific timings of acceleration and deceleration phases (Angel, 1974; Leib, Russo, D'Avella, & Nisky, 2020) of the reaching movement, whose kinematics is characterized by a bell-shaped velocity profile and consecutive peaks for acceleration and deceleration (Flash & Hogan, 1985; Mira et al., 2021). In fact, during reaching movements muscle activations are related to the acceleration and deceleration of shoulder and elbow joints (Tokuda et al., 2016) and are characterized by sequential bursts of agonist and antagonist muscle activations (Cooke & Brown, 1994).

On the other hand, the tonic activity is related both to joint stabilization and gravity compensation. Tonic control strategy may be related to an impedance control mechanism, needed to modify the mechanical properties of the limb or support the weight against gravity (Yadav & Sainburg, 2011). The resulting tonic synergies are fewer and less sparse than phasic ones, and they do not consist of specific muscle activations; on the contrary, more muscles are activated together, suggesting that muscles co-contract during tonic activation (Prange et al., 2009). Co-contraction of agonist and antagonist muscles can be clearly related to the joint stabilization against perturbations (Borzelli, Cesqui, Berger, Burdet, & D'Avella, 2018; Hasan, 1986), increasing joint stiffness. It may also contribute to anti-gravity function, since for holding upper limbs in static posture, gravity is counteracted by muscle co-contractions (Hogan, 1984; Latash, Aruin, & Shapiro, 1995). However, in our analysis, the two contributions of the tonic EMG cannot be

quantitatively separated, although they have different physiological functions.

Finally, shared synergies are less differentiated with respect to phasic synergies and they are less numerous, indicating that the shared space between phasic and tonic synergies is limited. Although the CNS seems to employ different motor control strategies for controlling phasic and tonic components, the two circuits of motor control may overlap and interact (Franklin et al., 2008), originating a reduced set of shared synergies that can be recruited with both phasic and tonic functions.

4.3. Applications of separated phasic and tonic components

Since phasic and tonic synergies are in general different, extracting synergies from the whole EMG could lead to a misinterpretation of the results. In fact, in many applications, the number and the composition of phasic and tonic synergies is related to specific physiological functions that may be equivocated if synergies are extracted from the whole EMG. This could happen especially for movements performed against gravity, such as upper limb movements. Movements with a limited tonic component, such as locomotion, are probably less affected by the separation of the components. The separation of phasic and tonic components of the EMG signal could be useful to assess the motor control in patients with hypertonia, like dystonic or stroke patients (Pisano et al., 2000), in which the excessive muscular tonic activity could hide the voluntary phasic activity. Analyzing only the phasic component could help the assessment of the effective muscular activity used for the movement and the evaluation of the effects of rehabilitation. Furthermore, the identification of phasic components could support the assessment of the effects of devices and exoskeletons used for neuromotor rehabilitation in neurological patients (Cancrini et al., 2022; Coscia et al., 2014) in a more effective way than using the whole EMG signal, as the analysis would target only the motion-related components. These devices compensate the gravity force, facilitating movement performance during therapy and improving recovery. Neural modification induced with neuromotor rehabilitation could probably be assessed with phasic synergies (Dipietro et al., 2007). Indeed, since the phasic activity of muscles is directly related to the kinematic patterns of the movement, the analysis of phasic synergies may allow to correctly assess and evaluate the coordination of activation patterns and to achieve more effective rehabilitation therapies for motor recovery.

Another application for the separation of phasic and tonic components regards the use of exoskeletons and devices for weight compensation in industrial scenarios for providing support to the back and the limbs reducing the effort of muscles and joints (de Looze, Bosch, Krause, Stadler, & O'Sullivan, 2016). These devices are usually employed to reduce the risk of musculoskeletal injuries in weight lifting (Abdoli-E & Stevenson, 2008) and overhead repetitive tasks (Kim et al., 2018). Ideally, they should act on the tonic EMG components reducing their activations without interfering with phasic components. With the proposed separated model, experimenters would be able to understand how exoskeletons or weight support devices reduce tonic components and if the devices modify the control structure underlying movement (phasic components). Since these kind of devices have been tested also in functional and repetitive dynamic tasks (Pacifico et al., 2020), this approach might also evaluate the ergonomics of the movement identifying the modification induced also in the phasic synergies. Finally, the separation of phasic synergies from tonic ones could be useful for the implementation of synergy-based control of neuroprosthesis (Cole & Ajiboye, 2019; Piazza et al., 2012) allowing to separate the components related to the desired movement in order to facilitate the movement classification (Li et al., 2022) and to reproduce it with a smaller number of command inputs (Dellacasa Bellingegni et al., 2017).

4.4. Methodological strengths and limitations

In this study, several original contributions were made. First, we demonstrated that phasic and tonic muscle activity are related to distinct phasic and tonic synergies, which are different in both number and structure. Synergies extracted from the whole EMG signals (i.e., combined phasic and tonic) are "hybrid" synergies that cannot be separated into phasic synergies and tonic synergies with standard NMF. Therefore, their different physiological function cannot be detected. Analyzing phasic or tonic synergies separately could be beneficial for a wide variety of applications. Moreover, our analysis included a variety of movements that explore the upper limb workspace, and represent typical movements in practical applications such as rehabilitation and industrial scenarios. The employed experimental protocols elicit proximal muscle activations (at shoulder and elbow level), with large variability of both phasic and tonic EMG activities. Furthermore, we provided a multiresolution analysis extracting synergies at three different levels of reconstruction quality (R^2 threshold of 0.80, 0.85, 0.90). In the literature, many different methods have been used in previous studies to determine the optimal number of synergies to extract (Ranaldi et al., 2021) and, therefore, we extracted synergies at three levels of reconstruction quality in order to guarantee the validity of our analysis, regardless of the reconstruction quality level. Finally, we employed a novel approach to estimate the dimensionality of the shared synergy space between the phasic and the tonic dataset with a bootstrap procedure aimed at evaluating the differences of the two subspaces spanned by phasic and tonic synergies with principal angles computed between the subspaces.

Despite the novel approaches, our analysis has some limitations. First, we investigated the effects of EMG tonic component removal only on the extraction of spatial synergies. Future work could include the extension of this methodology to other synergy models such as temporal synergies (Ivanenko, Cappellini, Dominici, Poppele, & Lacquaniti, 2005; Ivanenko, Poppele, & Lacquaniti, 2006), spatiotemporal (D'Avella et al., 2006; D'Avella, Saltiel, & Bizzi, 2003) or space-by-time (Delis, Panzeri, Pozzo, & Berret, 2014, 2015), to evaluate how the tonic component influences the temporal patterns. In addition, it could be interesting to explore the comparison between spatial and temporal models (Brambilla, Atzori, Müller, D'Avella, & Scano, 2023), or the comparison with synergistic approaches in the kinematic domain (Bockemühl, Troje, & Dürr, 2010). Another limitation of this study is that only reaching movements were analyzed. Other upper limb or whole body movement could be included to analyze the role of tonic synergies in more complex movements, that require more refined motor control. Furthermore, the tonic EMG signal was approximated with the linear ramp

model. It could be interesting to remove the tonic component with other models, as identifying the principal component related to the tonic activity (Flanders & Herrmann, 1992), extracting the tonic spatiotemporal synergies (D'Avella et al., 2008) or estimating the tonic component from muscle torques (Olesh, Pollard, & Gritsenko, 2017).

Finally, the negative components of the phasic signal, resulting from the tonic component removal, were set to zero in order to apply the NMF algorithm. The negative components of the phasic activity could be related to a motor control strategy that exploits gravity to reduce the activity of muscles. Gaveau, Grospretre, Berret, Angelaki, and Papaxanthis (2021) demonstrated that motor system takes advantage of the mechanical effects of gravity to accelerate downward and decelerate upward movements, and of inertial forces in different ways in upward or downward movements (Papaxanthis, Pozzo, & Schieppati, 2003), saving muscle effort. Thus, the exploitation of inertial and gravity forces reduces the activity of some muscles and generates negative phasic EMG components when the tonic EMG is removed. The contribution of the negative phasic components of the EMG signal will be analyzed in details in future work, using algorithms that allow to factorize the negative components as the Mixed Matrix Factorization (MMF) algorithm (Scano, Mira, & d'Avella, 2022), in order to understand better the role of the negative components of the phasic signal.

5. Conclusions

In this paper, we demonstrated that phasic and tonic activity are generated by distinct phasic and tonic synergies and we provided a detailed characterization of phasic and tonic synergies, showing that they are different in number and structure, and that their shared subspace is limited. We compared the goodness of reconstruction between phasic and tonic EMG datasets, and we assessed the number of clusters, the intra-cluster similarity, and the sparseness of shared, phasic-specific and tonic-specific synergies. We showed that a given number of synergies achieve a higher reconstruction of the tonic EMG signals than of the phasic signals. Moreover, phasic-specific synergies were grouped in more clusters, suggesting that they are more differentiated and variable between subjects than shared and tonic-specific synergies. Phasic-specific synergies were sparser than tonic-specific synergies, suggesting that phasic synergies identify specific pattern related to the movement, while tonic synergies capture the activation of multiple muscles co-contracting for joint stabilization. These results can be used to improve muscle synergy analysis targeting several fields such as basic human motor control, neuromotor rehabilitation, and prosthesis control.

CRediT authorship contribution statement

Cristina Brambilla: Data curation, Formal analysis, Investigation, Methodology, Software, Validation, Visualization, Writing – original draft, Writing – review & editing. **Marta Russo:** Investigation, Supervision, Writing – original draft, Writing – review & editing. **Andrea d'Avella:** Conceptualization, Investigation, Methodology, Software, Supervision, Writing – original draft, Writing – review & editing. **Alessandro Scano:** Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing.

Declaration of Competing Interest

None.

Data availability

Data will be made available on request.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.humov.2023.103148.

References

Abdoli-E, M., & Stevenson, J. M. (2008). The effect of on-body lift assistive device on the lumbar 3D dynamic moments and EMG during asymmetric freestyle lifting. *Clinical biomechanics, 23*(3), 372–380. https://doi.org/10.1016/j.clinbiomech.2007.10.012

Albert, S. T., Hadjiosif, A. M., Jang, J., Zimnik, A. J., Soteropoulos, D. S., Baker, S. N., … Shadmehr, R. (2020). Postural control of arm and fingers through integration of movement commands. *ELife, 9*. https://doi.org/10.7554/eLife.52507

Angel, R. W. (1974). Electromyography during voluntary movement: The two-burst pattern. *Electroencephalography and Clinical Neurophysiology, 36*(C), 493–498. https://doi.org/10.1016/0013-4694(74)90206-5

Arthur, D., & Vassilvitskii, S. (2007). K-means++: The advantages of careful seeding. In *Proceedings of the annual ACM-SIAM symposium on discrete algorithms, 07-09-Janu* (pp. 1027–1035).

Bizzi, E., Cheung, V. C. K., D'Avella, A., Saltiel, P., & Tresch, M. (2008). Combining modules for movement. In *, 57, issue 1. Brain research reviews* (pp. 125–133). Brain Res Rev https://doi.org/10.1016/j.brainresrev.2007.08.004.

Bockemühl, T., Troje, N. F., & Dürr, V. (2010). Inter-joint coupling and joint angle synergies of human catching movements. *Human Movement Science, 29*(1), 73–93. https://doi.org/10.1016/j.humov.2009.03.003

Borzelli, D., Cesqui, B., Berger, D. J., Burdet, E., & D'Avella, A. (2018). Muscle patterns underlying voluntary modulation of co-contraction. *PLoS One, 13*(10), Article e0205911. https://doi.org/10.1371/journal.pone.0205911
Brambilla, C., Atzori, M., Müller, H., D'Avella, A., & Scano, A. (2023). Spatial and temporal muscle synergies provide a dual characterization of low-dimensional and intermittent control of upper-limb movements. *Neuroscience, 514*, 100–122. https://doi.org/10.1016/j.neuroscience.2023.01.017
Cancrini, A., Baitelli, P., Nicora, M. L., Malosio, M., Pedrocchi, A., & Scano, A. (2022). The effects of robotic assistance on upper limb spatial muscle synergies in healthy people during planar upper-limb training. *PLoS One, 17*(8 August), 1–25. https://doi.org/10.1371/journal.pone.0272813
Cheung, V. C. K., D'Avella, A., Tresch, M. C., & Bizzi, E. (2005). Central and sensory contributions to the activation and organization of muscle synergies during natural motor behaviors. *Journal of Neuroscience, 25*(27), 6419–6434. https://doi.org/10.1523/JNEUROSCI.4904-04.2005
Cole, N. M., & Ajiboye, A. B. (2019). Muscle synergies for predicting non-isometric complex hand function for commanding FES neuroprosthetic hand systems. *Journal of Neural Engineering, 16*(5), Article 056018. https://doi.org/10.1088/1741-2552/ab2d47
Cooke, J. D., & Brown, S. H. (1994). Movement-related phasic muscle activation - III. The duration of phasic agonist activity initiating movement. *Experimental Brain Research, 99*(3), 473–482. https://doi.org/10.1007/BF00228984
Coscia, M., Cheung, V. C., Tropea, P., Koenig, A., Monaco, V., Bennis, C., … Bonato, P. (2014). The effect of arm weight support on upper limb muscle synergies during reaching movements. *Journal of Neuroengineering and Rehabilitation, 11*(1), 1–15. https://doi.org/10.1186/1743-0003-11-22
D'Avella, A., & Bizzi, E. (2005). Shared and specific muscle synergies in natural motor behaviors. *Proceedings of the National Academy of Sciences of the United States of America, 102*(8), 3076–3081. https://doi.org/10.1073/pnas.0500199102
D'Avella, A., Fernandez, L., Portone, A., & Lacquaniti, F. (2008). Modulation of phasic and tonic muscle synergies with reaching direction and speed. *Journal of Neurophysiology, 100*(3), 1433–1454. https://doi.org/10.1152/jn.01377.2007
D'Avella, A., & Lacquaniti, F. (2013). Control of reaching movements by muscle synergy combinations. *Frontiers in Computational Neuroscience, 0*(APR 2013), 42. https://doi.org/10.3389/fncom.2013.00042
D'Avella, A., Portone, A., Fernandez, L., & Lacquaniti, F. (2006). Control of fast-reaching movements by muscle synergy combinations. *Journal of Neuroscience, 26*(30), 7791–7810. https://doi.org/10.1523/JNEUROSCI.0830-06.2006
D'Avella, A., Saltiel, P., & Bizzi, E. (2003). Combinations of muscle synergies in the construction of a natural motor behavior. *Nature Neuroscience, 6*(3), 300–308. https://doi.org/10.1038/nn1010
Delis, I., Panzeri, S., Pozzo, T., & Berret, B. (2014). A unifying model of concurrent spatial and temporal modularity in muscle activity. *Journal of Neurophysiology, 111*(3), 675–693. https://doi.org/10.1152/jn.00245.2013
Delis, I., Panzeri, S., Pozzo, T., & Berret, B. (2015). Task-discriminative space-by-time factorization of muscle activity. *Frontiers in Human Neuroscience, 9*(JULY). https://doi.org/10.3389/fnhum.2015.00399
Dellacasa Bellingegni, A., Gruppioni, E., Colazzo, G., Davalli, A., Sacchetti, R., Guglielmelli, E., & Zollo, L. (2017). NLR, MLP, SVM, and LDA: A comparative analysis on EMG data from people with trans-radial amputation. *Journal of Neuroengineering and Rehabilitation, 14*(1). https://doi.org/10.1186/s12984-017-0290-6
Dipietro, L., Krebs, H. I., Fasoli, S. E., Volpe, B. T., Stein, J., Bever, C., & Hogan, N. (2007). Changing motor synergies in chronic stroke. *Journal of Neurophysiology, 98*(2), 757–768. https://doi.org/10.1152/jn.01295.2006
Flanders, M. (1991). Temporal patterns of muscle activation for arm movements in three-dimensional space. *Journal of Neuroscience, 11*(9), 2680–2693. https://doi.org/10.1523/jneurosci.11-09-02680.1991
Flanders, M., & Herrmann, U. (1992). Two components of muscle activation: Scaling with the speed of arm movement. *Journal of Neurophysiology, 67*(4), 931–943. https://doi.org/10.1152/jn.1992.67.4.931
Flash, T., & Hogan, N. (1985). The coordination of arm movements: An experimentally confirmed mathematical model. *Journal of Neuroscience, 5*(7), 1688–1703. https://doi.org/10.1523/jneurosci.05-07-01688.1985
Franklin, D. W., Burdet, E., Keng, P. T., Osu, R., Chew, C. M., Milner, T. E., & Kawato, M. (2008). CNS learns stable, accurate, and efficient movements using a simple algorithm. *Journal of Neuroscience, 28*(44), 11165–11173. https://doi.org/10.1523/JNEUROSCI.3099-08.2008
Gaveau, J., Grospretre, S., Berret, B., Angelaki, D. E., & Papaxanthis, C. (2021). A cross-species neural integration of gravity for motor optimization. *Science Advances, 7*(15). https://doi.org/10.1126/SCIADV.ABF7800/SUPPL_FILE/ABF7800_SM.PDF
Golub, G. H., & Van Loan, C. F. (1996). Matrix computations. In (3rd editio), *Vols. 208–209. Linear algebra and its applications*. Johns Hopkins University Press.
Hartigan, J. A. (1975). *Clustering algorithms*.
Hasan, Z. (1986). Optimized movement trajectories and joint stiffness in unperturbed, inertially loaded movements. *Biological Cybernetics, 53*(6), 373–382. https://doi.org/10.1007/BF00318203
Hermens, H. J., Freriks, B., Disselhorst-Klug, C., & Rau, G. (2000). Development of recommendations for SEMG sensors and sensor placement procedures. *Journal of Electromyography and Kinesiology, 10*(5), 361–374. https://doi.org/10.1016/S1050-6411(00)00027-4
Hogan, N. (1984). Adaptive control of mechanical impedance by Coactivation of antagonist muscles. *IEEE Transactions on Automatic Control, 29*(8), 681–690. https://doi.org/10.1109/TAC.1984.1103644
Hollerbach, J. M., & Flash, T. (1982). Dynamic interactions between limb segments during planar arm movement. *Biological Cybernetics, 44*(1), 67–77. https://doi.org/10.1007/BF00353957
Hoyer, P. O. (2004). Non-negative matrix factorization with sparseness constraints. *Journal of Machine Learning Research, 5*, 1457–1469.
Huang, B., Chen, W., Liang, J., Cheng, L., & Xiong, C. (2022). Characterization and categorization of various human lower limb movements based on kinematic synergies. *Frontiers in Bioengineering and Biotechnology, 9*(January), 1–12. https://doi.org/10.3389/fbioe.2021.793746
Ivanenko, Y., Cappellini, G., Dominici, N., Poppele, R. E., & Lacquaniti, F. (2005). Coordination of locomotion with voluntary movements in humans. *Journal of Neuroscience, 25*(31), 7238–7253. https://doi.org/10.1523/JNEUROSCI.1327-05.2005
Ivanenko, Y., & Gurfinkel, V. (2018). Human postural control. *Frontiers in Neuroscience, 12*(MAR), 171. https://doi.org/10.3389/fnins.2018.00171
Ivanenko, Y., Poppele, R., & Lacquaniti, F. (2006). Motor control programs and walking. *Neuroscientist, 12*(4), 339–348. https://doi.org/10.1177/1073858406287987
Kim, S., Nussbaum, M. A., Mokhlespour Esfahani, M. I., Alemi, M. M., Alabdulkarim, S., & Rashedi, E. (2018). Assessing the influence of a passive, upper extremity exoskeletal vest for tasks requiring arm elevation: Part I – "Expected" effects on discomfort, shoulder muscle activity, and work task performance. *Applied Ergonomics, 70*, 315–322. https://doi.org/10.1016/j.apergo.2018.02.025
Latash, M. L., Aruin, A. S., & Shapiro, M. B. (1995). The relation between posture and movement: A study of a simple synergy in a two-joint task. *Human Movement Science, 14*(1), 79–107. https://doi.org/10.1016/0167-9457(94)00046-H
Lee, D. D., & Seung, H. S. (1999). Learning the parts of objects by non-negative matrix factorization. *Nature, 401*(6755), 788–791. https://doi.org/10.1038/44565
Leib, R., Russo, M., D'Avella, A., & Nisky, I. (2020). A bang-bang control model predicts the triphasic muscles activity during hand reaching. *Journal of Neurophysiology, 124*(1), 295–304. https://doi.org/10.1152/jn.00132.2020
Li, Z., Zhao, X., Wang, Z., Xu, R., Meng, L., & Ming, D. (2022). A hierarchical classification of gestures under two force levels based on muscle synergy. *Biomedical Signal Processing and Control, 77*, Article 103695. https://doi.org/10.1016/j.bspc.2022.103695
de Looze, M. P., Bosch, T., Krause, F., Stadler, K. S., & O'Sullivan, L. W. (2016). Exoskeletons for industrial application and their potential effects on physical work load. *Ergonomics, 59*(5), 671–681. https://doi.org/10.1080/00140139.2015.1081988
McLean, R. A., Sanders, W. L., & Stroup, W. W. (1991). A unified approach to mixed linear models. *The American Statistician, 45*(1), 54. https://doi.org/10.2307/2685241
Mira, R. M., Molinari Tosatti, L., Sacco, M., & Scano, A. (2021). Detailed characterization of physiological EMG activations and directional tuning of upper-limb and trunk muscles in point-to-point reaching movements. *Current Research in Physiology, 4*, 60–72. https://doi.org/10.1016/J.CRPHYS.2021.02.005
Olesh, E. V., Pollard, B. S., & Gritsenko, V. (2017). Gravitational and dynamic components of muscle torque underlie tonic and phasic muscle activity during goal-directed reaching. *Frontiers in Human Neuroscience, 11*, 474. https://doi.org/10.3389/fnhum.2017.00474
Pacifico, I., Molteni, F., Giovacchini, F., Vitiello, N., Crea, S., Scano, A., … Colombina, G. (2020). An experimental evaluation of the proto-mate: A novel ergonomic upper-limb exoskeleton to reduce workers' physical strain. *IEEE Robotics and Automation Magazine, 27*(1), 54–65. https://doi.org/10.1109/MRA.2019.2954105

Pale, U., Atzori, M., Müller, H., & Scano, A. (2020). Variability of muscle synergies in hand grasps: Analysis of intra-and inter-session data. *Sensors, 20*(15), 1–27. https://doi.org/10.3390/s20154297

Papaxanthis, C., Pozzo, T., & Schieppati, M. (2003). Trajectories of arm pointing movements on the sagittal plane vary with both direction and speed. *Experimental Brain Research, 148*(4), 498–503. https://doi.org/10.1007/s00221-002-1327-y

Piazza, S., Torricelli, D., Brunetti, F., Del-Ama, A. J., Gil-Agudo, A., & Pons, J. L. (2012). A novel FES control paradigm based on muscle synergies for postural rehabilitation therapy with hybrid exoskeletons. In *, 2012. Proceedings of the Annual International Conference of the IEEE Engineering in Medicine and Biology Society* (pp. 1868–1871). EMBS. https://doi.org/10.1109/EMBC.2012.6346316.

Pisano, F., Miscio, G., Del Conte, C., Pianca, D., Candeloro, E., & Colombo, R. (2000). Quantitative measures of spasticity in post-stroke patients. *Clinical Neurophysiology, 111*(6), 1015–1022. https://doi.org/10.1016/S1388-2457(00)00289-3

Prange, G. B., Kallenberg, L. A. C., Jannink, M. J. A., Stienen, A. H. A., van der Kooij, H., IJzerman, M. J., & Hermens, H. J. (2009). Influence of gravity compensation on muscle activity during reach and retrieval in healthy elderly. *Journal of Electromyography and Kinesiology, 19*(2), e40–e49. https://doi.org/10.1016/J.JELEKIN.2007.08.001

Ranaldi, S., De Marchis, C., Severini, G., & Conforto, S. (2021). An objective, information-based approach for selecting the number of muscle synergies to be extracted via non-negative matrix factorization. *IEEE Transactions on Neural Systems and Rehabilitation Engineering, 29*, 2676–2683. https://doi.org/10.1109/TNSRE.2021.3134763

Russo, M., D'Andola, M., Portone, A., Lacquaniti, F., & D'Avella, A. (2014). Dimensionality of joint torques and muscle patterns for reaching. *Frontiers in Computational Neuroscience, 8*(MAR), 24. https://doi.org/10.3389/fncom.2014.00024

Scano, A., Dardari, L., Molteni, F., Giberti, H., Tosatti, L. M., & D'Avella, A. (2019). A comprehensive spatial mapping of muscle synergies in highly variable upper-limb movements of healthy subjects. *Frontiers in Physiology, 10*(SEP), 1231. https://doi.org/10.3389/fphys.2019.01231

Scano, A., Mira, R. M., & d'Avella, A. (2022). Mixed matrix factorization: A novel algorithm for the extraction of kinematic-muscular synergies. *Journal of Neurophysiology, 127*(2), 529–547. https://doi.org/10.1152/JN.00379.2021/ASSET/IMAGES/MEDIUM/JN-00379-2021R01.PNG

Sylos-Labini, F., La Scaleia, V., Cappellini, G., Fabiano, A., Picone, S., Keshishian, E. S., … Lacquaniti, F. (2020). Distinct locomotor precursors in newborn babies. *Proceedings of the National Academy of Sciences of the United States of America, 117*(17), 9604–9612. https://doi.org/10.1073/pnas.1920984117

Tokuda, K., Lee, B., Shiihara, Y., Takahashi, K., Wada, N., Shirakura, K., & Watanabe, H. (2016). Muscle activation patterns in acceleration-based phases during reach-to-grasp movement. *Journal of Physical Therapy Science, 28*(11), 3105–3111. https://doi.org/10.1589/jpts.28.3105

Torres-Oviedo, G., & Ting, L. H. (2010). Subject-specific muscle synergies in human balance control are consistent across different biomechanical contexts. *Journal of Neurophysiology, 103*(6), 3084–3098. https://doi.org/10.1152/jn.00960.2009

Yadav, V., & Sainburg, R. L. (2011). Motor lateralization is characterized by a serial hybrid control scheme. *Neuroscience, 196*, 153–167. https://doi.org/10.1016/j.neuroscience.2011.08.039

Zhao, K., Zhang, Z., Wen, H., & Scano, A. (2021). Intra-subject and inter-subject movement variability quantified with muscle synergies in upper-limb reaching movements. *Biomimetics, 6*(4), 63. https://doi.org/10.3390/biomimetics6040063