Age-Related Modifications of Muscle Synergies and Their Temporal Activations for Overground Walking

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Abstract—Healthy ageing modifies neuromuscular control of human overground walking. Previous studies found that ageing changes gait biomechanics, but whether there is concurrent ageing-related modulation of neuromuscular control remains unclear. We analyzed gait kinematics and electromyographic signals (EMGs; 14 lower-limb and trunk muscles) collected at three speeds during overground walking in 11 healthy young adults (mean age of 23.4 years) and 11 healthy elderlies (67.2 years). Neuromuscular control was characterized by extracting muscle synergies from EMGs and the synergies of both groups were *k*-means-clustered. The synergies of the two groups were grossly similar, but we observed numerous cluster- and muscle-specific differences between the age groups. At the population level, some hip-motion-related synergy clusters were more frequently identified in elderlies while others, more frequent in young adults. Such differences in synergy prevalence between the age groups are consistent with the finding that elderlies had a larger hip flexion range. For the synergies shared between both groups, the elderlies had higher inter-subject variability of the temporal activations than young adults. To further explore what synergy characteristics may be related to this inter-subject variability, we found that the inter-subject variance of temporal activations correlated negatively with the sparseness of the synergies in elderlies but not young adults during slow walking. Overall, our results suggest that as humans age, not only are the muscle synergies for walking fine-tuned in structure, but their temporal activation patterns are also more heterogeneous across individuals, possibly reflecting individual differences in prior sensorimotor experience or ageing-related changes in limb neuro-musculoskeletal properties.

Index Terms—Muscle synergy, ageing, gait, electromyographic signals (EMGs), motor variability.

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I. INTRODUCTION

D URING ageing, gait characteristics may gradually change as a result of age-related changes in the neuro-musculoskeletal system, including reduced muscle strength, decreased joint power, and deteriorated sensorimotor functions [1], [2]. Such changes in gait may increase the risk of injury or even reduce mobility [3], [4]. Some past sensorimotor experience or any history of neuromuscular impairment may also lead to the expression of different learned and compensatory patterns in the person's gait. For instance, when compared with healthy elderlies, disabled elderlies have significantly higher mid-stance hip mechanical energy expenditure related to compensatory gait strategies [5]. Presumably, these altered gait patterns, be they related to age-dependent changes in biomechanics, prior learning, or past injuries, are accompanied by alterations in the neuromuscular gait control strategy implemented by the central nervous system [6].

How is the neural control of gait adjusted during ageing to ensure efficient dynamic motor control as the vast number of internal and external variables fluctuate over the years? Answering this question would necessitate a thorough, mechanistic understanding of how the immensely variable locomotor muscle patterns are constructed by the motor system. One way to study the neural implementation of gait control is to reduce the observed high-dimensional motor patterns into a low-dimensional set of motor modules representable as muscle synergies [7], [8]. When a muscle synergy is recruited, multiple muscles are activated as specific spinal premotor interneurons [9], [10] and/or motor cortical neurons [11]

synergistically activate different motoneuronal pools. The muscular compositions of the muscle synergies encoded by these neurons may be identified as vectors decomposed from electromyographic signals (EMGs) using factorization algorithms. As putative representations of neuromotor modules, muscle synergies and their temporal activations can be regarded as markers that reflect modifications of the motor control policy over time.

Recent theoretical and experimental studies have suggested that EMG-derived muscle synergies may be updated as limb biomechanical properties change. In a study that simulated gait using a realistic neuro-musculoskeletal model, the different biomechanical demands of walking are indeed reflected by the muscular compositions of the synergies [12], thus suggesting that for consistent gait, the synergies' structures should depend on limb biomechanics. Consistent with the notion that muscle synergies and their activations exhibit considerable plasticity, studies in early motor development have revealed that as the anthropometry of infants and children mature, some early muscle synergies fractionate into units with fewer muscles [13], [14]. Other results have shown that even though some synergies may remain invariant in structure over the years, their temporal control patterns still change throughout life [15], [16]. Muscle synergies can also be modified after motor learning [17] or neurological injuries. In stroke patients, gait impairment appeared to be related to the expression of abnormal synergies that could be explained by merging multiple normal synergies [18]. Similar processes of synergy merging have also been reported in stroke survivors with upper-limb impairment [19] and runners undergoing motor training [14].

In the literature, there are numerous but scattered studies on the relationship between ageing and synergy modifications in locomotor tasks. Monaco *et al.* [20] found that the gross structures of the muscle synergies and their temporal activations were unchanged by ageing, but they suggested that ageing may impact how spinal circuits integrate peripheral afference and descending inputs, resulting in modification of the final motor output in the older subjects. Baggen *et al.* [16], on the other hand, showed that synergy structures and complexities changed during ageing even though the between-task synergy similarity was higher in the older group than in the younger group. The above studies prove that a few basic muscle synergies for gait can be identified from both younger and older adults, but it remains unclear whether the between-group differences in motor outputs may be attributed to changes in specific muscle synergies, their temporal activities, or both.

Here, we aim to clarify whether the neural control of gait is altered during ageing by comparing the locomotor muscle synergies obtained from younger adults with those obtained from older adults. To reveal how the synergies may differ between the two age groups at the population level, we examined the inter-subject variability of the synergies in both groups and correlated them with kinematic measures. To further understand any potential difference in gait control stability between the two groups, we characterized both the within-subject and inter-subject variability of the synergies' temporal activations. Our analysis argues that as humans age, not only are the muscle synergies fine-tuned in structure, but
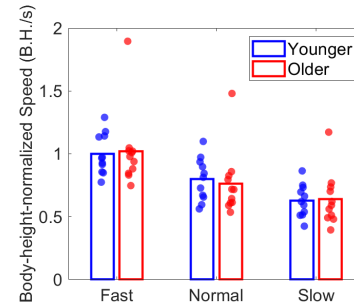

Fig. 1. The body-height normalized walking speeds selected by the subjects. The between-group differences in three walking speeds are not statistically significant. (fast: $p = 0.85$; normal: $p = 0.71$; slow: $p = 0.88$; ANOVA).

the activation patterns of specific synergies also change. These modifications of muscle synergies may originate from both age-dependent changes in gait biomechanics and the varied sensorimotor experience of the subjects through the years.

II. MATERIALS AND METHODS

A. Participants

Two groups of healthy subjects, a younger (N = 11; 7 females; age 23.4 ± 2.5 years) and an older (N = 11; 7 females; age 67.2 ± 4.3 years) group, participated. All subjects had no history of any musculoskeletal or neurological injury or surgery. Informed consent was obtained from each subject before experimentation. All procedures were approved by The Joint Chinese University of Hong Kong - New Territories East Cluster Clinical Research Ethics Committee (no. 2019. 498).

B. EMG and Kinematic Data Collection

Each subject was instructed to walk overground for 3.6 m at three self-selected speeds (slow, normal, and fast) (Fig. 1). As the subject walked, electromyographic signals (EMGs; sampling rate of 1000 Hz) (14 muscles on each side) were recorded by wireless surface EMG electrodes (Trigno, Delsys, Boston, MA, USA). Before EMG sensor attachment, the skin over the attachment positions was cleaned by alcohol (75%). The sensors were then placed according to SENIAM recommendations [21] and attached to skin surface with double-sided tape, and stabilized in position with self-adherent bandage wrap (3M Coban™). The muscles recorded included tibialis anterior (TA), medial (MG) and lateral heads of the gastrocnemius (LG), soleus (Sol), vastus lateralis (VL), vastus medialis (VM), rectus femoris (RF), hamstrings (Hams), adductor longus (AL), tensor fascia latae (TFL), gluteus maximus (GM), erector spinae (ES), external oblique (ExtO) and latissimus dorsi (LatDor).

During walking, full-body kinematics were also recorded using 3D motion capture cameras (Vicon, Oxford Metrics, Oxford, UK; 200 Hz). Infrared reflective markers (N = 39) were placed according to the requirements of the full-body Plug-in Gait model implemented by Vicon Nexus. The gait cycles for synergy and kinematics analysis were selected based on data quality. For synergy extraction on average,

19±3 and 30±2 cycles were selected for each young and old subject, respectively. Evidences have shown that 20 cycles are sufficient for synergy extraction for reconstructing EMGs with optimal quality [22]. For joint angle comparison, cycles were selected according to the quality of the marker trajectories. We excluded cycles with absent heel or toe markers; as a result, 11±3 and 19±3 cycles were selected for the young and old subjects, respectively. Kinematic data of specific joints would be further compared between subject groups based on the comparison results of the subsequent muscle-synergy analysis. We defined the hip angle to be the angle between the projected sagittal thigh axis and the sagittal pelvic axis. The hip flexion/extension angle during static standing was defined as the baseline and thus set to 0°. A positive/negative (flexion/extension) angle corresponds to one with the hip positioned anterior/posterior to the body. The peak hip flexion and extension were then conveniently obtained from the maximum flexion and extension angles, respectively (Fig. 6, 7). Bending the knee by knee flexors is referred to as knee flexion, so the opposite direction is knee extension. Thus, increase and decrease of the angle represent knee flexion and extension, respectively. Peak knee flexion is equivalent to the knee range of motion.

C. EMG Preprocessing and Muscle Synergy Extraction

The EMGs collected from the right side of each subject were preprocessed with the following steps: removal of noise from powerline interference [23], [24], high-pass filtering (cutoff at 40 Hz), rectification, low-pass filtering (40 Hz), integration (over 20-ms intervals) and variance normalization of each muscle [25]. The cutoff frequencies of the high- and low-pass filters were selected to preserve the most information in the EMGs while keeping the number of extracted synergies the same even when higher or lower cutoff frequencies were used for the high- or low-pass filters, respectively. This was followed by applying the non-negative matrix factorization algorithm (NNMF) to extract time-invariant muscle synergies from the EMGs [26]. Let $\mathbf{D}$ be a non-negative $m \times n$ data matrix comprising n samples of an m-dimension data vector. The NNMF models $\mathbf{D}$ to be a linear combination of two matrices $\mathbf{W}$ and $\mathbf{C}$, such that

$$\mathbf{D} = \mathbf{W}\mathbf{C}^T + \mathbf{R} = \sum \mathbf{w}_i \mathbf{c}_i^T + \mathbf{R} \qquad (1)$$

where vector $\mathbf{w}_i$ is the ith column of $\mathbf{W}$ denoting the ith muscle synergy, and vector $\mathbf{c}_i$, i^{th} column of $\mathbf{C}$, is the temporal activation coefficients for $\mathbf{w}_i$. $\mathbf{R}$ is the residual unexplained by the model.

To identify the number of muscle synergies needed to reconstruct the EMGs adequately, we successively increased the number of synergies extracted from one to the number of muscles recorded and selected the minimum number of synergies required for an EMG-reconstruction R^2 of 80%. The R^2 was calculated as follows,

$$R^2 = 1 - SSE/SST \qquad (2)$$

$$SST = \sum_{ij} (D_{ij} - mD_i)^2 \qquad (3)$$

$$SSE = \sum_{ij} (D_{ij} - [\mathbf{W}\mathbf{C}^T]_{ij})^2 \qquad (4)$$
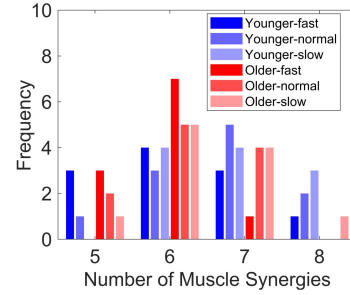

Fig. 2. The distribution of the dimensionality of the muscle synergy sets (i.e., the number of synergies) for both groups at the three speeds.

where SST is the sum squared total, D_{ij} is the EMG data of the ith muscle at the jth time point, mD_i is the average EMG value of the ith muscle, and SSE is the sum squared error.

To prevent the extracted synergy set from respresenting a suboptimal local minimum on the error surface, at each number of synergies, NNMF was run 20 times, each time with different initial parameters which were uniformly distributed between 0 and the maximum EMG amplitude. The run with the highest R^2 was selected for further analyses. For both the younger and older groups, the numbers of synergies thus selected ranged from five to eight (Fig. 2).

D. Clustering Muscle Synergies

To characterize the muscle synergy patterns of both groups, the synergies of all subjects in both groups were k-means clustered together using Matlab. We verified that analogous results were obtained when the synergies of the two groups were separately clustered. The algorithm was initialized with random cluster centroids. The number of clusters was determined as the smallest number that yielded a local maximum of average silhouette value (across 1 to 14 clusters). Each run of the k-means algorithm was repeated 100 times to ensure robustness in our determination of the number of clusters. To quantify the between-group synergy similarity at the population level, the scalar product (SP) between every pair of synergy vectors from both groups was calculated in each cluster; the SP of synergy pairs within the younger and older groups were also calculated as baselines. The scalar product ranges from 0 to 1, with 1 indicating two identical vectors and 0 indicating two vectors with no correlation.

E. Variability of Synergies' Temporal Activations

The variability of synergies' activation coefficients across subjects in each group was evaluated after the muscle synergies shared by both age groups were extracted. In this way, we ensured that the temporal activations from the two groups being compared were coefficients for the exact same set of basis vectors. Pre-processed EMGs of all subjects of all three speeds in both groups were concatenated into a single EMG matrix, which was then factorized using NNMF. The minimum number of synergies that yielded an EMG-reconstruction R^2 of 80% was selected as the number of synergies for this analysis.

After extracting the shared synergies, the temporal activation of each synergy was segmented into individual gait cycles using the timings of heel strike and toe off for each subject [27], with each cycle time-normalized and resampled
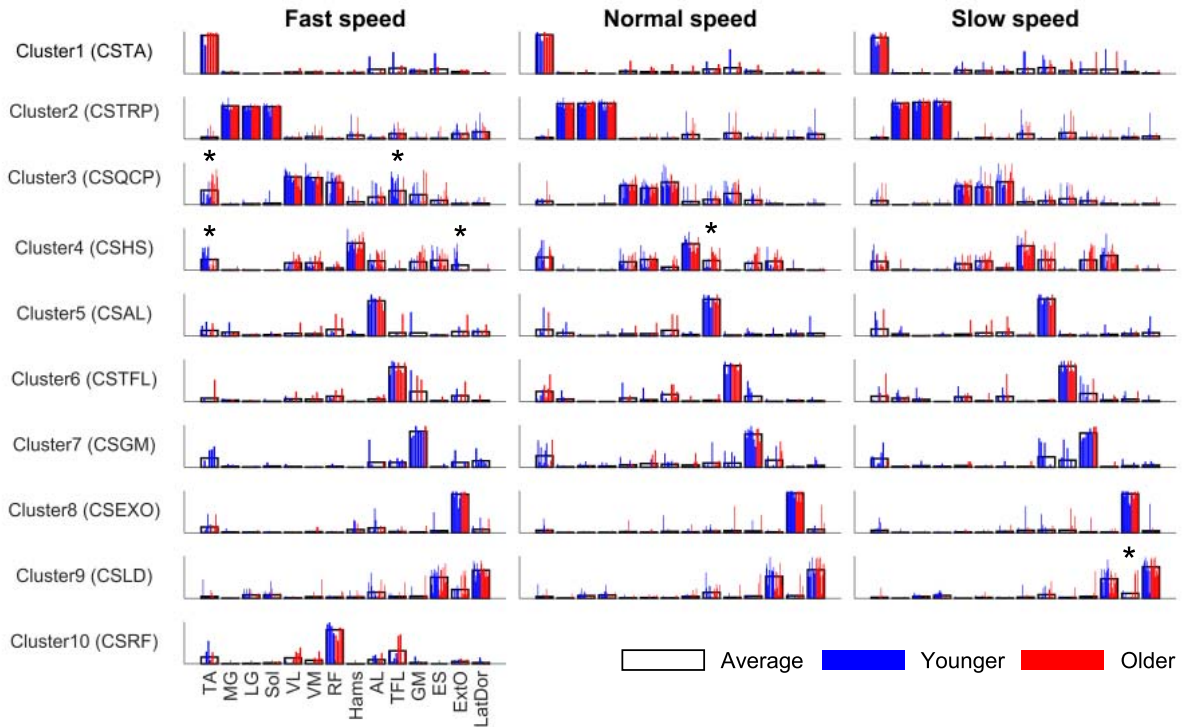
Fig. 3. Cluster analysis of muscle synergies during overground walking at three self-selected speeds for both age groups. 10, 9 and 9 clusters were identified for fast, normal and slow speed, respectively. Despite between-group differences in some specific muscle components (marked with ∗, $p < 0.05$; ANOVA) and an additional 10th cluster involving RF (CSRF, CS standing for cluster of synergy) in fast speed, all the nine clusters have grossly similar synergies between the two age groups.
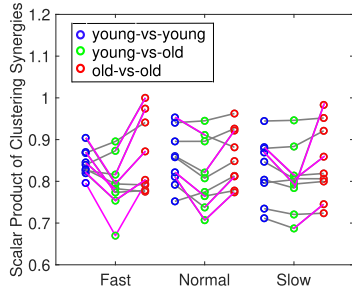

Fig. 4. The similarity of muscle synergies in each cluster. In each cluster, scalar product values were calculated for young-old, young-young and old-old synergy pairs, respectively, with their means connected by a continuous line. The significant differences are shown as pink lines ($p < 0.05$, ANOVA).

into 200-time points (time points: 1-100 for stance; 101-200 for swing), and then averaged across cycles. Each time point of the resampled temporal activation was regarded as an independent variable, and the variance of each time point across subjects was calculated. We referred to this variance as inter-subject variability of the temporal activations.

F. Sparseness of Muscle Synergy

Following earlier works [14], [28], we also explored whether the sparseness of the synergy vector, may be related to other attributes of the synergy or its coefficient. The sparseness of each synergy vector was evaluated by:

$$sparseness(\mathbf{x}) = \frac{\sqrt{n} - (\sum |\mathbf{x}_i|)/\sqrt{\sum \mathbf{x}_i^2}}{\sqrt{n} - 1}$$

where $\mathbf{x}_i$ is the ith muscle component of synergy $\mathbf{x}$, and n is the number of EMG channels. Based on this definition, a vector with only one active component is a sparse vector with a sparseness of 1 while a non-sparse vector with the same value in all components has a sparseness 0.

G. Statistical Parametric Mapping

For every synergy, we applied statistical parametric mapping (SPM) to further compare the temporal activations (averaged across subjects) between the subject groups. The SPM is a topological methodology for detecting field changes in smooth n-dimensional continuous signals. It detects the regions of interest of the continuous signals [29] by applying statistical tests, such as subtraction, correlation, regression, t-test, and ANOVA. The SPM is frequently used in the statistical analysis of neuroimaging voxel data for functional mapping and functional connectivity investigations [30]. Here, we employed the SPM in a fashion analogous to that used in neuroimaging to identify the temporal intervals of the temporal activations with significant between-group differences ($p < 0.05$).

III. RESULTS

A. Some Hip-Related Muscle Synergies Tended to Be More Age-Group Specific

We began by verifying that there was no significant difference between the body-height normalized walking speed of the two age groups at all three self-selected speeds (Fig. 1). Thus, any between-group differences in synergies or kinematics should more likely originate from ageing effects rather than
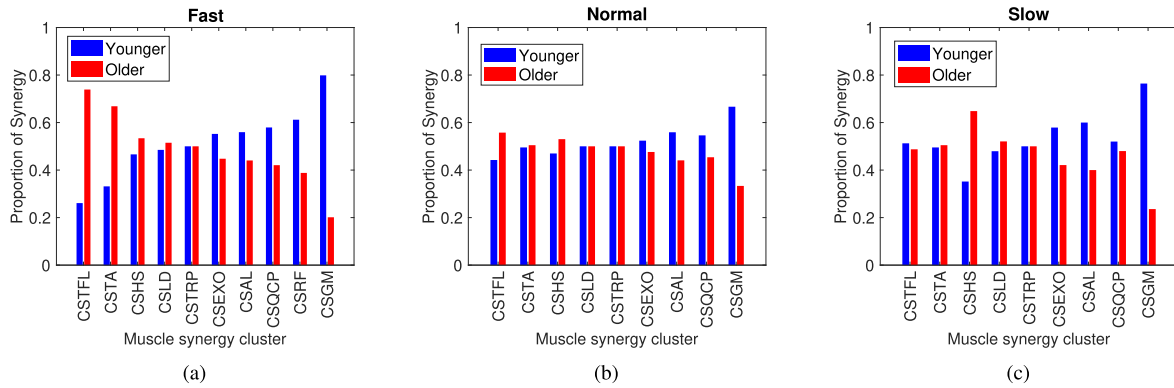
Fig. 5. The proportion of synergies belonging to both groups at self-selected fast (a), normal (b), and slow speed (c) in the synergy clusters. Across all clusters, the younger group had the highest proportion in the cluster involving GM at all three speeds while the older group was better represented in clusters involving TFL and Hams at fast and slow speed, respectively.

gait speed difference. After extracting the muscle synergies of each subject, for each gait speed, we k-means clustered the synergies of all subjects of both groups to assess if there was any age-related change in the muscle synergies for overground walking. For all walking speeds, the synergies of both groups could be grouped into 9-10 clusters, with 1 cluster involving muscle RF specific to fast walking (Fig. 3). Within individual clusters, between-group differences were speed-, cluster-, and muscle-specific (e.g., ExtO in cluster 9, slow speed; TA and ExtO in cluster 4, fast speed; and AL in cluster 4, normal speed) (Fig. 3). For all walking speeds and each cluster, we compared the similarity of young-versus-old synergy pairs against baseline similarity from within-young and within-old synergy pairs (Fig. 4). The young-versus-old similarity was significantly lower than both baselines in 3 clusters for fast speed, 1 cluster for normal speed, and none in slow speed, thus further supporting the result that there were speed-specific between-group differences in some of the synergy clusters.

Beyond the muscle- and speed-specific between-group differences noted above, synergies of the two groups were also different in the sense that some clusters comprised more synergies from one of the age groups. Fig. 5 shows the proportions of synergies from the younger (blue) and older (red) groups within each synergy cluster. For fast speed, a cluster involving TFL (CSTFL) was much more frequently identified in the older (74%) than younger group (26%) (Fig. 5(a)); another involving GM (CSGM) was much less frequently identified in elderlies (20%, 33%, 24%) in all speeds. For slow walking, a Hams-related cluster (CSHS) was likewise more noticeable in the older group (65%) (Fig. 5(c)). Note that TFL, GM, and Hams are all related to hip motion, and Hams to knee motion as well. We therefore proceeded to examine the hip- and knee-joint kinematics of the two age groups in more detail.

B. The Range of Hip Motion Differed Between Age Groups

To investigate if the between-group differences in muscle synergies noted above may correlate with differences in gait biomechanics, we compared the gait kinematics between the two age groups. Indeed, over a gait cycle, older adults had higher hip flexion angle but lower hip extension angle when

compared with younger subjects at all speeds of overground walking (Fig. 6). The peak hip flexion of the older group was significantly higher than that of the younger group at all speeds (Fig. 7(a)) whereas the peak hip extension of the older, significantly smaller than those of the younger (Fig. 7(b)) ($p < 0.05$, ANOVA). On the other hand, knee flexion was not significantly different between two groups.

C. At Normal and Slow Speeds, Older Adults Exhibited Higher Inter-Subject Variability in Temporal Activations

To compare the muscle synergies' temporal activations between the two groups, we enforced the EMGs of both groups to be explained by the same set of synergies (Fig. 8, column 1) so that for each synergy, the activations being compared represented coefficients for the same basis vector. This enforcement is justified given the synergies of the two groups were grossly similar (Fig. 3, 4). As shown in Fig. 8, the average temporal activation coefficients of the two groups were different in amplitude at certain phases of the gait cycle in the synergies involving GM (SGM) and TA (STA) (phases with significant between-group differences are highlighted in grey ($p < 0.05$, SPM)). To further contrast the temporal activations of the two groups, we also calculated and compared the across-subject variance of the coefficients at each time point of both groups. For the normal and slow speeds, the inter-subject temporal-activation variability of the older adults was significantly higher than that of younger subjects in 6 synergies involving the TA (STA), triceps surae (STRP), quadriceps (SQCP), Hams (SHS), LatDor (SLD), and TFL (STFL), respectively ($p < 0.05$, ANOVA) (Fig. 9). To further confirm the validity of this analysis, we compared the variances of both groups after excluding an older-group subject whose speed was an outlier. The results for fast and slow speeds remained the same as before, but the differences in SQCP and SHS at normal speed became insignificant after outlier exclusion.

For completeness, we also considered the intra-subject cycle-to-cycle variability of the temporal activations (Fig. 10). The intra-subject temporal-activation variability of the elderlies was statistically higher than that of young adults in SGM at fast speed, STRP and SHS at normal speed, and STA at
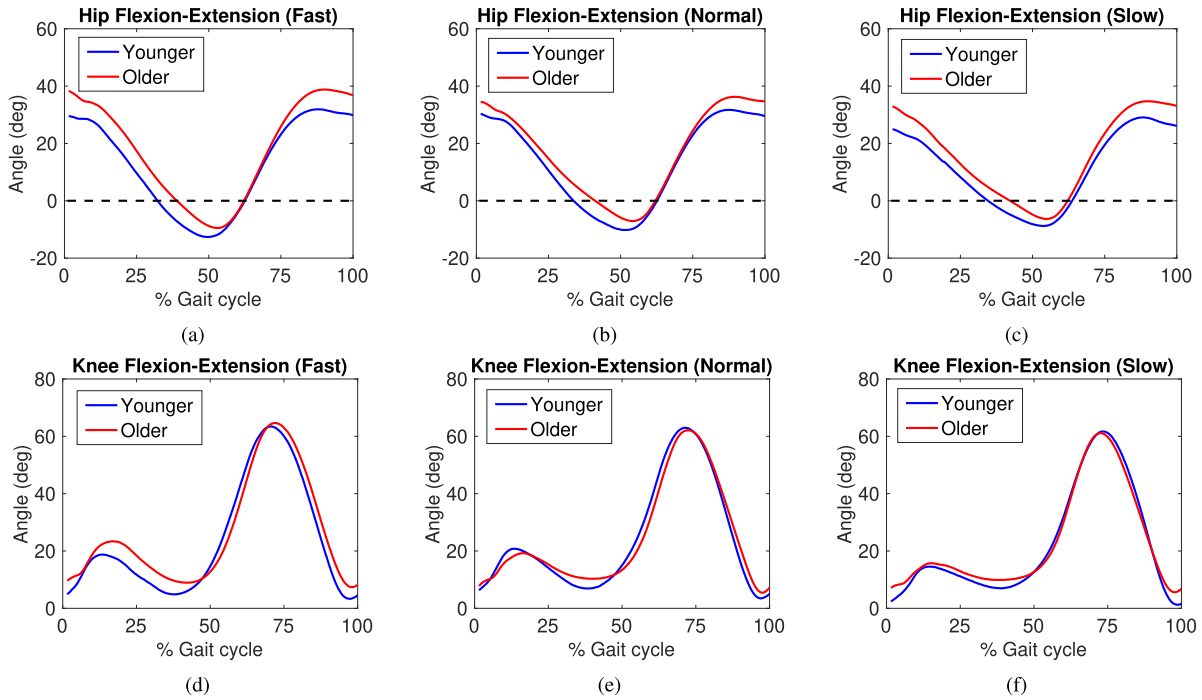
Fig. 6. The 2D-sagittal projection of the hip and knee flexion-extension angles in a gait cycle for both age groups. For hip motion, positive angles correspond to flexion and negative angles to extension.
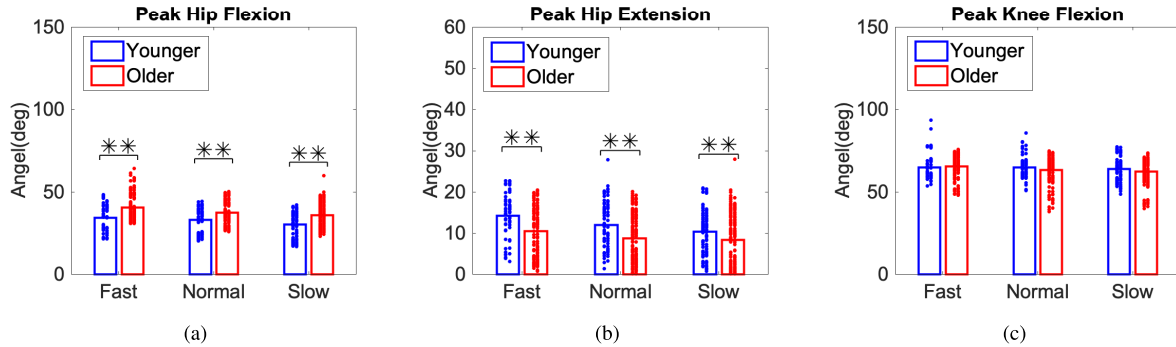

Fig. 7. Peak hip flexion, hip extension and knee flexion (the highest joint angle in the phase of gait) in each gait cycle for both age groups. The older group had significantly higher hip flexion (a) but lower hip extension (b) compared with the younger group ($* p < 0.05$, $** p < 0.01$, ANOVA).

slow speed ($p < 0.05$, ANOVA), but lower in SEXO, SLD at normal and slow speeds ($p < 0.05$, ANOVA).

D. Negative Correlations Between **W**-Sparseness and **C**-Variability

We sought to investigate further what properties of the muscle synergies (**W**) may be related to the inter- or intra-subject variability of the temporal activations (**C**). Inspired by previous works that characterized **W** by the sparseness of the synergy vectors [14], [28], we correlated the sparseness of the shared synergy vectors (Fig. 8) with the inter-subject variability of the temporal activations of the same synergies. Surprisingly, we found a significant negative correlation at slow speed in the older group ($r = -0.84$, $p = 0.001$, Pearson's r) but not in the younger group ($p = 0.8$, Pearson's r) (Fig. 11). Such negative correlation between the sparseness of **W** and the variance of corresponding **C** became more obvious when the within-subject cycle-to-cycle variability of

the temporal activations was considered across all subjects. In both age groups and at all gait speeds, the **W**-sparseness and **C**-variance of the synergies extracted from the subjects exhibited statistically significant negative correlations (Fig. 12) (young, (fast, $r = -0.48$, $p \ll 0.05$; normal, $r = -0.53$, $p \ll 0.05$; slow, $r = -0.23$, $p = 0.03$); old, (fast, $r = -0.30$, $p = 0.01$; normal, $r = -0.52$, $p \ll 0.05$; slow, $r = -0.44$, $p \ll 0.05$), Pearson's r). Similarly, across all gait speeds and across subjects, the across-time variance of the temporal activations of each synergy ($\text{Var}(\mathbf{C}_t)$) also correlated negatively with the sparseness of the same **W** in both age groups (Fig. 13) (old, $r = -0.78$, $p \ll 0.05$; young, $r = -0.79$, $p \ll 0.05$, Pearson's r). For completeness, We also performed additional analyses on finding potential correlations between the sparseness of synergies and their activation peak and average activations, respectively. Our results show that for activation peak, significant correlations were found in both groups at all three speeds, but for average activations, only at the fast speed of the younger group.
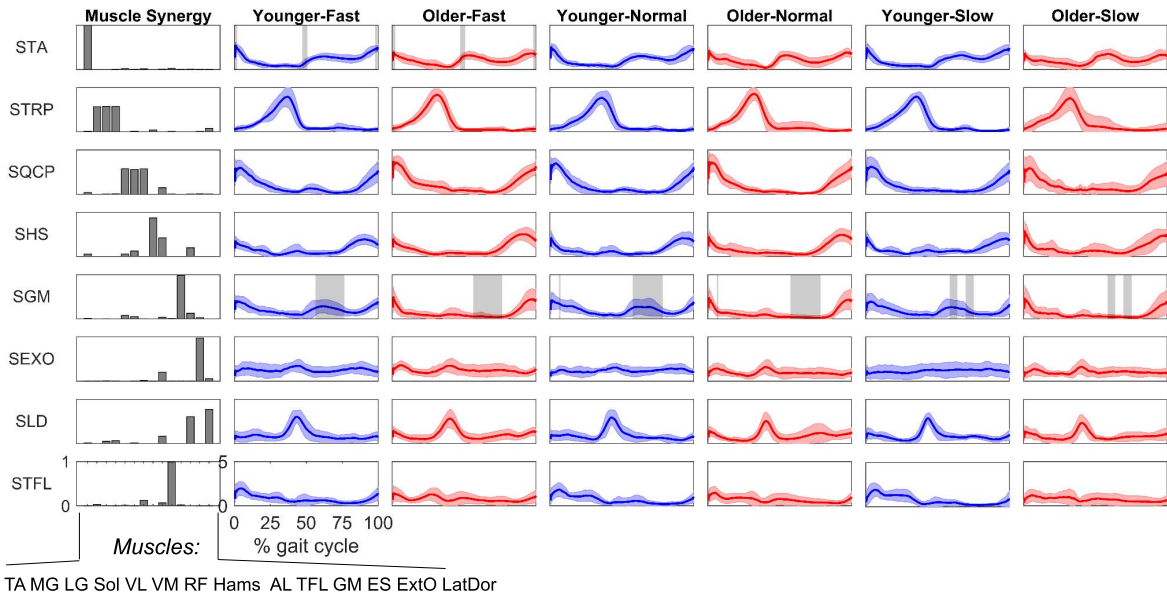
Fig. 8.   Shared muscle synergies and their temporal activation coefficients. The muscle synergies shared between both age groups are shown in the first column and their corresponding temporal activations at three speeds for young and old adults are shown in the remaining columns. Solid curves are the average temporal activations across subjects for a single gait cycle and the red/blue shadow of curve represents the standard deviation of activations. Between-group differences of activations (grey shadow, $p < 0.05$, SPM) in certain phases of a gait cycle were found at fast speed (row 1, column 2, 3) for STA (synergy involving TA), and all three speeds for SGM (row 5, column 2-7).
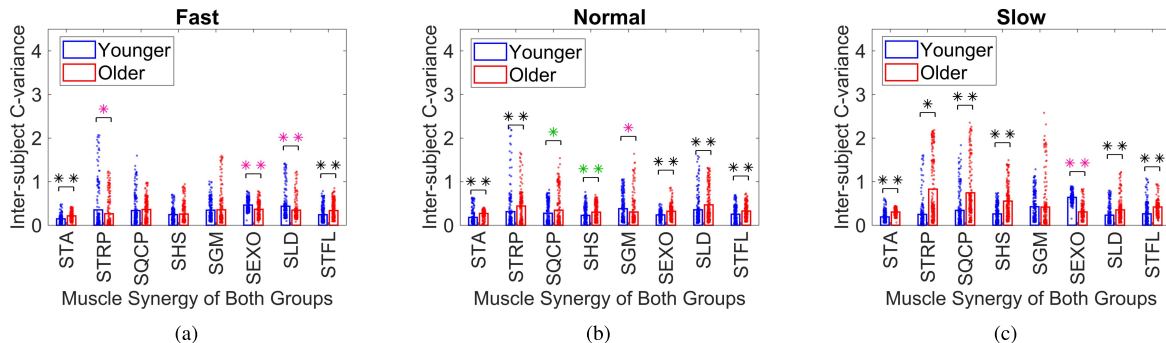

Fig. 9.   The variance of temporal activations across subjects ($*$ $p < 0.05$, $**$ $p < 0.01$, ANOVA). The instances with the younger group showing higher and significant inter-subject C-variance are marked with pink stars. The differences that lost statistical significance after outlier exclusion are highlighted with green stars.

IV. DISCUSSION

A. Age-Related Modifications of Locomotor Muscle Synergies

Our k-means clustering identified 9 basic locomotor muscle synergies that were utilized by both age groups for overground walking at different self-selected gait speeds (Fig. 3). At fast speed, one additional synergy (CSRF) was recruited, agreeing with the previous result that in young male subjects, the number of synergies activated was proportional to the instantaneous speed of treadmill locomotion [31]. Nevertheless, excepting CSRF and other cluster- and speed-specific between-group differences in certain muscles components, overall, the observed muscle synergies were grossly similar across age groups and speeds. The gross structure of the synergy clusters identified here may therefore represent elementary building blocks of locomotor patterns that are sufficient for generating overground walking at a range of speeds and conditions [32], [33]. Recent data from rodents have suggested that the basic locomotor muscle synergies are encoded in the spinal cord, developed early in life, and preserved into adulthood [15], but are nonetheless subject to being fine-tuned during development, possibly through supraspinal modulation of the spinal synergy-encoding networks [15], [32]. The consistency of the overall compositions of the synergies observed here agrees well with this result, but the small between-group differences noted (Fig. 3) may reflect age-related fine-tunings of the synergies structures already reported in the rodents.

Beyond the small amplitude differences in certain muscle components, the synergies of the younger and older groups were also different in the sense that certain synergy clusters were dominated by synergies from one of the two age groups (Fig. 5). The clusters involving TFL (CSTFL) and Hams (CSHS) included more older-subject synergies at fast and slow speeds, respectively, but another cluster involving GM (CSGM) was dominated by younger-subject synergies at all speeds. We can infer that at the population level, older subjects are more likely to utilize CSTFL and CSHS than younger subjects, while younger adults are more likely to use CSGM
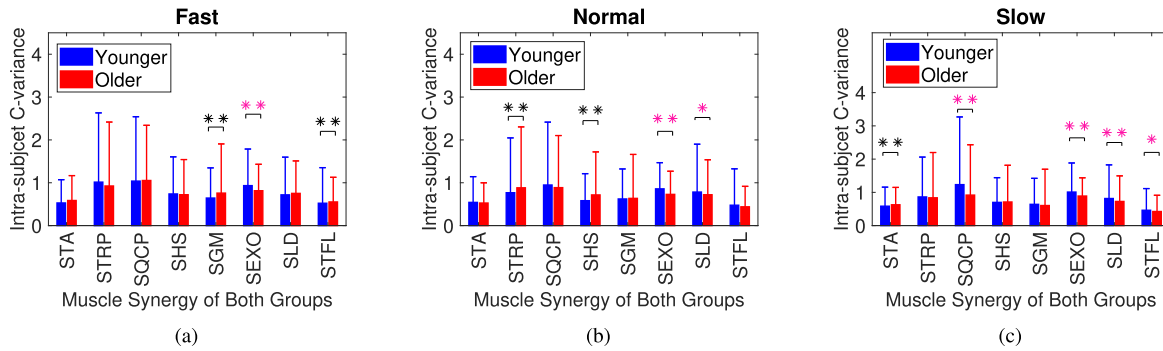
Fig. 10. The within-subject variance of temporal activations across gait cycles ($*$ $p < 0.05$, $**$ $p < 0.01$, ANOVA). The instances with the younger group showing higher intra-subject C-variance are marked with pink stars.
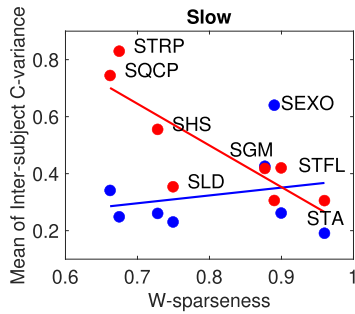

Fig. 11. Correlation between W-sparseness and inter-subject C-variance. The red line is the linear regression for the older group ($r = -0.84$, $p \ll 0.05$, Pearson's r) while the blue line for young adults ($r = 0.21$, $p = 0.8$, Pearson's r).
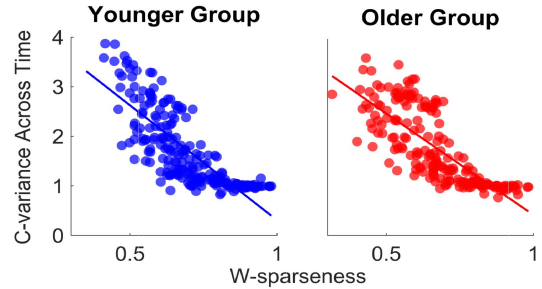

Fig. 13. Correlation between W-sparseness and Var($\mathbf{C}_t$). Each line is the linear regression of W-sparseness and the across-time variance of the synergies' corresponding Cs across all subjects and speeds (young, $r = -0.79$, $p \ll 0.05$; old, $r = -0.78$, $p \ll 0.05$, Pearson's r).
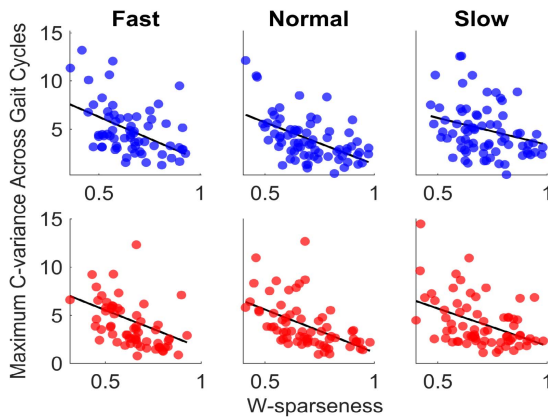

Fig. 12. Correlation between W-sparseness and intra-subject C-variance. Each line is the linear regression of W-sparseness and the within-subject maximum C-variance across gait cycles (blue: young (fast, $r = -0.48$, $p \ll 0.05$; normal, $r = -0.53$, $p \ll 0.05$; slow, $r = -0.23$, $p = 0.03$); red: old (fast, $r = -0.30$, $p = 0.01$; normal, $r = -0.52$, $p \ll 0.05$; slow, $r = -0.44$, $p \ll 0.05$), Pearson's r).

during overground walking. Consistent with our results, a previous study [16] assessed age-related modifications of the synergies for step ascent and likewise found subtle differences between the synergies of the younger and older groups, with the latter relying on somewhat more complex synergy patterns. It is therefore likely that even for locomotion, the muscle weightings of the synergies are fine-tuned or reshaped as people age, with the times of change coinciding with the maturation of the neuromusculoskeletal system, and with

the synergy-encoding networks being constantly adjusted by sensory and descending signals [34] throughout the lifetime.

Interestingly, the age-specific synergy clusters are related to muscles TFL, GM, and Hams which are hip flexor, hip extensor, and knee flexor, respectively. From this observation, we can infer the potential of the Hams to compensate GM for weak hip extension during gait. This inference agrees with the previous result that biceps femoris (whose long head is a part of Hams) contributes to stance hip extension in the presence of a weak GM [35]. Also, the prevalence of the TFL synergy in the older group during fast walking may reflect the use of TFL for generating additional hip flexor torque due to weakness of other hip flexors (e.g. iliacus and psoas) in older subjects.

B. Origins of Age-Related Muscle Synergy Modifications

The identified changes in the muscle synergies may reflect age-related changes in the biomechanical requirements of walking. Certainly, ageing is associated with changes in biomechanical properties [3], [36], [37], which may impose a different set of biomechanical constraints to functional gait [12], thus necessitating muscle synergy modifications [38]. In our findings, at all speeds, the older subjects had larger ranges of hip flexion but smaller ranges of hip extension (Fig. 7). Previously, Judge *et al.* found that elderlies tended to use more hip flexor power to compensate for the insufficient ankle plantarflexor power to ensure gait performance [3], [4]. The between-group difference in the frequencies of use of the hip-related muscle synergies we report here (Fig. 5) may well reflect such compensations occurring in most, but not all of the older subjects. Changes in the gait biomechanical constraints

may also change the biomechanical functions subserved by the same muscle synergies. For instance, muscle GM typically provides stability to the sacroiliac joint during walking [39]; however, its function and properties would be altered when changes occur as a result of joint injury. Specifically, people with patellofemoral pain syndrome have a shorter activation duration of GM muscle activity when running compared to normal people [40].

Alternatively, the observed muscle-synergy modification may result from age-related changes in the central nervous system (CNS). It has been shown that age-related structural and chemical changes of the motor regions in the brain lead to the use of different functional networks even for simple motor skills [41]. Also, the spinal motor circuitries, and in particular the arrangements of the reflex pathways, are certainly modified in older humans [42]. Gradual age-related damage of spinal interneurons due to environmental factors has also been reported [43]. All of the above could potentially account for the muscle-synergy changes reported here.

It should be noted that the state of the CNS, the structures of the muscle synergies deployed, the biomechanical functions subserved by the synergies, and limb biomechanical properties all mutually influence each other. The changes in the synergies originating from age-related alterations in neuronal circuitry would affect gait kinematics, which may elicit other necessary compensatory changes in the synergies. Meanwhile, long-term physical changes of the legs may also induce reshaping of the muscle synergies through the sensory afferents [44].

C. Elderlies Have Higher Inter-Subject Variability of the Synergies' Temporal Activations

As an individual grows from a toddler to an adult, early muscle synergies are fine-tuned to accommodate the developing neuro-musculoskeletal system, and the precise temporal activations of the locomotor synergies are also reshaped gradually [14], [45]. This gradual sculpting of the activations is presumably also dependent on the individual's history of sensorimotor experience and motor learning [14], [17], [32], [46]. Our result here reveals a higher inter-subject variability of the synergies' temporal activations in elderlies (Fig. 9), indicating a higher heterogeneity of locomotor control strategies within the older cohort. The result may simply be the consequence of the older subjects having had more years of life than the younger ones for accumulating different patterns of motor-control adjustments from their variable sensorimotor histories or different levels of degeneration that altered neuromuscular control. Our result also agrees with the conclusion of Baggen *et al.* [16], that the organization or activation timing of the synergies for step ascent may be altered through the lifetime, thus inevitably leading to higher across-subject variability in the older group.

D. Negative Correlation Between Sparseness of Synergies and Variability of Their Temporal Activation

Muscle synergies and their temporal activation were extracted from the EMGs using NNMF. While the synergies have been interpreted as neuromotor modules that correspond to how discrete spinal or cortical premotor networks co-activate the motoneuronal pools of multiple muscles, the temporal activation may reflect the dynamic neural activities that drive the recruitment of these networks [32]. The sparseness of the synergy vectors studied here, on the other hand, quantifies the degree of muscle co-activations in each motor module. The synergy with the highest sparseness would involve the activation of only 1 muscle, while the synergy with the lowest sparseness, co-activation of all recorded muscles. Presumably, the synergy's sparseness should reflect the connectivity between the premotor neurons encoding the synergy and the moto neurons [10].

As an attempt to relate properties of the synergy vectors ($\mathbf{W}$) to characteristics of their temporal activations, we correlated $\mathbf{W}$-sparseness with both the inter- and intra-subject variability of $\mathbf{C}$ (Fig. 11, 12, 13) and surprisingly found a negative correlation between them. To the best of our knowledge, our finding is the first demonstration that variability of the synergies' drives could be related to the numbers of muscles coordinated by the synergies. Thus, synergies with lower sparseness values (i.e., more muscle components) have more diverse temporal activations, both within and across subjects. We speculate that the premotor networks that coordinate larger numbers of muscles are also susceptible to modulation by feedback signals coming from more muscles, thus giving them greater variability of activations. Such feedback modulation can be underpinned either by intraspinal reflex pathways or long-loop reflex circuits that involve the descending pathways. Indeed, it has been shown that synergy-encoding premotor interneurons are directly contacted by both proprioceptive and descending synaptic terminals [47]. In one of our recent works, we showed that muscle synergies that exhibit higher variability in their activations might play a more important role in driving changes in motor outputs during early motor skill learning [17]. Therefore, our demonstration of the negative correlation between synergy sparseness and activation variability implies that the synergies that are less sparse may play a more critical role in helping the CNS arrive at the appropriate motor outputs during the initial phase of locomotor adaptation or gait retraining. Whether muscle synergies that are less sparse should represent better targets of intervention awaits further study. We do not know the functional implications of this arrangement. Perhaps it reflects how motor-output variability is maximized for functional flexibility when the outputs themselves are constrained by the structures of the muscle synergies and the connectivity of the premotor networks.

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