Statistical Parametric Mapping (SPM) for alpha-based statistical analyses of multi-muscle EMG time-series

Mark A. Robinson^a, Jos Vanrenterghem^a, Todd C. Pataky^b

a Research Institute for Sport and Exercise Science, Liverpool John Moores University, UK b Department of Bioengineering, Shinshu University, Japan

Journal of Electromyography and Kinesiology

doi:10.1016/j.jelekin.2014.10.018

Abstract

Multi-muscle EMG time-series are highly correlated and time dependent yet traditional statistical analysis of scalars from an EMG time-series fails to account for such dependencies. This paper promotes the use of SPM vector-field analysis for the generalised analysis of EMG time-series. We reanalysed a publicly available dataset of Young versus Adult EMG gait data to contrast scalar and SPM vector-field analysis. Independent scalar analyses of EMG data between 35-45% stance phase showed no statistical differences between the Young and Adult groups. SPM vector-field analysis did however identify statistical differences within this time period. As scalar analysis failed to consider the multi-muscle and time dependence of the EMG time-series it exhibited Type II error. SPM vector-field analysis on the other hand accounts for both dependencies whilst tightly controlling for Type I and Type II error making it highly applicable to EMG data analysis. Additionally SPM vector-field analysis is generalizable to linear and non-linear parametric and non-parametric statistical models, allowing its use under constraints that are common to electromyography and kinesiology.

Keywords: biomechanics, multivariate statistics, vector-field analysis, random field theory, experimental error.

Corresponding Author:

Mark Robinson m.a.robinson@ljmu.ac.uk

Tom Reilly Building, Byrom Street Campus, Liverpool, Merseyside, L3 3AF, United Kingdom.

1. Introduction

EMG waveforms are complex time-series signals that describe localised electrical activity of individual muscles. The synthesis of multi-muscle time-series is common as it provides insight into motor control [Gribble and Ostry, 1998, 1999], clinical pathology [Frigo and Crenna, 2009], sports performance [Trevithick et al., 2007] and musculo-skeletal simulations [Hamner et al., 2010, Thelen et al., 2003]. Often in the above contexts, due to the exploratory nature of biomechanical research, no specific hypotheses are made regarding either individual EMG time-series or their temporal characteristics. Instead, in classical hypothesis testing a "non-directed null hypothesis" [Pataky et al., 2013] such as "there are no differences between Young and Adult EMG time series during gait" is tested. The consequences of such hypotheses require that all EMG signals should be statistically evaluated across the whole time-series (e.g. a gait cycle) as the hypothesis pertains neither to a specific muscle or time point. In contrast to this, classical hypothesis testing of EMG time-series tends to involve the extraction of summarizing scalar parameters of individual muscles [Houck, 2003] and qualitative interpretation [e.g. Bovi et al., 2011, Koshland et al., 2005]. Scalar or qualitative analyses fail to consider the characteristics of inter-muscle dependence or time dependence in EMG time-series.

Inter-muscle dependence: Inter-muscle dependence is evidenced by inter-muscle covariance and has been extensively illustrated by the effective management of, for example, inter-muscle co-activation [Gribble and Ostry, 1998], net joint moments [Gribble and Ostry, 1999] and multi-muscle synergy [d'Avella et al., 2003]. The independent statistical treatment of scalar values from EMG waveforms is unable to account for the complexity of multi-muscle EMG time-series as it fails to consider inter-muscle covariance. Whilst mean single muscle EMG time-series have their own inherent variability, inter-muscle time-series may also co-vary (Figure 1). If single-muscle variance was much larger than inter-muscle (co-) variance for example, it is

unlikely that scalar analysis would detect this. Hypothesis testing methods that omit covariance are inherently biased because they fail to consider inter-muscle dependence.

Time dependence: The evidence for time dependence is based on coordinated joint movements, which are the consequence of smooth synergistic muscle-tendon forces. The smoothness of coordinated motion results from the time-dependent activation of individual motor units. The combination of a sequential recruitment of muscle fibers [De Luca et al., 1982] and biological elasticity allows smooth muscle forces to be generated. Raw EMG signals themselves are not smooth, so well established signal processing techniques are used to reduce the noise associated with signal acquisition and to better represent underlying muscle forces, although the quality of the representation is influenced by many factors [Disselhorst-Klug et al., 2009]. Time-dependence is therefore manifest in smooth EMG time-series which, from a statistical perspective, implies non-random temporal neighborhood covariance. Hypothesis testing of single-instant parameters and integrals are also therefore biased because they disregard time-dependence [Pataky et al., 2013].

Qualitative interpretation or scalar extraction are, of course, not exclusive EMG analysis methods. Other more complex analyses of EMG time-series include principal component analysis [Brandon *et al.*, 2013], cross-correlation [Wren *et al.*, 2006] or wavelet transform for time/frequency analysis [von Tscharner, 2000]. These methods may consider inter-muscle and/or time dependence yet these methods do not directly provide the necessary objective statistics with which a non-directed null hypothesis could be rejected.

In contrast to qualitative interpretation and scalar extraction, Statistical Parametric Mapping (SPM) [Pataky *et al.*, 2013] regards the multi-muscle EMG signal, subsequently referred to as the EMG vector-field, as the sole unit of observation. This allows both inter-muscle and time dependence to be incorporated directly into statistical testing. Moreover, and in distinction

even to other methods, SPM exploits the well-documented probabilistic behaviour of smooth Gaussian continua [Adler & Taylor, 2007], provides tight control of Type I and Type II statistical error, and provides an objective framework for hypothesis evaluation.

The purpose of this paper is to provide a statistical solution for the objective classical hypothesis testing of such non-directed null hypotheses in multi-muscle EMG time-series. Through alignment of EMG time-series analysis with SPM by testing the hypothesis "there are no differences between Young and Adult EMG time series during gait", we intend to show that bias associated with scalar EMG analysis can be mitigated. Using public data, we illustrate that the Gaussian vector-field more appropriately models variance in multi-muscle EMG time series than do scalar summary metrics. We specifically aim to (a) promote a new understanding of the EMG vector-field as the indivisible unit of observation, and (b) describe a new technique for comprehensive, generalized EMG analysis.

2.0 Methods

We considered the public dataset of Bovi *et al.* (2011). This is a comprehensive dataset including 3D multi-joint kinematics, kinetics and EMG for a variety of gait-related tasks from 40 healthy subjects subcategorised into 20 "young" (aged 6–17) and 20 "adult" (aged 22–72). Present focus was on mean EMG time-series of the Anterior Tibialis, Soleus, Gastrocnemius Medialis and Peroneus Longus muscles (Figure 2) calculated from their walking trials (as labelled N, XS, S, M, L in their supplementary data file). Four muscles were chosen for brevity. We filtered the data using simple convolution and a relatively narrow Gaussian kernel (FWHM=2.0%, SD= 4.7%). Parameters for this simple filter were selected iteratively, through qualitative visualisation to maximise group trajectory divergence. While simple, post-hoc analyses found qualitative effects of neither filtering parameters nor methods,

including more common bandpass filtering methods. While we acknowledge that filtering choices can affect physiological interpretations of EMG data [Hodges and Bui, 1996], the goal of this paper was not to make physiological conclusions but rather to highlight potential problems with using traditional (0-D) hypothesis testing to make inferences regarding general 1-D EMG data.

2.1 Scalar Analysis

To test the null hypothesis we exemplified a qualitative/scalar analysis by selecting ten scalar values from each EMG time-series at 35-45% gait cycle, which was the region where there appeared to be the greatest qualitative difference between groups. These scalar values were then statistically compared using a two-sample t-test with one test for each instance in time and for each muscle separately. To retain a Type I family-wise error rate of $\alpha = 0.05$ we adopted a Šidák corrected threshold of 0.012 for the comparison of four muscles. We did not correct alpha for the ten time points because we chose ten time points for illustrative purposes; a typical scalar analysis would examine one time point only.

2.2 Statistical Parametric Mapping (SPM)

We used SPM to test the null hypothesis by statistically examining the whole EMG time-series. All SPM analyses were implemented in Python 2.7 using Canopy 1.1 (Enthought Inc., Austin, USA). Conceptually, the SPM analysis process is similar to the calculation and interpretation of a scalar two-sample t-test. Importantly however we employ a SPM Hotelling's T^2 statistic to account for covariance between the EMG time-series (Figure 1), A SPM Hotelling's T^2 test is the vector-field equivalent to the two-sample t-test [Cao & Worsley, 1999; Pataky $et\ al.$, 2013]. The EMG time-series were analysed as a four-component vector-field I=4, J=40, Q=101, where I, J and Q were the number of vector components, responses and time points respectively.

(1)
$$SPM\{T^2\} \equiv T^2(q) = \frac{J_1 J_2}{J_1 + J_2} \left(\overline{\mathbf{y}}_1(q) - \overline{\mathbf{y}}_2(q) \right)^{\mathsf{T}} \mathbf{W}(q)^{-1} \left(\overline{\mathbf{y}}_1(q) - \overline{\mathbf{y}}_2(q) \right)$$

(2)
$$\mathbf{W} = \frac{1}{J_1 + J_2 - 2} \left(\sum_{j=1}^{J_1} (\mathbf{y}_{1j} - \overline{\mathbf{y}}_1) (\mathbf{y}_{1j} - \overline{\mathbf{y}}_1)^{\mathrm{T}} + \sum_{j=1}^{J_2} (\mathbf{y}_{2j} - \overline{\mathbf{y}}_2) (\mathbf{y}_{2j} - \overline{\mathbf{y}}_2)^{\mathrm{T}} \right)$$

Subscripts "1" and "2" index the two groups and **W** is the pooled covariance matrix. The domain "(q)" is omitted for readability.

The scalar output statistic, SPM $\{T^2\}$, is calculated separately at each individual time point (q)and is termed a statistical parametric map. At this stage it is worth noting that SPM refers to the overall methodological approach, and SPM $\{T^2\}$ to the scalar trajectory variable. The calculation of SPM $\{T^2\}$ simply indicates the magnitude of the Young-Adult differences. therefore at this stage we do not accept or reject our hypothesis. To test our null hypothesis we next calculated the critical threshold at which only α % (5%) of smooth random curves would be expected to traverse. Like all classical hypothesis testing methods SPM produces Type I error at a rate of alpha, SPM does not prevent Type I error but tightly controls its rate of occurrence. The critical threshold calculation is based upon estimates of trajectory smoothness via temporal gradients [Friston et al., 2007] and, based on that smoothness, Random Field Theory expectations regarding the field-wide maximum [Adler and Taylor, 2007]. If any values of SPM $\{T^2\}$ exceed the critical threshold, then the EMG time-series are considered significantly different. Typically, due to waveform smoothness and the interdependence of neighboring points, multiple adjacent points of the SPM $\{T^2\}$ curve often exceed the critical threshold, we therefore call these "supra-threshold clusters". SPM then uses Random Field Theory expectations regarding supra-threshold cluster size to calculate cluster specific p-values which indicate the probability with which supra-threshold clusters could have been produced by a random field process with the same temporal smoothness [Adler and Taylor, 2007]. The calculation of cluster specific p-values demonstrates therefore

that SPM is sample-rate independent: measuring at 1 kHz or 1 GHz would not change the temporal extent of the supra-threshold cluster with respect to the field size (provided both are above the Nyquist frequency).

2.2 Post-hoc scalar field SPM

Post-hoc analysis should only take place if the vector-field SPM{ T^2 } result was significant i.e. the critical threshold was exceeded. This is comparable with the hierarchical testing procedure of ANOVA followed by post-hoc t-tests. When overall significance is achieved in the vector-field ($\mathbf{y}(\mathbf{q})$) analysis, individual vector components ($\mathbf{y}_i(\mathbf{q})$) may then be compared. In the example dataset, post-hoc analysis was conducted on individual vector component pairs using the two-sample t-test. This test initially calculates the time-varying statistical parametric map SPM{t}, the significance of which is determined in the same way as described in section 2.2. To retain a Type I family-wise error rate of $\alpha = 0.05$ for these post-hoc analyses we adopted a Šidák corrected threshold of 0.012 for four comparisons.

3.0 Results

3.1 Scalar Analysis: Two-sample t-test

Statistical testing on EMG data at 35-45% gait cycle supported the null hypothesis as no significant differences between Young and Adult EMG magnitudes were observed (table 1).

3.2 Statistical Parametric Mapping: Hotelling's T^2 test

In contrast to the scalar two-sample t-test results, SPM found a highly significant difference between the Young and Adult groups (p<0.05) (Figure 3). One supra-threshold cluster, which peaked at 43% gait cycle was identified. On this evidence, SPM analysis recommends that

the null hypothesis is rejected as significant differences between the Adult and Young groups were observed. As the supra-threshold cluster included times which were not significant in the two-sample t-test analyses, the scalar analysis interpretation therefore resulted in Type II error. The reason for the discrepancy between analyses is due to inter-muscle dependence and this is addressed further in the discussion. As SPM is not restricted to analysing discrete time points, consideration of the EMG time-series as an EMG vector-field allowed SPM to detect statistical differences, whereas all except the one chosen time point would have been ignored in the scalar analysis. As the vector-field T^2 test showed a significant difference between the Young and Adult vector-fields, post-hoc two-sample SPM $\{t\}$ tests were conducted on individual muscles. No muscles showed a statistical difference between the Young and Adult groups (Figure 4). The discrepancy between vector-field and scalar analysis results can be explained by muscle covariance (see Discussion).

4.0 Discussion

Two different statistical approaches (scalar and SPM analysis) were used to test the null hypothesis that Young and Adult EMG time series were identical. The two approaches led to different conclusions; the scalar analysis provided insufficient evidence to reject the null hypothesis whereas the SPM analysis rejected the null hypothesis. The scalar analysis, by failing to consider inter-muscle and time dependence, also led to Type II statistical error. SPM vector-field analysis by contrast considered both inter-muscle and time dependence and whilst maintaining a constant error rate of α .

4.1 Experimental (Type I and Type II) errors

Type II statistical errors occurred in the scalar analysis where the SPM analysis showed significant group differences. The reason for the discrepancy between the scalar and SPM

analyses is due to inter-muscle dependence. In the scalar analysis intra-muscle differences are small with respect to intra-muscle variance, but the inter-muscle (vector) effect is large with respect to inter-muscle covariance. Vector-field analysis considers the maximum difference between the groups using the resultant vector difference between the inter-muscle Young and Adult vector-fields (Figure 5). Considering that the magnitude of the resultant vector difference will always be greater than individual muscle components, vector-field analysis is more robust to Type II error.

In the scalar analysis, where typically only one time point is typically selected, failure to test the null hypothesis throughout the time-series meant that significant group differences could be missed at other time points. Even if EMG magnitudes were small at other time points and even if the biomechanical implications may also be small or negligible; testing the null hypothesis of equivalent Young and Adult EMG requires one to report all effects because significant effects refute the null hypothesis. To ignore low-magnitude EMG one must derive a null hypothesis which, based on biomechanical or neuromuscular rationale, justifiably pertains only to the EMG magnitudes of interest. A threshold of 10% max, for example, may or may not be theoretically justified. So although the intra-muscle effects were small, the inter-muscle effects were large enough to produce statistical differences. Simply, SPM finds statistical differences because it considers temporal covariance, as the EMG signals of the gastrocnemius medialis, peroneus longus and soleus in particular are highly time dependent (highly correlated). The difference between the Young and Adult groups was therefore stronger in the EMG vector-field than in each EMG waveform separately. The testing of nondirected hypotheses should not assume EMG waveform independence as independent scalar analysis does.

In addition to scalar analyses being susceptible to Type II error, Type I error can also be easily demonstrated because single-muscle scalar analyses often focus on particular portions

of the time series in an *ad hoc* manner [Pataky *et al.*, 2013]; this is inconsistent with an *a priori* null hypothesis of EMG equivalence. More specifically, scalar parameters assume a point-process Gaussian variance model, but Gaussian random field variance [Adler & Taylor, 2007] more accurately models variance in smooth time series [Friston *et al.*, 2007; Pataky, 2010; Pataky *et al.*, 2013]. SPM, through random field theory [Worsley *et al.*, 2004], therefore retains tighter control over both Type I and Type II statistical error.

4.2 Post-hoc testing

In this study, given the null hypothesis of no difference between Young and Adult EMG, the vector-field analysis alone sufficiently refutes the null hypothesis. If however the hypothesis pertained to individual components of the vector-field i.e. individual muscle time-series, posthoc scalar trajectory SPM analysis may be justified. The lack of significant differences in the post-hoc two-sample t-tests is not unexpected because the two-sample SPM t-test does not consider muscle covariance. In this case, the SPM post-hoc analysis paralleled the results of the two-sample t-tests which resulted in Type II error. In this case SPM post-hoc analysis provides no additional explanation for the significant vector-field analysis result which indicates that it is not one individual muscle that distinguishes between the Young and Adult groups but a combination of muscles. One may therefore prefer to formulate null hypotheses for which post-hoc data exploration is unimportant or redundant for example, "there is no significant difference between Young and Adult quadriceps EMG", which is entirely testable by vector-field analysis and needs no further investigation of individual quadriceps muscles. Hypotheses concerning individual muscles are likely better suited to scalar field SPM or would require examination of the resultant vector difference to establish which individual components contributed most to the Hotelling's T^2 statistic.

4.3 The generalisability of SPM

The applicability of SPM to EMG is vast. SPM fully supports both all linear and non-linear parametric statistical models (regression, ANOVA, MANCOVA, etc.) and their non-parametric variants [Friston $et\ al.$, 2007; Worsley $et\ al.$, 2004] as well as other statistical concepts such as the False Discovery Rate [Benjamini & Hochberg, 1995] and Bayesian inference. The two-sample analysis within this study fails to describe the applicability of vector-field EMG to the investigation of differences between muscle groups. One example would be the popular comparison of hamstrings versus quadriceps muscle activations for which a paired Hotelling's T^2 test would be suitable.

SPM has become the gold standard in neuroimaging [Friston *et al.*, 2007], and it also has the potential to standardize hypothesis testing of time-normalizable EMG waveforms, because in both cases the null hypothesis pertains to null continuum effects. Since SPM generalizes to *n*-D spatiotemporal neural [Friston *et al.*, 2007; Worsley *et al.*, 2004] and biomechanical continua [Pataky, 2010], it may also be able to unify discrete- (0-D) and high-density (2-D) EMG analyses. Grounded in RFT's expectations of smooth, random continuum behavior, SPM promises to improve our ability to objectively quantify, and therefore understand coordinated muscle activity.

5.0 Conclusions

Reanalysis of a public dataset study showed that vector-field SPM more appropriately accounts for inter-muscle dependence and time dependence which are present within EMG continua. Scalar analyses that only consider discrete values are more likely to lead to Type II error. One should therefore consider time-normalized EMG waveforms as an indivisible unit

of observation. The applicability of vector-field SPM analysis is broader than is shown in this paper and is proposed for consideration in future EMG analyses.

References

Adler RJ, Taylor JE. Random Fields and Geometry. New York: Springer – Verlag, 2007.

Benjamini Y, Hochberg Y. Controlling the false discovery rate: A practical and powerful approach to multiple testing. Journal of the Royal Statistical Society. 1995; 57(1):289-300.

Bovi G, Rabuffetti M, Mazzoleni P, Ferrarin M. A multiple-task gait analysis approach: kinematic, kinetic and EMG reference data for healthy young and adult subjects. Gait and Posture. 2011;33:6-13.

Brandon SCE, Graham RB, Almosnino S, Sadler EM, Stevenson JM, Deluzio KJ. Interpreting principal components in biomechanics: Representative extremes and single component reconstruction. Journal of Electromyography and Kinesiology. 2013;23(6):1304-10.

Cao J, Worsley KJ. The detection of local shape changes via the geometry of Hotelling's T2 fields. Annals of Statistics. 1999;27(3),925–942.

d'Avella A, Saltiel P, Bizzi E. Combinations of muscle synergies in the construction of a natural motor behavior. Nature neuroscience. 2003;6:300-8.

De Luca CJ, LeFever RS, McCue MP, Xenakis AP. Behaviours of human motor units in different muscles during linearly varying contractions. Journal of Physiology. 1982;329:113-28.

Disselhorst-Klug C, Schmitz-Rode T, Rau G. Surface electromyography and muscle force: Limits in sEMG–force relationship and new approaches for applications. Clinical Biomechanics. 2009;24:225–35.

Frigo C, Crenna P. Multichannel SEMG in clinical gait analysis: a review and state-of-the-art. Clinical Biomechanics. 2009;24:236-45.

Friston KJ, Ashburner JT, Kiebel SJ, Nichols TE, Penny WD (2007). Statistical Parametric Mapping: The Analysis of Functional Brain Images. London: Elsevier, 2007.

Gribble PL, Ostry DJ. Independent coactivation of shoulder and elbow muscles. Experimental Brain Research. 1998;123:355-60.

Gribble PL, Ostry DJ. Compensation for Interaction Torques During Single- and Multijoint limb movement. Journal of Neurophysiology. 1999;82:2310-26.

Hamner SR, Seth A, Delp SL. Muscle contributions to propulsion and support during running. Journal of Biomechanics. 2010;43:2709-16.

Hodges PW, Bui BH. A comparison of computer-based methods for the determination of onset of muscle contraction using electromyography. Electroencephalography and clinical Neurophysiology. 1996;101:511-19.

Houck J. Muscle activation patterns of selected lower extremity muscles during stepping and cutting tasks. Journal of Electromyography and Kinesiology. 2003;13:545-54.

Koshland GF, Galloway JC, Farley B. Novel muscle patterns for reaching after cervical spinal cord injury: a case for motor redundancy. Experimental Brain Research. 2005;164:133-47.

Pataky TC. Generalized n-dimensional biomechanical field analysis using statistical parametric mapping. Journal of Biomechanics. 2010;43(10):1976–82.

Pataky TC, Robinson MA, Vanrenterghem J. Vector field statistical analysis of kinematic and force trajectories. Journal of Biomechanics. 2013;46:2394-401.

Thelen DG, Anderson FC, Delp SL. Generating dynamic simulations of movement using computed muscle control. Journal of Biomechanics. 2003;36:321-8.

Trevithick BA, Ginn KA, Halaki M, Balnave R. Shoulder muscle recruitment patterns during a kayak stroke performed on a paddling ergometer. Journal of Electromyography and Kinesiology. 2007;17:74-9.

Von Tscharner V. Intensity analysis in time-frequency space of surface myoelectric signals by wavelets of specified resolution. Journal of Electromyography and Kinesiology. 2000;10(6),433-45.

Worsley KJ, Taylor JE, Tomaiuolo F, Lerch J. Unified univariate and multivariate random field theory. NeuroImage. 2004;23:S189–95.

Wren TAL, Do KP, Rethlefsen SA, Healey B. Cross-correlation as a method for comparing dynamic electromyography signals during gait. Journal of Biomechanics. 2006;39(14):2714-18.

Tables

Table 1. Statistical results from a two-sample *t*-test comparing the Young and Adult EMG amplitudes at 35-45% gait cycle for four muscles separately.

time	Gastroc. Medialis		Peroneus longus		<u>Soleus</u>		<u>Tibialis anterior</u>	
%	<i>t</i> -value	<i>p</i> -value	<i>t</i> -value	<i>p</i> -value	<i>t</i> -value	<i>p</i> -value	<i>t</i> -value	<i>p</i> -value
35	-0.62	0.56	-0.63	0.54	-1.15	0.28	0.53	0.61
36	-0.63	0.55	-0.56	0.59	-1.08	0.31	0.43	0.68
37	-0.67	0.52	-0.50	0.63	-1.00	0.35	0.40	0.70
38	-0.73	0.49	-0.46	0.66	-0.98	0.36	0.43	0.68
39	-0.82	0.44	-0.43	0.68	-0.95	0.37	0.52	0.62
40	-0.96	0.37	-0.39	0.71	-0.94	0.37	0.64	0.54
41	-1.17	0.28	-0.32	0.76	-0.93	0.38	0.78	0.46
42	-1.41	0.20	-0.22	0.83	-0.89	0.40	0.92	0.38
43	-1.70	0.13	-0.08	0.94	-0.84	0.42	1.10	0.31
44	-1.94	0.09	0.08	0.94	-0.81	0.44	1.30	0.23
45	-2.09	0.07	0.21	0.84	-0.76	0.47	1.51	0.17

Figures

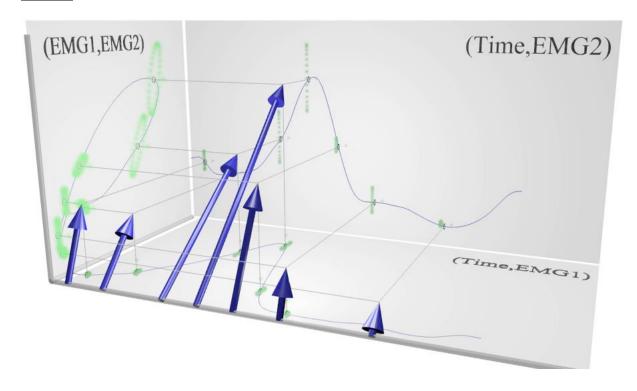


Figure 1. Vector-field schematic, depicting a mean two-muscle EMG waveform in blue (Young: gastrocnemius medialis & peroneus longus, Fig.2), along with inter-muscle dependence (EMG1-EMG2 covariance) and time-dependence (TIME-EMG smoothness). Here vertical dotted lines depict the magnitude of standard deviations. Projection of EMG1 and EMG2 onto the (EMG1, EMG2) plane results in covariance ellipses, where ellipse orientation indicates the direction of maximum covariance.

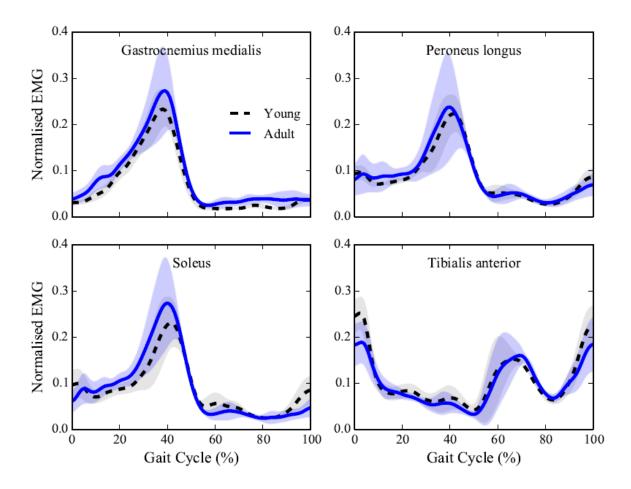


Figure 2. Mean filtered Young (black – dashed) and Adult (blue) gait EMG time-series from Bovi *et al.* (2011). The shaded standard deviation clouds, although typically assumed to be independent, actually arise from time-dependent inter-muscle covariance (Fig.1).

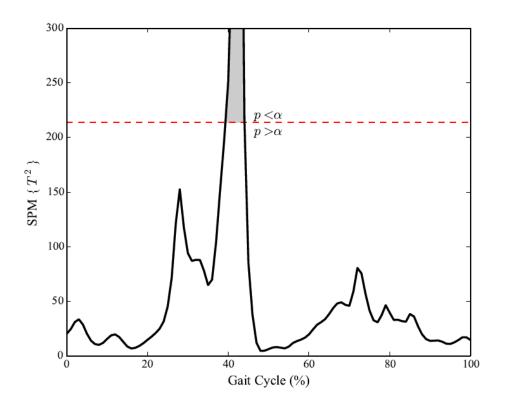


Figure 3: SPM results (Hotelling's T^2 test statistic trajectory) depicting Young–Adult differences. The critical threshold (red dashed line) was 213.7. One region of the T^2 trajectory (a supra-threshold cluster - shaded) exceeded the critical threshold. SPM therefore finds a significant group difference (p<0.05) but scalar analyses did not.

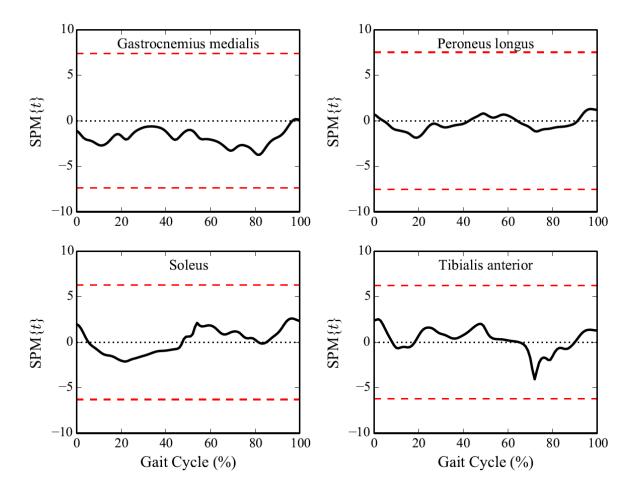


Figure 4. Post-hoc two-sample SPM t-test results comparing Young versus Adult groups for individual muscles. No SPM $\{t\}$ values reached the critical threshold (dashed line) for significance. Post-hoc tests are provided for example only as the null hypothesis tested is completely answered by the independent Hotelling's T^2 test.

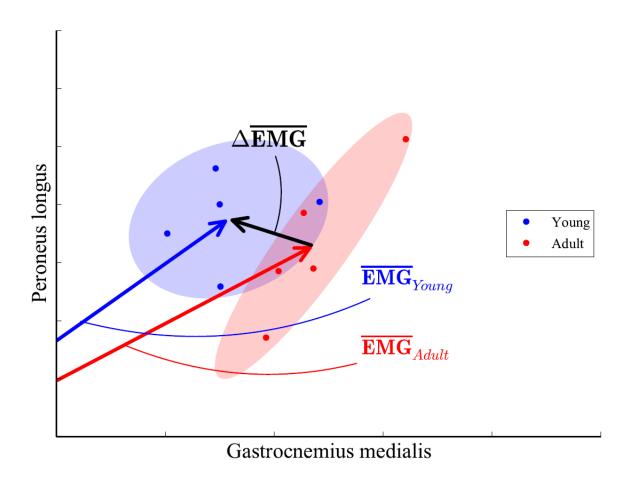


Figure 5: Example inter-muscle dependence between the gastrocnemius medialis and the peroneus longus at the time of the greatest vector difference (time=43%). Ellipses depict covariance. The small variance in the ΔEMG direction leads to null hypothesis rejection (see Fig.3), but intra-muscle analysis fails to reject the null hypothesis.