



Stride-to-stride fluctuations and temporal patterns of muscle activity exhibit similar responses during walking to variable visual cues

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ARTICLE INFO

Keywords:

Walking
EMG
Complexity
Fractals
Variability
Cueing

ABSTRACT

Incorporating variability within gait retraining approaches has been proposed and shown to lead to positive changes. Specifically, submitting the individuals to walk in synchrony to cues that are temporally organized with a fractal-like patterns, promotes changes at the stride-to-stride fluctuations closer to those typically find in young adults. However, there is still a need to understand the underlying neuromuscular mechanisms associated to such improvement. Thus, this study aimed to investigate whether changes in the temporal structure of the variability in gait patterns are accompanied by changes in muscle activity patterns. Fourteen young individuals walked synchronized to one uncued (UNC) and three cued conditions: isochronous (ISO), fractal (FRC) and random (RND). Inter-stride intervals were determined from an accelerometer placed on the lateral malleoli. Inter-muscle peak intervals were obtained from the electromyographic signal from the gastrocnemius muscle. Fractal scaling, obtained through detrended fluctuation analysis, and coefficient of variation were calculated. Repeated measures ANOVAs were used to identify differences between conditions. Significant main effect was observed for both fractal scaling and coefficient of variation. Both shown no differences between UNC and FRC conditions, while ISO and RND were significantly lower compared to UNC and FRC conditions. In addition, a Pearson's Correlation was used to test the correlation between variables. A strong correlation was found the temporal structure of gait and muscle activity patterns. These findings strengthen the current literature regarding the incorporation of variability within cued approaches. Specifically, it shows that such an approach allows the modification of the neuromuscular processes underlying the stride-to-stride fluctuations.

1. Introduction

Fluctuations in biological signals are increasingly being investigated given its clinical potential and added value to the study of disease and pathology. Therefore, a substantial amount of research has been conducted on this subject by several research teams within the last two decades. This work has well established the clinical value of these fluctuations in some biological signals such as heart rate (Costa et al., 2002; Goldberger, 2001; Goldberger et al., 2002; Ivanov et al., 1999; Peng et al., 1995). However, further research is needed to establish such relevance to the health professionals and researchers' community in other areas such as in human movement and specifically in locomotion (Almurad et al., 2018, 2017; Delignières and Torre, 2009; Ezzina et al.,

2021; Hausdorff, 2007, 2009; Hausdorff et al., 2001, 1998, 1996, 1994; Hausdorff et al., 1997a; Hausdorff et al., 1997b; Herman et al., 2005; Hunt et al., 2014; Jordão et al., 2022; Kaipust et al., 2013, 2012; Marmelat et al., 2014; Roerdink et al., 2015; Vaz et al., 2024; Vaz et al., 2020a; Vaz et al., 2020b; Vaz et al., 2019).

What we do know thus far is that stride-to-stride fluctuations present in gait patterns in healthy walkers exhibit non-random fluctuations (Hausdorff et al., 1998, 1996, 1995; Hausdorff et al., 1997a; Hausdorff et al., 1997b). Specifically, they have a fractal-like pattern. Briefly, a fractal is classically described as a geometric object with self-similarity over multiple scales. In biological signals there is a power-law relation where oscillations appear self-similar when observed over seconds, minutes, hours or days.

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<https://doi.org/10.1016/j.jbiomech.2024.111972>

Accepted 29 January 2024

Available online 3 February 2024

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More importantly, previous research also reported that such healthy, fractal-like, patterns are not present in neurological patients and are affected by ageing (Hausdorff et al., 1997b). In both situations, they typically switch towards random type of fluctuations. In the early 2000's, it has been proposed that these fractal-like patterns characterize a complex system that presents high levels of adaptability (Stergiou et al., 2006). Conversely, when these patterns alter towards randomness or become highly repeatable, the adaptability of the system decreases. Specifically, an individual that exhibits complex, fractal-like, patterns during walking contains sufficient motor solutions and plasticity to deal with sudden events that, for example, may cause a fall. A neurological patient or an older adult, that present random fluctuations (i.e., less complex structure), will be less likely to adapt to the same event leading to a fall (Hausdorff, 2007; Herman et al., 2005). In fact, it has previously been reported that fallers present more randomness in their gait patterns compared to non-fallers (Ravi et al., 2021).

Although these changes in gait patterns with pathology and ageing are well established, research is limited regarding how rehabilitation or training may harness such a fundamental feature of human movement. Nevertheless, recent research has shown the potential of sensorimotor synchronization to recover healthy gait patterns (Hunt et al., 2014; Jordão et al., 2022; Marmelat et al., 2014; Vaz et al., 2020a; Vaz et al., 2020b; Vaz et al., 2019). Particularly, these studies suggested that incorporating a fractal based cueing system to walk with, could enable the individual to exhibit fractal-like patterns. Our group has recently shown that gait patterns can be modified, restored, and even be retained after such a cueing system is turned off (Vaz et al., 2020b). Other rehabilitation approaches have also shown some promising results in the restoration of these patterns, such as using Tai-Chi (Gow et al., 2017) and interpersonal coordination during walking (Almurad et al., 2018; Ezzina et al., 2021).

Despite these recent advances, we still lack a clear comprehension regarding the underlying neurophysiological and control mechanisms of these complex, fractal-like, gait patterns. Theoretically, the presence of adaptive and flexible behaviors emerges from the dynamic interaction between the nervous system, the body, and the environment (Cavanaugh et al., 2017; Harrison and Stergiou, 2015). However, this organism-environment interaction is only the observable one. In other words, the emergence of an adaptive behavior is the result of a dynamic multilevel and multiscale interaction. It is, therefore, plausible to think that the kinematics of gait (e.g., stride time) is determined by a complex and dynamic interaction from different muscles and each muscle's activity and activation is the result of the interaction of several other microlevel processes (e.g., motor unit recruitment, stretch-reflexes, etc.). This points to an important experimental next step; whether these complex, fractal-like, gait patterns are only present at the outcome, or we can find them in parts of the process, e.g., in movement effectors such as muscles. In other words, we question whether these patterns in human gait, typically observed in stride time parameters, also emerge at the muscle activity level. If that would be the case, when we manipulate gait patterns through cueing, the same patterns as those observed in parameters such the stride time interval, would also be present at the muscle activity level.

Therefore, the present study investigated whether changes in the temporal structure of the variability (i.e., how fluctuations change over time) in gait patterns are accompanied by changes in the temporal structure of the variability in muscle activity. According to the above presented theoretical framework, we hypothesized that the changes in the temporal structure of the variability in muscle activity, would follow the direction of the gait manipulation. In other words, the muscle activity patterns would become more random while the individual synchronizes to a random-like cues. Additionally, we investigated if the temporal structure of gait and muscle activity are related. We hypothesized that a positive correlation between the temporal structure of the variabilities in gait and muscle activity, will suggest that the neuromuscular control of movement incorporates the observed changes in the

gait patterns. To accomplish the above, we experimentally manipulated the temporal structure of the variability present in gait using different cued conditions. Specifically, we manipulated the temporal structure of the cueing, as we previously performed successfully to demonstrate that such manipulations could lead to similar changes in the temporal structure of the variability present in gait patterns (Vaz et al., 2020a; Vaz et al., 2020b; Vaz et al., 2019).

2. Methods

2.1. Participants

A priori sample size calculation was determined based on the primary hypothesis. Based on previous related research (Vaz et al., 2019), the sample size calculation revealed fourteen individuals would provide an 80 % power to detect an effect size of 0.92 at a significance level of 0.05.

Participants had no medical history of cardiovascular or metabolic disease/disorders, nor history of musculoskeletal disorders in the past 6 months. Participants signed an informed consent that was previously approved by the Institutional Review Board.

2.2. Experimental procedures

Testing was completed in one session. The session began with the determination of the preferred walking speed (PWS). For PWS determination, participants were asked to start walking on the treadmill and indicate when comfortable with the treadmill's speed, while the treadmill speed was increased in increments of 0.1 km/h. The participants were allowed to remain walking at the selected speed for familiarization. Then, the first trial was an uncued condition (UNC) needed for the calculation of the preferred stride time. The mean and standard deviation of the stride time from this condition was then used to design individualized stimuli for the three randomly presented cued conditions: Isochronous (ISO), Fractal (FRC) and Random (RND). The stimuli were scaled using the mean and standard deviation of each participant's preferred stride-time. This scaling generated a set of participant-specific stimuli, but also maintained the consistency of stimulus patterns across participants. Each of the four conditions lasted 10-minutes and there was a minimum of 5-minutes resting period between trials.

The stimulus for the cued conditions was provided via a moving horizontal bar projected on a screen in front of the participant (Fig. 1). Participants were instructed to synchronize their right heel strike to the top of the moving bar's path (Jordão et al., 2022; Vaz et al., 2020b; Vaz et al., 2019). The moving indicator turned red when reaching the top of the display. The FRC stimulus was generated using an approximation of a -10 dB/decade filter with a weighted sum of first order filters. The RND stimulus was generated using a normal distribution of random numbers. These stimuli were previously validated using Detrended Fluctuation Analysis (DFA) which allows the calculation of the fractal scaling exponent alpha (α) (FRC: $\alpha = 1$; RND: $\alpha = 0.5$) (Ravi et al., 2020). The ISO stimulus was generated using each participant's mean self-paced stride-time and a standard deviation of zero. As mentioned above, these stimuli were scaled using the mean and standard deviation of each participant's preferred stride-time (UNC condition). This scaling generated a set of participant-specific stimuli, but also maintained the consistency of stimulus patterns across participants.

2.3. Data collection

A triaxial miniaturized accelerometer sampled at 1000 Hz (Plux, Lisbon, Portugal) was placed at the lateral malleoli of the right foot and was used to determine gait events. Electromyography (EMG) was collected from the Gastrocnemius Medialis (GM) through a telemetric system (Plux, Lisbon, Portugal) at 1000 Hz. First, the skin was shaved and cleaned with an alcohol solution to minimize skin impedance. Disposable pre-gelled Ag/AgCl electrodes with inter-electrode distance

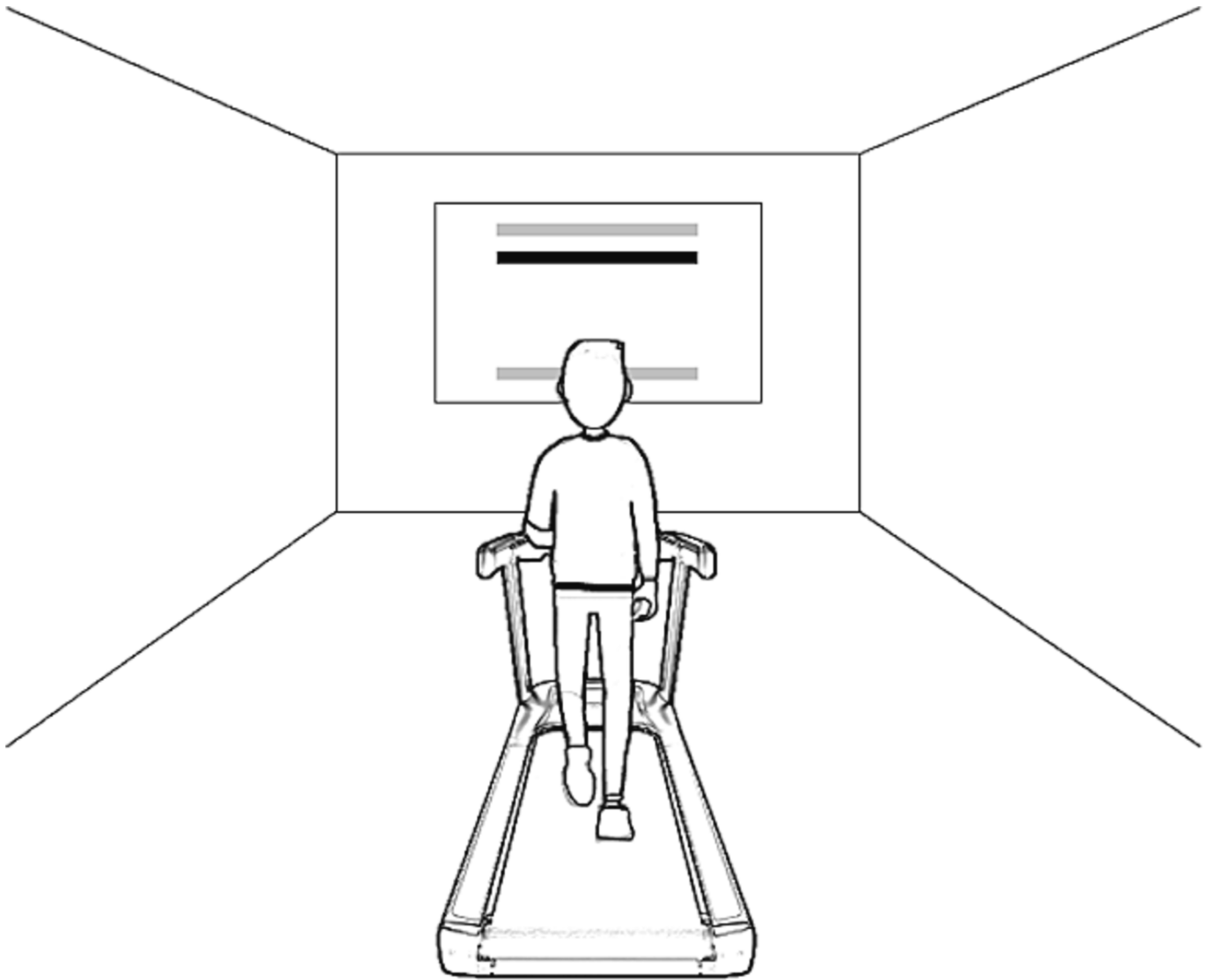


Fig. 1. Schematic representation of the experimental setup. The treadmill was placed in front of the screen. The stimulus for the cued conditions was provided via a black moving horizontal bar projected on a screen in front of the participant. Participants were instructed to synchronize their right heel strike to the top of the moving bar's path.

of 20 mm were placed according to SENIAM Project recommendations (Hermens et al., 2000). The wire was securely fixed with tape to avoid movement interference. Acceleration and EMG signals were synchronized.

2.4. Data analysis

The first 15-seconds of each trial were discarded prior to analysis to avoid transient effects of familiarization to the stimulus. A 4th order, zero lag low-pass Butterworth filter with a cutoff frequency of 20 Hz was applied to the accelerometer signal. Filtering cutoff frequency was determined according to Winter (Winter, 2009). A custom Matlab® (version 2019b) code was used to determine inter-stride intervals (ISIs), which was defined as the time difference between two consecutive heel strikes of the same foot. Heel strike events were detected using a previously reported approach (Jasiewicz et al., 2006). Raw EMG signals were band-pass filtered (20–500 Hz), full-wave rectified and smoothed with a low-pass filter (12 Hz, 4th order Butterworth), following the recommendations from the International Society of Electromyography and Kinesiology (Merletti, 1999). Then, the peak maximum from each gait cycle was identified and the time difference between two

consecutive peaks was determined: inter muscle peak intervals (IMPIs). The coefficient of variation (CV) and the fractal scaling exponent α were calculated for each ISIs and IMPIs time series. The CV was used as a measure of the magnitude of variability, while the fractal scaling exponent α was used as a measure of the temporal structure of variability. DFA was used to determine the fractal-scaling exponent α for ISIs and IMPIs time series (Hunt et al., 2014; Marmelat et al., 2014; Vaz et al., 2019). Window sizes of 16 to $N/9$ were used, where N is the length of the data (Damouras et al., 2010).

2.5. Statistical analysis

Analyses were performed using R (R Core Team; Vienna, Austria) with the level of significance set *a priori* to 0.05. Descriptive means, standard deviations, and confidence intervals were calculated for ISIs and IMPIs for each condition. To study concomitant changes between conditions, repeated measures analysis of variance was used to determine if differences existed in participants' mean values across conditions. The assumption of normality was tested through Shapiro Wilk's Test. Mauchly's Test of Sphericity was used to test the assumption of equal variance. When the assumption of sphericity was not met, the

Greenhouse-Geisser adjustment was used. For variables found to exhibit differences among conditions, Bonferroni-adjusted pairwise comparisons were used to determine which condition produced significantly different mean values from others. For main and interaction effect sizes were calculated as generalized eta squared. As for multiple comparison, Cohen's d was calculated as a measure of effect size. When nonparametric tests were used, Cliff's delta (δ) was used. Pearson's Correlation Coefficient was used to test the association between ISIs and IMPIs.

3. Results

3.1. Participants

Fourteen male young adults (age: 22.08 ± 3.80 yrs, height: 1.75 ± 0.07 m, body mass: 69.75 ± 8.49 kg) participated in this study.

3.2. Inter stride intervals (ISIs)

For α -ISIs, a significant main effect was observed for condition ($F_{27.501,2.115} = 29.288$, $p < 0.001$, $\eta_p^2 = 0.693$). Pairwise comparisons revealed that α -ISIs were significantly higher in UNC (0.91 ± 0.16) as compared to ISO (0.52 ± 0.14 , $p < 0.001$, $d = 2.59$) and RND (0.63 ± 0.12 , $p = 0.002$, $d = 1.98$). The α -ISIs were also significantly higher in FRC (0.83 ± 0.11) as compared to ISO ($p < 0.001$, $d = 2.46$) and RND ($p < 0.001$, $d = 2.61$; Fig. 2 upper panel).

For CV-ISIs, a significant main effect was observed for condition ($\chi^2(3) = 19.791$, $p < 0.001$). Pairwise comparisons showed higher CV-ISIs in the FRC as compared to the UNC ($p = 0.041$, $\delta = 0.857$) and ISO ($p < 0.001$, $\delta = 0.714$) conditions (Table 1). No other significant differences were found between conditions.

3.3. Inter muscle peak intervals (IMPIs)

For α -IMPIs, a significant main effect was observed for condition ($F_{27.888,2.145} = 29.280$, $p < 0.001$, $\eta_p^2 = 0.693$). Pairwise comparisons revealed that α -IMPIs were significantly higher in UNC (0.80 ± 0.19) than ISO (0.41 ± 0.14 , $p < 0.001$, $d = 2.34$) and RND (0.55 ± 0.12 , $p = 0.005$, $d = 1.57$). The α -ISIs was also significantly higher in FRC (0.76 ± 0.13) as compared to ISO ($p < 0.001$, $d = 2.59$) and RND ($p < 0.001$, $d = 1.68$; Fig. 2 lower panel).

For CV-IMPIs, a significant main effect was observed for condition ($\chi^2(3) = 16.727$, $p = 0.001$). Pairwise comparisons showed higher CV-IMPIs in the FRC than during the UNC ($p = 0.016$, $\delta = 0.857$) and ISO ($p = 0.001$, $\delta = 0.714$) conditions (Table 1). No other significant differences were found between conditions.

3.4. Correlation Analysis

Pearson's Correlation Analysis showed that α -ISIs and α -IMPIs were strongly and positively correlated ($r_{(56)} = 0.911$ [CI_{95%}: 0.852 to 0.947], $r^2 = 0.830$, $p < 0.001$). Fig. 3 presents a scatter plot illustrating this correlation.

4. Discussion

This study aimed to investigate whether changes in the temporal structure of the variability present in gait patterns are accompanied by changes in the temporal structure of the variability of muscle activity patterns, while young healthy adults walked in synchronization to different temporally structured cues. Additionally, it aimed to investigate if the temporal structure of variability of gait and muscle activity patterns are positively correlated. The present study findings supported our hypotheses. We have observed that the temporal structure of gait and muscle activity patterns exhibited the same trend. They exhibited fractal-like patterns during the uncued and the fractal cueing conditions.

In addition, we found a change in these patterns towards random fluctuations while walking to an isochronous or a random cueing condition.

It is well established in the literature that changes in the temporal structure of variability in gait patterns could occur as a function of the temporal structure of the cues the individuals walk to. Our fractal scaling exponent findings agree with other studies which have shown that young adults present fractal-like patterns in their stride-to-stride fluctuations during uncued gait (Hunt et al., 2014; Marmelat et al., 2014; Rhea et al., 2014a, 2014b; Roerdink et al., 2015; Vaz et al., 2020a; Vaz et al., 2019). This is an expected result as young adults should have fractal-like patterns due to their optimal healthy status. Furthermore, our results demonstrate that these patterns are also present when young adults walk in sync to fractal-like cues as it was also found by others (Hunt et al., 2014; Marmelat et al., 2014; Rhea et al., 2014a, 2014b; Roerdink et al., 2015; Vaz et al., 2020a; Vaz et al., 2019). Conversely, when walking to an isochronous or random-like cues, these patterns change towards random white-noise like (Hunt et al., 2014; Marmelat et al., 2014; Vaz et al., 2020a; Vaz et al., 2019). Again, this result confirms similar findings from other studies (Hunt et al., 2014; Marmelat et al., 2014; Rhea et al., 2014a, 2014b; Roerdink et al., 2015; Vaz et al., 2020a).

Where the novelty of our work lies, is in the fact that our experimental paradigm was able to produce similar results in terms of the fractal scaling exponent of the muscle activity. Recent research has also examined the fractal properties of EMG signals during locomotion, however, the authors applied these methods to the entire continuous EMG signal (Mileti et al., 2020; Santuz et al., 2020; Santuz and Akay, 2020). Our approach extracted the fractal properties from a discrete time series. As described above, we identified the activation per stride and determined the inter muscle peak intervals, similarly to what is typically conducted for gait kinematic parameters such as stride time. The reason for such an approach was twofold: i) to study if the duration of muscle activity events would follow the same dynamics as previously described for gait kinematic parameters, and ii) to investigate the interdependency of gait and muscle activity patterns. Importantly, this collects the global motor control strategy rather than the potential individual processes that occur at the millisecond level. We have recently reported the first ever evidence of the presence of fractal patterns at the muscle activity level in human locomotion (Jordão et al., 2023). Specifically, we found a strong correlation between the fractal patterns of gait and muscle activity in older adults. The present study adds an important next step where we show that by manipulating gait patterns, alterations in muscle activity follow the temporal structure of the manipulation applied.

Our findings also support the theoretical framework that proposed the existence of multiscale and multilevel interactions at the temporal domains of the physiological systems. Furthermore, the fact that our results demonstrated that muscle activity patterns follow the same direction of gait patterns when individuals sync their strides to different temporally structured cues, also support this multi-level interaction in the emergence of adaptable flexible behaviors. Similarly, as far as we know, this is the first study that brought evidence that the temporal structure of the variability of muscle activity could be altered as a function of cues' temporal structure.

Our findings that muscle activity patterns exhibit fractal properties during human gait and are modifiable through cueing together with our recent findings (Jordão et al., 2023) indicate that gait training methods could lead to changes that are intrinsic to muscular control. One could speculate, for example, that gait training using our experimental paradigm could also lead to positive adaptations in the presence of muscle related pathology. That may explain the findings of carryover effects we have previously observed using our experimental paradigm (Vaz et al., 2020b).

Our findings also fill an important knowledge gap by connecting neurophysiological processes with stride-to-stride fluctuations of gait patterns, bringing new evidence regarding the dynamic interaction

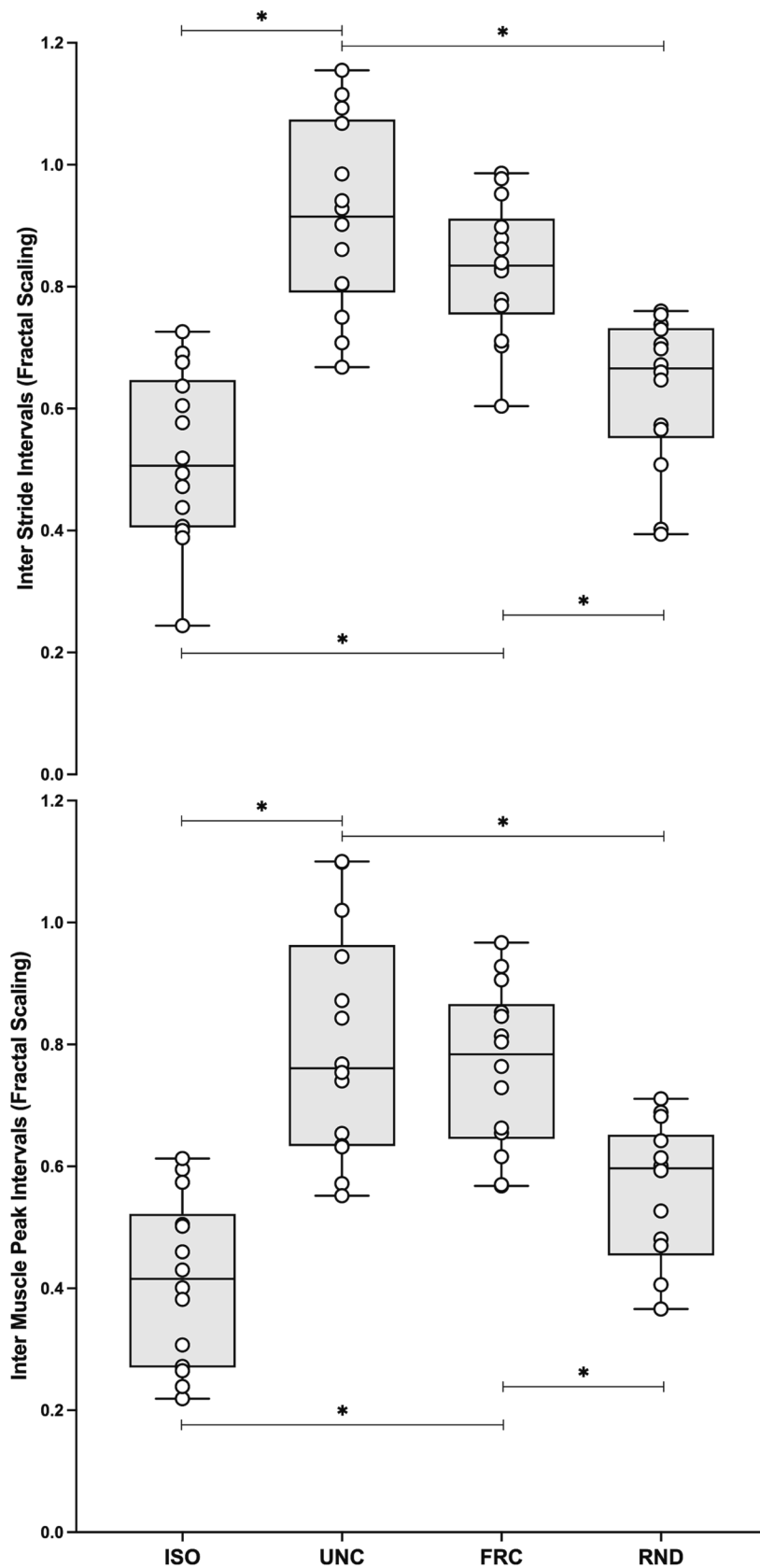


Fig. 2. Boxplots of Inter Stride Intervals (upper panel) and Inter Muscle Peak Intervals (lower panel) fractal scaling (α) values across all walking conditions. Data are presented as $M \pm 95\%$ CI. Bullets represent individual data points. UNC – Uncued; ISO – Isochronous; RND – Random; FRC – Fractal; * $p < 0.05$.

Table 1
Descriptive statistics for all dependent variables. Data are presented as Mean ± SD.

	Isochronous	Uncued	Fractal	Random
Inter Stride Intervals (ISIs)				
Fractal scaling (∞)	0.52 ± 0.14	0.91 ± 0.16	0.83 ± 0.11	0.63 ± 0.12
Coefficient of Variation (%)	1.3 ± 0.3	1.7 ± 0.6	2.4 ± 0.4	1.8 ± 1.1
Inter Muscle Peak Intervals (IMPIs)				
Fractal scaling (∞)	0.41 ± 0.14	0.80 ± 0.19	0.76 ± 0.13	0.55 ± 0.12
Coefficient of Variation (%)	1.6 ± 0.4	1.7 ± 0.4	2.5 ± 1.4	2.3 ± 1.4

between different levels of the physiological system. In fact, previous research suggested such type of an interaction is possible. For example, previous research has shown that the fractal properties of gait are stronger during overground walking than walking on a treadmill where additional constraints are in place (Terrier and Dériaz, 2011). On a recent study, Meliti et al. (2020) showed that the motor modules (i.e., synergistic organization of several muscles) become more regular during treadmill walking. This suggests that the level of flexibility of the motor modules likely affects the flexibility of the locomotor system, which is at a macrolevel of analysis. Furthermore, some preliminary findings showed that the complexity of heart rate and brain activity signals followed the complexity of walking paths that individuals walked to (Kamal et al., 2020a, 2020b), suggesting these systems were coupled at the temporal structure domain.

Important next steps are certainly necessary to either support or

refute the present findings. On one hand, incorporating the measurement of other muscles can provide more robust information regarding potential changes at the motor control level. Our study is limited to the activity of the gastrocnemius. While the gastrocnemius muscle is a strong contributor during the gait cycle, particularly during the push-off phase, it is certainly not the only muscle involved during gait. For example, looking at gastrocnemius-tibialis anterior co-activation may provide additional relevant information. We anticipate that greater co-activation, typically present in older adults' gait (DaSilva et al., 2021), lead to more rigid and constrained motor solutions. In other words, it turns the physiological system as less adaptable. However, this needs to be experimentally tested. Similarly, investigating the muscle activity patterns in older adults' walking, that typically present more random type of gait patterns, is also needed to validate, and expand the present findings to clinical populations.

This fractal-like training approach can be seen as a mechanistic method that aims to change an outcome measure – stride time fluctuations – rather than modifying motor control strategies. However, we believe that our approach, a visual continuous moving bar, provides both feedback and feedforward information and the users can adapt their gait strategies to follow it. Importantly, the users are not instructed to match on a fractal or an isochronous way. They hardly perceive those as different conditions since the difference is very subtle. This means that the present approach is a goal-oriented type of training. In other words, the users are focused on external information to perform the task: heel strike matching the moving bar reaching the top of its path.

5. Conclusion

The present study showed that the temporal structure of variability

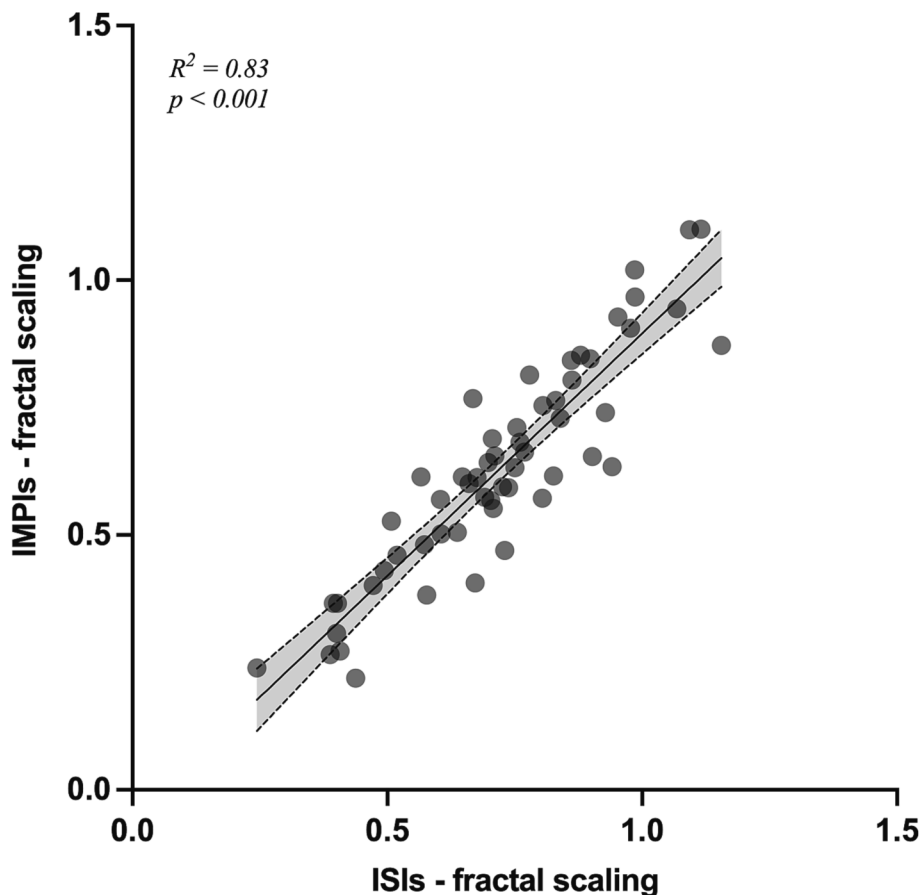


Fig. 3. The correlation between Inter Muscle Peak Intervals (IMPIs) and Inter Stride Intervals (ISIs) fractal scaling (∞) values. The grey area represents 95 % confidence intervals. The individual data points represent each participant value per condition.

present in the gait patterns is strongly correlated with the temporal structure of variability of the gastrocnemius muscle activity. We have experimentally manipulated gait patterns through a cued methodology and showed that regardless the direction of the manipulation, muscle activity patterns followed the changes also observed in the gait patterns. These results provide important neurophysiological insights to this field of research, suggesting that the proposed approach likely modifies the individual's motor control.

6. Declarations

6.1. Ethics approval and consent to participate

The studies involving human participants were reviewed and approved by the Ethics Committee from the Faculty of Human Kinetics, University of Lisbon. The participants provided their written informed consent to participate in this study.

7. Consent for publication

Not applicable.

8. Availability of data and material

The datasets generated for this study are available on request to the corresponding author.

JRV was partly supported by the Fundação para a Ciência e Tecnologia under grant numbers UIDB/00447/2020 and UIDB/04585/2020. NS was supported by the Center for Research in Human Movement Variability and the National Institutes of Health (P20GM109090, R15AG063106, and R01NS114282).

9. Authors' contributions

The conception and design of the experiments were undertaken by JRV, JSG and NS. Data collection was undertaken by JRV, JG and SJ, while assembly, analysis, and interpretation of data were undertaken by JRV, NC, JS and NS. Drafting the article or revising it critically for important intellectual content was undertaken by JRV, NC and NS. All authors have contributed to the review and improved the final version of this manuscript. All authors read and approved the final manuscript.

CRedit authorship contribution statement

João R Vaz: Conceptualization, Data curation, Writing – original draft, Visualization, Investigation, Formal analysis, Methodology. **Nelson Cortes:** Writing – review & editing, Visualization, Formal analysis. **João Sá Gomes:** Conceptualization, Data curation, Writing – review & editing, Investigation. **Sofia Jordão:** Writing – review & editing, Visualization, Investigation. **Nick Stergiou:** Conceptualization, Writing – review & editing, Formal analysis.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

JV was supported by Fundação para a Ciência e Tecnologia (UIDB/04585/2020, UID/DTP/UI447/2019); NS was supported by National Institutes of Health (P20GM109090, R15AG063106, R01NS114282).

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