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Minimal effects of age and prolonged physical and mental exercise on healthy adults' gait

Paulo Cezar Rocha dos Santos[a,b,*], Tibor Hortobágyi[a], Inge Zijdewind[c], Lilian Teresa Bucken Gobbi[b], Fabio Augusto Barbieri[d], Claudine Lamoth[a]

[a] University of Groningen, University Medical Center Groningen, Center for Human Movement Sciences, Groningen, the Netherlands
[b] São Paulo State University (UNESP), Post-graduation Program in Movement Sciences, Institute of Biosciences, Posture and Gait Studies Laboratory (LEPLO), Rio Claro, Brazil
[c] Department of Biomedical Sciences of Cells and Systems, University of Groningen, University Medical Center Groningen, Groningen, the Netherlands
[d] São Paulo State University (UNESP), Post-graduation Program in Movement Sciences, Human Movement Research Laboratory (MOVI-LAB), Department of Physical Education, Bauru, Brazil

A R T I C L E I N F O

Keywords:
Performance fatigability
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A B S T R A C T

Background: Gait adaptability in old age can be examined by responses to various perturbations. Fatigability due to mental or muscle exercises can perturb internal cognitive and muscle resources, necessitating adaptations in gait.
Research question: What are the effects of age and mental and muscle fatigability on stride outcomes and gait variability?
Methods: Twelve older (66–75yrs) and twelve young (20–25 yrs) adults walked at 1.2 m/s before and after two fatigue conditions in two separate sessions. Fatigue conditions were induced by repetitive sit-to-stand task (RSTS) and by 30-min of mental tasks and randomized between days (about a week apart). We calculated the average and coefficient of variation of stride length, width, single support, swing time and cadence, and the detrended fluctuations analysis (DFA) based on 120 strides time intervals. We also calculated multi-scale sample entropy (MSE) and the maximal Lyapunov exponent (λmax) of mediolateral (ML) and anteroposterior (AP) of the Center of Pressure (CoP) trajectories.
Results: In both age groups, RSTS modestly affected stride length, single support time, cadence, and CV of stride length ($p \leq 0.05$), while the mental task did not affect gait. After fatigability, λmax - ML increased ($p \leq 0.05$), independent of fatigue condition. All observed effects were small (η^2: 0.001 to 0.02).
Significance: Muscle and mental fatigability had minimal effects on gait in young and healthy older adults possibly because treadmill walking makes gait uniform. It is still possible that age-dependent muscle activation underlies the uniform gait on the treadmill. Age- and fatigability effects might be more overt during real life compared with treadmill walking, creating a more effective model for examining gait and age adaptability to fatigability perturbations.

1. Introduction

Natural aging modifies gait [1]. Older compared to young adults walk slower, with shorter strides, longer single-support and swing time, and higher gait variability [1]. Age-typical changes on gait also involve gait dynamics outcomes in which reflect complementary characteristics of gait performance. Whereas Detrended Fluctuations Analysis (DFA) indicates absence/presence of correlation between strides [2], Multi-scale Sample Entropy (MSE) and maximal Lyapunov exponent (λmax) measure the complexity/regularity of signal and the capacity to resist to

Abbreviations: *AP*, anteroposterior; *CoP*, Center of Pressure Position; *CPT*, continuous performance test; *CV*, coefficient of variation; *DFA*, detrended fluctuation analysis; *MFI*, multidimensional fatigue inventory; *ML*, mediolateral; MMSE, mini-mental state examination; *MSE*, multi-scale sample entropy; *MVF*, maximum voluntary force; *PVT*, psychomotor vigilant test; *RT*, reaction time; *SPPB*, short physical performance battery; *SL*, stride length; *SST*, single support time; *RSTS*, repetitive sit-to-stand task; *SW*, step width; *SwT*, swing time; *V*, vertical; *VAS*, visual analogue scale; η^2, eta Squared; λmax, maximum Lyapunov exponent

* Corresponding author. Present Adress: University of Groningen, University Medical Center Groningen, Center for Human Movement Sciences, A. Deusinglaan 1, 9713 AV Groningen, the Netherlands.
E-mail address: p.c.rocha.dos.santos@umcg.nl (P.C.R.d. Santos).

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small perturbation, respectively [3,4]. These outcomes are associated with a decline in cognitive function, mobility disability, and higher fall risk [3–5]. An important question is how older adults adapt their gait to external perturbations. Mechanical/external perturbations, as models [6], evoke age-specific adaptations in gait. Another model of gait perturbation is fatigue that interferes with internal resources [7]. There is still an inadequate understanding of how behavioral interventions, such as fatigue by enrollment in physical and cognitive exercise, may affect gait outcomes in older adults. Fatigue, as a state, emerges from a decline in an objective measure of performance over time (performance fatigability), and from a subjective increase in difficulty to continue a task (perceived fatigability) [7]. Protocols to induce fatigability involve sustained muscle/physical and/or demanding mental tasks. The responses to fatigability due to cognitive and/or physical exercises could be of interest in the assessment and development of interventions aiming to improve mobility in older adults.

Repetitive sit-to-stand (RSTS) is a model to induce fatigability and to examine age-adaptations in gait [8–10]. After RSTS, there is a decrease in maximal voluntary isometric force (MVF) of the quadriceps muscle by 9% in older adults [8,9] and an increase in the level of activation of the fatigued quadriceps coupled with compensatory increases in plantarflexor activation [11]. After RSTS, older adults walk overground with 4% shorter stride duration, 2 cm wider steps [9,10], and increase step length and trunk variability [10]. Endurance activity [12,13] and leg press and calf raise exercise [14] produced inconsistent changes in local dynamic stability during treadmill walking.

Gait is not a fully automated motor act involving cortical and cognitive control [5]. Interference with executive function and attention, as in dual-tasking, slows gait and increases stride duration variability [5]. Specifically, performing a demanding mental task for a prolonged period can reduce intrinsic motivation, executive function, and attention and increase in up 2 times the CV of stride outcomes in older adults [15]. Because older adults may have impaired cognition, it is conceivable that fatigability-induced by demanding mental tasks would amplify age-differences in gait.

Therefore, we determined the effects of age, RSTS, and a prolonged mental effort on the spatial and temporal stride outcomes and gait dynamics during treadmill walking. For gait dynamics, we quantified the center of pressure (CoP) variability and stability. We also examined the effects of age and sustained periods of mental task on stride outcomes and gait dynamics. We hypothesized that both forms of fatigability would decrease stride length and stability, increase cadence, and variability of step times and CoP trajectories. However, we expected larger effects in older compared with young adults and greater effects on gait as a result of the RSTS compared with a sustained period of demanding mental task.

2. Method

2.1. Participants

Twelve older and twelve young adults were recruited by word of mouth from the community to participate in the study (Table 1). Exclusion criteria were: neurological and cardiovascular disease; self-reported pain, musculoskeletal injury or surgery in the lower extremities that could affect the protocol; inability to walk without an assistive device; high-level of self-reported fatigue. All participants signed an informed consent form approved by the Ethical Committee of the Center for Human Movement Sciences at the UMCG.

2.2. Procedure

Participants visited the laboratory on two days about 6–8 days apart at the same time of the day for ~2 h each. They were instructed to avoid exhaustive exercises the day before testing. During the first visit, participants completed Mini-Mental State Examination (MMSE) [16], Multidimensional Fatigue Inventory (MFI) [17], the Short Physical Performance Battery (SPPB) [18], and had the demographic characteristics recorded. The experimental conditions were randomized between sessions: Session A: RSTS; Session B: Mental Task. In both Sessions, participants walked on the treadmill and performed the maximum voluntary quadriceps force (MVF) before and after experimental conditions.

Table 1
Participants Characteristics and score on questionnaires.

	Older		Young		p-value
Groups' characteristics					
N (male and female)	12 (7 and 5)		12 (7 and 5)		–
Age (yrs)	71	± 3.76	22.45	± 1.69	< 0.01
Height (cm)	173.13	± 7.70	177.45	± 9.17	0.17
Body mass (kg)	73.92	± 10.15	69.81	± 11.38	0.71
SPPB (scores)	12.00	± 0.00	12.00	± 0.00	1.00
MFI (scores)	34.58	± 9.81	38.18	± 9.36	0.52
STS					
Repetition (rep)	134.12	± 114.71	583.3	± 173.62	< 0.01
Duration (min)	4.47	± 3.99	19.48	± 5.92	

Values are means and SDs. SPPB: Short Physical Performance Battery; MFI: Multidimensional fatigue inventory; STS: Sit-to-Stand.

2.3. Walking condition

Without holding the handrail but wearing a harness, participants walked on a treadmill with two embedded force plates (M-gait, Motekforce, Amsterdam, NL) that measured 3-D ground reaction forces (N) and moments of force (Nm) under each leg at 1 kHz. Participants walked for 3 min at a fixed speed of 1.2 m/s. This speed was chosen to be similar to the older adults' comfortable speed [19,20] and to test both age groups in the same condition, eliminating a speed-effect on gait [19,20].

2.4. MVF and electrical stimulation

MVF and twitch interpolation were done on a custom-built dynamometer [21]. Participant's non-dominant leg was strapped to the lever arm with the knee 90° flexed. Placement of electrodes, the procedure to determine the stimulation intensity, and the stimulator were described previously [21]. Participants then performed maximum voluntary contractions before and after each experimental condition. Participants were instructed to contract the quadriceps as rapidly and forcefully as possible and maintain it for 5 s. Double electrical pulses were discharged on the plateau of MVF (superimposed twitch), followed by two twitches at rest. We calculated the MVF before twitch and the voluntary activation (1-(superimposed twitch /potentiated twitch))*100 [21]

2.5. Experimental conditions

Participants performed RSTS, with the arms crossed at the chest, at 30 beats/min (chair: 0.43 × 0.41 × 0.42 m). The protocol was stopped either when participants indicated an inability to continue or after 30 min. Duration and number of repetitions were recorded.

In session B, participants performed three mentally demanding tasks on a computer for 10 min each [22]: the Psychomotor Vigilance Task (PVT) [23], the Continuous Performance Test (CPT) and the Stroop test [22]. RT was assessed for all tasks and the accuracy (% of the correct answer) for Stroop (congruent and incongruent responses) test and CPT. Outcomes of the mental tasks were averaged over windows of 1 min, then an overall mean was calculated considering Time 1 (2–5 min) and Time 2 (6–10 min). Participants, after familiarized with the scales, reported perceived fatigue (Visual Analogue Scale - VAS, 0mm = no perceived fatigue 100mm = completely fatigued) for session B [22], and rate of perceived exertion (6–20 Borg scale) [24], for both sessions, before and immediately after experimental conditions.

2.6. Gait analysis and outcomes

We combined the data from the two force plates to identify heel contact and toe-off from ground forces and, combined with the moments data, we computed the Center of Pressure Position (CoP) in AP and ML directions [6].

Taking the middle 120 strides, mean and CV was calculated for Stride length (SL), Step width (SW), Single support time (SST), Swing time (SwT), Cadence and, DFA. DFA quantifies absence/presence of long-range correlations between stride time intervals using a window of length n (n = N/4), being N = 120 strides that are considered as acceptable power in between- and within-subjects designs involving older adults [2]. Values of $\alpha > 0.5$ indicate persistence in the stride time intervals and < 0.5 indicate alternation of larger- and smaller-than the average values.

Based on CoP trajectories in ML and AP direction, we calculated: 1) MSE, as an indication of gait complexity. It implies predictability of fluctuation patterns over time by increasing the length τ ($\tau = 7$) in the average of data point non-overlapping window ($r = 0.02$).MSE = Zero represents data predictability [25]; 2) The λmax was quantified by the log of the expansion between the CoP trajectories by using Wolf algorithm (embedding of $n = 7$ dimensions, delay τ of 10 samples), which is the most appropriated to evaluate λmax from relatively small data sets [26]. Large λmax indicate lower local dynamic stability (smaller ability to resist perturbations) [26]. Both methods, MSE and λmax, were previously employed in relatively small lengths, i.e., 3 min walking [4,26].

2.7. Statistical analysis

Power calculation (G*Power software) required a minimum of 24 participants (probability of 82% to detect a difference in SL at 5% of significance). Using, SPSS (SPSS Inc., USA), when the Shapiro-Wilk test revealed non-normal distribution, data were log-transformed. A Student's *t*-test between groups was applied to compare the characteristics, questionnaires and SPPB, and RSTS outcomes. To compare mental task performance, an ANOVA was applied with between-factors Group (young vs. older) and within factor Time (time 1 vs. time 2). ANOVAs with between-factor Group and within-factors Experimental conditions (RSTS vs. mental), and Time (pre- vs. post-experimental conditions) were applied for the Borg scale, MVF and voluntary activation, and the gait outcomes. Tukey's post hoc contrast was used to identify significant differences. The level of significance adopted was $p \leq 0.05$. Effect sizes were estimated concerning eta square (η^2). $\eta^2 \geq 0.02$ indicate small, ≥ 0.13 intermediate, and ≥ 0.26 large effects [27].

3. Results

3.1. Participants

Table 1 shows that the two age groups had similar characteristics (all $p > 0.05$).

3.2. Experimental conditions

Young performed up 4x longer RSTS than older participants (Table 1, $T_{22} = 7.40$, $p < 0.01$, $T_{22} = 7.39$, $p < 0.01$ respectively). Fig. 1a and Table 2 show the Group by Experimental condition by Time interaction for the Borg scale ($p < 0.01$), indicating an increase after RSTS ($p < 0.01$) but not after the mental tasks ($p > 0.05$). Older vs. young adults reported higher exertion levels pre-fatigue. Fig. 1b and Table 2 show Time by Experimental conditions interaction for MVF indicated a decrease by 17% in both groups after RSTS ($p < 0.05$) but only ˜2% after the mental tasks ($p > 0.05$). Group main effect revealed that older adults performed ˜40% less MVF than young ($p < 0.01$).

Fig. 1c shows Time main effect ($p < 0.05$) with a longer RT for PVT (˜7%) and Stroop test (˜15% for congruent and incongruent), and Accuracy for incongruent responses in Stroop decreased by 5% ($p < 0.05$), and CPT RT was ˜9% shorter at time 2 compared with time 1. ($p < 0.05$). The VAS was significantly higher after (˜30 mm) than before the mental task. Group main effects ($p < 0.05$) indicated that the older had longer RTs for the PVT (12%), and the Stroop test (congruent responses by 56%; incongruent responses by 65%) than young adults (Table 3). There was no Time by Group interaction ($p > 0.05$).

3.3. Effects of the experimental condition and age on gait

As a secondary analysis, we separately compared the effects of experimental conditions on gait outcomes in a speed condition from 20 to 30% faster than the comfortable. For both gait speed, detailed ANOVA and outcomes are presented in Supplementary Material T1, T2, and T3.

Fig. 2a shows the Time by Experimental condition interaction in stride outcomes. Post hoc revealed significant effects of RSTS on SL, CV of SL, SST, and cadence ($p \leq 0.05$). No significant changes were observed after the mental task ($p > 0.05$) (η^2: 0.001 to 0.007).

Group main effects revealed that older adults walked with a higher CV of SL, CV of SST, CV of SwT, and lower SwT ($p < 0.05$ for all) than young adults (η^2: 0.10 to 0.155).

3.4. Gait dynamics

Time main effect ($p < 0.05$) indicated an increase by 11% in ML λmax after the experimental condition ($p < 0.05$, η^2: 0.020). Time by Experimental conditions interaction for MSE of ML-CoP ($p < 0.05$, Fig. 2b) showed a 5% decrease after the mental tasks (η^2: 0.015).

Group main effects revealed up to 50% higher in AP and ML λmax in older than young adults ($p < 0.05$ for all, η^2: 0.20 and 0.215).

4. Discussion

We determined the effects of age, RSTS, and prolonged mental effort on gait. RSTS reduced MVF and increased perceived exertion in both age groups. After the mental task, perceived fatigue was higher, and RT of PVT and Stroop tests lower. While both manipulations effectively induced fatigability, these interventions produced minimal effects on healthy adults' treadmill walking.

While the mental tasks did not affect stride outcomes, after RSTS SL and single ST decreased by ˜2 cm and ˜5 ms, respectively and cadence increased by ˜1.5 steps/min. Corresponding effect sizes suggested minimal functional effects (Fig. 2). These results agree with data collected after sustained endurance exercise, showing small (Cohen d: 0.1–0.4) or no effects on stride outcomes, gait variability [28], or on local dynamic stability of foot contact velocity and trunk accelerations [12–14] during treadmill walking. In contrast, when older adults walked overground, RSTS did affect stride outcomes and gait variability [9,10]. Repetitive leg-press plus calf and toe rises also increased the average and variability of the margin of stability during treadmill walking at a comfortable speed [14]. The inconsistent results between studies may be due to differences in the assessment of walking (treadmill vs. overground) and models to induce fatigability (muscles, tasks).

While treadmill vs. overground walking has the advantage of examining gait under a standardized condition, there are biomechanical differences in gait under in the two conditions [29]. Overground walking requires the participants to actively adjust features of gait, whereas the belt movement presets steps during treadmill walking. Treadmill walking is more reactive, which, compared with overground walking, necessitates lower muscle activation to generate force to

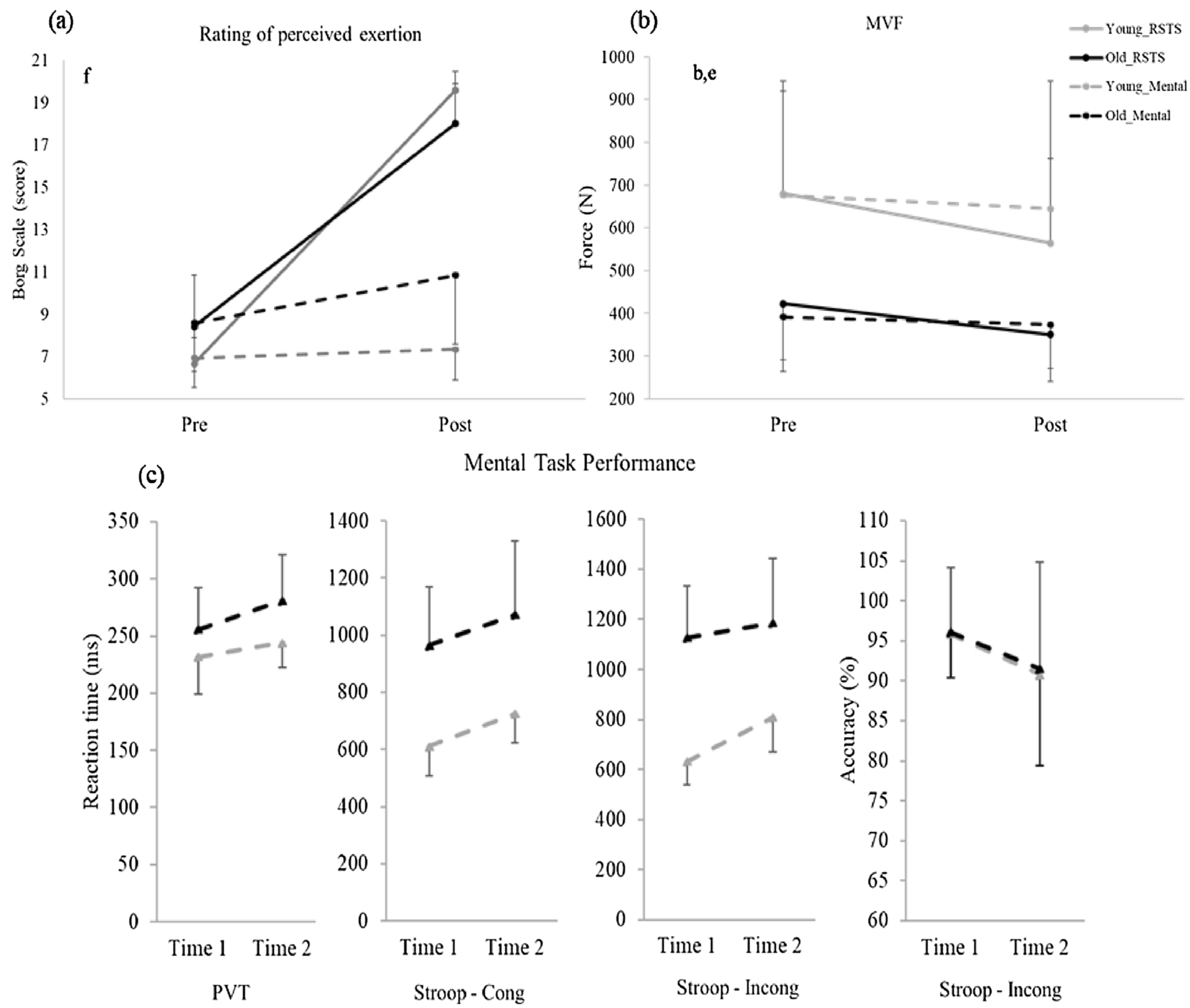

Fig. 1. Average and standard deviation of young (grey line) and older (black line) adults: (a) rating of perceived exertion and (b) MVF pre- and post-fatigue, and (c) mental task performance for the RT of PVT and Stroop test (congruent and incongruent answers) and for accuracy on Stroop test, considering time 1 (first minutes) and time 2 (last minutes). Continuous and dashed lines represent RSTS and mental task, respectively. [a]Time main effect, [b]Group main effect, [e]Time by Experimental conditions interaction, [f]Group by Experimental conditions by Time interaction.

advance the legs [29], making the cyclical movements of the legs uniform [28] and, compressing stride variability by 15% [30]. Thus, treadmill walking could minimize fatigue-effects on gait [28].

Muscle contributions to gait mechanics can also account for the small perturbation effects on gait. RSTS tends to reduce MVF and median frequency of the EMG of the knee extensors but not of the ankle muscles [9]. During walking, the knee extensors decelerate the center of mass and stabilize the leg at heel strike [31]. However, the joint power generated by the knee extensors compared with the plantarflexors is ˜4 times lower and plantarflexor power correlates ($r^2 > 0.5$) with the age-related decrease in SL and gait speed [32]. These factors support prior data [14], showing the effects of combined knee and ankle fatiguing

Table 2
Mean and standard deviation of experimental conditions outcomes in Older and Young groups.

Experimental conditions outcomes		RSTS		Mental tasks	
		Pre	Post	Pre	Post
Borg Scale [f]	Older	8.42 ± 2.42	19.58 ± 9.0	8.58 ± 2.39	10.83 ± 3.21
	Young	6.67 ± 1.23	18.00 ± 1.91	6.92 ± 1.38	7.33 ± 1.43
MVF (N) [b, e, f]	Older	422.08 ± 133.17	354.83 ± 116.33	391.25 ± 188.57	373.92 ± 102.41
	Young	676.25 ± 244.71	563.27 ± 208.72	676.17 ± 270.81	657.83 ± 284.57
VC (%)	Older	86.50 ± 11.22	87.56 ± 13.21	85.50 ± 14.78	89.33 ± 13.47
	Young	88.65 ± 17.65	87.93 ± 14.35	90.67 ± 11.22	90.58 ± 8.64

MVF, Maximum Voluntary Force; VC, Voluntary Contraction
[a]Time main effect, [b]Group main effect, [c]Experimental condition main effect, [d]Time by Group interaction, [e]Time by Experimental conditions interaction, [f] Group by Experimental conditions by Time interaction.

Table 3
Mean and standard deviation of mental tasks outcomes in Older and Young groups, considering time 1 (minutes 2–5) and time 2 (minutes 6–10).

Mental Tasks Outcomes		Time 1		Time 2	
RT (ms)					
PVT [a, b, d]	Older	256.02	± 37.78	281.05	± 41.85
	Young	231.68	± 34.44	244.15	± 23.11
Stroop – Cong [a, b, d]	Older	965.01	± 214.13	1130.87	± 420.33
	Young	611.43	± 91.45	725.71	± 108.09
Stroop – Incong [a, b, d]	Older	1127.92	± 286.75	1261.80	± 514.39
	Young	632.35	± 97.79	807.43	± 145.35
CPT [a, b]	Older	395.02	± 125.22	370.35	± 93.93
	Young	306.16	± 71.35	302.22	± 66.89
Accuracy (%)					
Stroop – Cong [a, b]	Older	99.08	± 1.49	96.06	± 4.81
	Young	94.93	± 4.31	95.66	± 4.98
Stroop – Incong [a, b, d]	Older	95.83	± 8.40	91.47	± 14.01
	Young	96.11	± 5.74	90.77	± 11.84
CPT [a, b, d]	Older	80.00	± 19.05	80.47	± 17.58
	Young	89.59	± 9.35	92.39	± 5.06
Visual Analogue Scale		Pre - mental tasks		Post - mental tasks	
(mm) [a]	Older	9.75	± 8.35	47.83	± 26.18
	Young	10.33	± 8.66	49.25	± 15.25

RT, Reaction Time; PVT, Psychomotor Vigilance Test; Cong, Congruent answer; Incong, Incongruent Answer; CPT, Continuous Performance Test; RT.
[a]Time main effect, [b]Group main effect, [c]Experimental condition main effect, [d]Time by Group interaction, [e]Time by Experimental conditions interaction, [f] Group by Experimental conditions by Time interaction.

exercises on gait variability (˜10% increase in variation of the AP and ML margin of stability and SW) and a ˜ 8% increase in SL.

Notwithstanding the lower contribution of knee than ankle muscles to limb mechanical work during walking, previous studies revealed that quadriceps muscle fatigue induced by RSTS modified contributions of the non-fatigued ankle muscles to gait [33]: Ankle joint work increased ˜2 times while stepping down from a curb after RSTS [33]. Additionally, prior EMG data indicated that older women increased quadriceps activity during the stance phase by ˜10% after 20 min of treadmill walking, suggesting that compensation was needed to maintain gait [34].

The small effects induced by the RSTS task on treadmill walking in our study could be related to joint work during gait being submaximal. Ample reserve is left in the quadriceps to respond to the fatigue-induced reductions in force-generating capacity after hundreds of STS movements. Previous data indicated that EMG activation of the quadriceps muscle during habitual gait was less than 25% of the maximal activation [35,36]. It thus appears that, while RSTS substantially reduced the MVF (Fig. 1) and its activation [11], these reductions were too small to necessitate changes in the mechanics of treadmill walking.

It is also conceivable that participants compensated for reduced quadriceps force by increasing the reliance on non-fatigued muscles [33] to keep up with belt speed on the treadmill. Increases in muscle activation by hip flexors and plantarflexors could increase stride length through an increased hip range of motion and more effective push-off, overriding any stride length shortening effects due to quadriceps fatigue. Future studies, thus, should determine if compensatory muscle activation appeared during walking after fatigue.

Our results indicate that sustained mental task did not affect gait outcomes, agreeing with previous data [15]. However, these authors reported an increase in the CV of speed, SL, stance phase, double support and swing time during dual-task walking, after the mental task [15]. These data seem to support the idea that prolonged mental activity is a reasonable model to study the effects of sustained mental task-induced fatigability on gait, resembling dual- but not single-task walking [15]. Moreover, after the mental tasks, perhaps participants rely on feedback from leg muscles and capitalize on the imposed pattern by the treadmill to compensate for the reduced availability of cognitive resources. Despite minimal fatigability effects on gait, this study proposed novel insights into the understanding how the mental fatigue-induced interference with cognitive functions would affect gait dynamics.

Although older vs. young adults walked with shorter SL (˜6 cm), higher cadence (˜6 steps /min), variability (˜20% of CV in SL and SwT), and λmax in AP and ML (˜30%), unexpectedly, there was no interaction between fatigue and task in our outcomes. Interaction involving age and sustained muscle activity reported previously [9,37], did result in more pronounced adjustments in stride length (˜4%), duration (˜4%) and speed (up to 8%) in older compared with young adults during overground walking [9] and dual-task walking [37]. It seems to complement the argument that overground compared with treadmill walking represents a more complex and challenging task, and dual-task walking requires greater sensory integration and executive function that could amplify the fatigability effects.

A limitation of this is that older and young were similar in physical performance, global cognition, and trait of fatigue, minimizing the effects of age and experimental manipulations on gait. Duration of the RSTS varied between participants. However, the 16% average force loss we observed was greater than reductions reported previously [8,9]. Fatigability in the plantarflexors or combined knee and ankle exercises

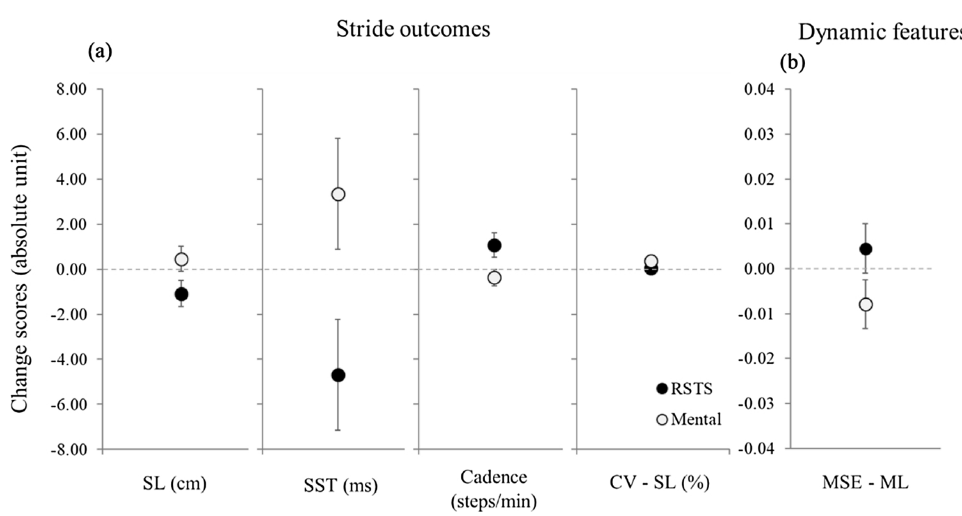

Fig. 2. Change score computed as post fatigue minus pre fatigue (Y-axis), considering (a) stride outcomes, and (b) dynamics features of gait (X-axis) for RSTS (black dots) and mental task (grey dots), independent of the age-groups.

could be effective to probe the effects of age and fatiguability on gait [14]. Concerning mental tasks, this protocol appears to be effective to examine age-related changes in dual-task walking [15]. A comparison between treadmill vs. real world walking in the fatigue context would provide information if the treadmill does indeed minimize fatigability-effects on gait.

5. Conclusion

Muscle and mental fatigability had minimal effects on gait in young and healthy older adults possibly because treadmill walking makes gait uniform. It is still possible that age-dependent muscle activation underlies the uniform gait on the treadmill. Age- and fatigability effects might be more overt during real life compared with treadmill walking, creating a more effective model for examining gait and age adaptability to fatigability perturbations.

CRediT authorship contribution statement

Paulo Cezar Rocha dos Santos: Conceptualization, Project administration, Data curation, Formal analysis, Investigation, Methodology, Visualization, Writing - original draft, Writing - review & editing. **Tibor Hortobágyi:** Conceptualization, Formal analysis, Methodology, Supervision, Visualization, Writing - review & editing. **Inge Zijdewind:** Conceptualization, Methodology, Writing - review & editing. **Lilian Teresa Bucken Gobbi:** Conceptualization. **Fabio Augusto Barbieri:** Conceptualization, Methodology, Writing - review & editing. **Claudine Lamoth:** Conceptualization, Data curation, Formal analysis, Visualization, Methodology, Supervision, Writing - review & editing.

Declaration of Competing Interest

None.

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Appendix A. Supplementary data

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