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ORIGINAL ARTICLE



The association between sedentary behavior and cognitive ability in older adults

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Abstract

Executive functions (EF) are a grouping of cognitive abilities essential for daily life. Previous research has shown that physical activity (PA) may in fact preserve EF in older adults, but the link between sedentary behavior (SB) and cognitive ability has been less explored. The purpose of this study was to examine the relationship between SB and cognition (executive function and memory) in older adults. Seventy five older adults (74.6 ± 9 years) self-reported their sedentary time (ST) and PA, as well as EF ability (paper-based measure of EF). Participants also completed several performance-based measures of EF and a memory task. Older adults who were less sedentary had superior EF and memory (e.g., Stroop time was significantly faster in less sedentary adults ($34.7 \text{ s} \pm 1.9$) compared to more sedentary adults ($39.6 \text{ s} \pm 1.8$), p = .02). Regression analysis showed that total ST was associated with several measures of EF after adjusting for age, and physical activity (e.g., Stroop time $\beta = .005$ (.002, .009). Less cognitively demanding SB (TV viewing and napping) was associated with worse performance on most EF and in the memory task. Performing a hobby was also associated with lower levels of EF and memory. For example, the building times for the Lego task were positively related to napping ($r^2 = .34$), watching TV ($r^2 = .27$), and performing a hobby ($r^2 = .46$). Associations of ST with cognitive abilities were more pronounced in older adults who engaged in less PA. These results suggest that SB may play an important role in cognitive abilities of older adults. Longitudinal studies using performance-based assessments of EF are needed. Lara Coelho and Kayla Hauck contributed equally to the manuscript.

Keywords Exectuive function · Sedentary behaviour · Aging · Memory

Lara Coelho and Kayla Hauck contributed equally to the manuscript.

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Introduction

Executive functions (EF), a subtype of cognitive ability, are a group of mental processes that are mediated by the prefrontal cortex [1]. These mental processes include: impulse control, task switching, working memory, inhibition, processing speed, planning, cognitive flexibility, self-regulation, and socio-emotional control [2–4]. Previous research has shown that EF declines as we age [5–7]. EF abilities are critical, as they serve to plan activities, shift attention, and organize our lives. Importantly, EF ability predicts the likelihood that older adults remain living independently [8]; it is, therefore, important to find ways to maintain EF as we age.

One potential strategy for preserving cognitive ability (both EF and memory) is physical activity (PA). There are numerous physical health benefits associated with regular PA in older adults [9], and some studies have also found a positive association between PA and EF (for review: [10]). For example, exercise-training programs have been shown to



improve processing speed, memory, and overall EF [11–13]. In addition to protecting EF, increased PA has been found to reduce the risk of dementia, therefore preserving memory in older adults [14].

Although the benefits of PA for healthy aging have long been recognized, the potential risks of prolonged periods of sedentary behavior have emerged more recently. Sedentary behavior is defined as any waking activity in a seated or reclined posture, with a low energy expenditure [<1.5 metabolic equivalents (METs; [15])]. Among older adults, sedentary time is associated with increased risk of cardiometabolic disease, all-cause mortality, and impaired physical function [16]. There also appears to be an inverse relationship between sedentary time and cognitive abilities, although reports regarding this association are inconsistent. One challenge is that not all sedentary behaviors are the same. Passive sedentary behaviors, such as TV viewing, appear to be detrimental [17]; while more cognitively stimulating activities, such as reading, using computers, or doing puzzles, may be less deleterious and even associated with better cognitive ability [17, 18]. Another issue is the significant heterogeneity across studies in the measurement of cognition [19]. A recent systematic review concluded that while the available evidence suggests that sedentary time is associated with cognition among older adults, the nature of the relationship is largely dependent on how cognitive ability is defined and measured [19]. Finally, there is considerable debate about whether or not sedentary time affects health independently of physical activity [20]. Among older adults, it appears that habitual physical activity provides protection against the effects of sedentary time on some health risks, including diabetes [21] and mortality [22]. While the negative consequences of prolonged sedentary time appear to be greatest among people who are inactive, to date few studies have examined the interaction between physical activity and sedentary time on cognitive ability among older adults.

In light of the limitations to the currently available evidence, the association between sedentary behavior and cognitive ability was identified as a research priority in an international consensus statement on sedentary behavior in older adults [23]. Thus, this study had two main objectives. The first was to examine the relationship between sedentary time (ST) and cognitive ability using a robust assessment of cognitive abilities. We included both paper- and performance-based tests to assess working memory and EF across several domains. The second objective was to determine whether the association between ST and EF is different among individuals who engage in regular physical activity compared to those who are inactive.



Participants

Seventy-seven adults volunteered to participate in this study. Participants were recruited via flyers and communication with local seniors groups and organizations. Two participants did not complete the entire study so they were subsequently excluded from the analyses. The remaining 75 participants (15 males and 60 females) were between 52 and 97 years (mean: 74.6±9 SD years). None of the participants were diagnosed with dementia or any other cognitive disorders. All procedures were approved by the University of Lethbridge Human Participant Research Committee (Protocol #2015-015), and all participants gave written informed consent.

Procedures

All participants completed a battery of paper-based and performance-based measures of EF, and a memory test, in addition to reporting their perceived handedness, physical activity, and sedentary behaviors. Participants completed the study in a single 1-h session either at the University of Lethbridge, or in their residence.

Measures

Sedentary time

Sedentary time (ST) was assessed using the Longitudinal Aging Study Amsterdam Sedentary Behavior questionnaire (LASA-SB; [24]). The LASA asks about time spent in 10 different sedentary behaviors on an average weekday (Monday to Friday) and weekend day (Saturday or Sunday). The questionnaire has been validated for use with older adults and shows acceptable reliability (r = .71) and moderate correlation with device-based measures of sedentary time (r=.46) [24]. Visser and Koster [24] found that six activities correlated best (r = .46, p < .05) with total device-measured sedentary time: napping, reading, listening to music, watching TV, performing a hobby, and talking to friends. We included these activities into our analyses. For the statistical analyses, we used both the total sedentary time (weekday + weekend), as well as the time spent on each of the six sedentary behaviors identified by Visser and Koster [24]. We also separated the participants by sedentary level using the LASA-SB questionnaire for further analysis. We averaged the total sedentary time on the week and weekend days for each participant; 9.4 h is the average sedentary time among older adults across 10 different countries. [25], so a score



of > 9.4 h was classified as "more sedentary" (n=32) and a score of \leq 9.4 h was classified as "less sedentary" (n=43).

Physical activity

PA was assessed using the Godin Leisure-Time Physical Activity Questionnaire (GLTE; [26]), which evaluates time spent in light-, moderate-, and strenuous-intensity activity during a typical 7-day period. The amount of times per week the participants engaged in strenuous activity (heart beats rapidly) was multiplied by 9 for the strenuous Godin scale score. For the moderate exercise (non-exhausting) Godin scale score, the amount of times per week were multiplied by 5. The 2 Godin scale scores (strenuous, moderate) were added together for the total leisure score index (LSI). A score > 24 was classified as "physically active" (n = 37; 9 males; age = 73.8 ± 8.6), while a score of of ≤ 24 was classified as "less active" (n = 38; 6 males; age = 75.3 ± 9.4) [27].

Paper-based Executive function

Participants completed the adult version of the Behavioral Rating Inventory of Executive Function (BRIEF-A), which measures one's self-reported EF [28, 29]. Participants completed 75 questions which asked them to rate how often each behavior had been a problem within the past month as either never, sometimes, or often a problem. Each reported behavior belongs to one of eight subscales: (1) Inhibit; (2) Shift; (3) Emotional Control; (4) Self-Monitor; (5) Initiate; (6) Working Memory; (7) Plan/Organize; (8) Task Monitor. See [29] for a detailed

description regarding of each subscale. The summed responses for the first four subscales (inhibit, shift, emotional control, and self-monitor) define the Behavioral Regulation Index (BRI) and the following five subscales (initiate, working memory, plan/organize, task monitor, and organization of materials) define the Metacognitive Index (MI). The BRI and MI together comprise the Global Executive Composite (GEC). Each raw score collected for the BRI, MI, and GEC categories was standardized for age using the BRIEF-A handbook [29]. The T-score and percentile rank generated for each subject were used in statistical analysis. A higher score on each of the subscales of the BRIEF indicates worse EF.

Performance-based assessment of executive function

All participants completed five performance-based assessments of EF (Stroop, tower of Hanoi, two block building tasks, and snap) that measure a different parameter of EF (see Table 1 for descriptions). All tasks were completed with the participant seated at a table and were video recorded with a Sony[®] Handycam. Details of each test can be found in the supplementary material.

Memory test

The logical memory test (from the third edition of the Wechsler Memory Scale)

The Logical Memory Test involved listening to two different short stories that were read by the experimenter, and then asking the participant to recall as much detail as possible both immediately after and after 30 min. As in [34],

Table 1 Description of performance-based assessments of EF

EF assessment	Description of game	EF targeted
Stroop [30]	Participants read the name of colors that are printed in different colored ink. Time to completion and number of errors are recorded	Behavioral inhibition
Tower of Hanoi [31]	Participants must move a stack of 3 blocks to a final location, only moving one block at a time and not placing a larger block on a smaller block. Time to completion and number of errors are recorded	Working memory, task switching, planning, and organization of materials
Mega and Lego blocks [32]	Participants must replicate several Mega and Lego models. Time to completion and number of errors are recorded. As well as the amount they use their right hand	Working memory, planning, organization of materials
Snap Modified version of Wisconsin card sort [33]	The Participants and experimenter took turns flipping over a card and saying the shape of the object on the card. If the experimenter and participant flipped over the same color card, they had to switch and say the color on the card. We recorded how many mistakes the participants made	Behavioral inhibition, task switch

For the measures of time to completion and errors, lower scores indicate superior EF. For the measure of right-hand use, higher scores indicate superior EF



we measured the amount of details correctly identified from both stories summed as the "first recall total score". After the second reading of the second story, the number of correct responses was summed and combined with their "first recall total score" to generate a "recall total score". After the 30-min break, the subjects were asked again to vocalize everything they could remember from the two stories successively. The amount of detail correctly identified from each story were summed together and labeled as the "delayed recall total score". Subjects were asked various questions regarding each story and they were required to provide a "yes" or "no" answer (and to guess if unsure). In total, participants answered 30 story-specific questions (15 questions referring to each story). Correct responses for both stories were summed and labeled as the subject's "recognition total" score. Lastly, we also calculated percent retention scores. These were defined as the amount of correct response after the 30-min delay, compared to the amount of correct response immediately following the stories being read. For more information on this task, please see [34].

Statistical analysis

Statistical analyses were completed using SPSS version 24 (SPSS Inc., Chicago, IL, USA). Statistical significance was set at p < .05. To examine if less sedentary participants performed differently than the more sedentary participants, we conducted non-parametric Kruskal-Wallis tests, as the data were not normally distributed. Pearson's r correlations were calculated to explore associations between time spent in different sedentary behaviors and cognitive abilities. This was completed to determine which variables to include as the dependent variables in our linear regression analyses. Lastly, we conducted the linear regression analyses to investigate if ST was a significant predictor of cognitive ability, with age and the LSI as covariates. Finally, as previous research has shown that PA can protect against the negative effects of sedentary time [21, 22], we conducted a stratified analysis to examine the association of ST with EF among inactive and active participants.

Table 2 Participant scores on tests of cognitive ability, grouped by self-reported sedentary time

		Less se (n=43	dentary		More seden- N ary $(n=32)$		Kruskal–Wal- lis test statistic	p
		Mean	SE	Mean	SE			
Sedentary time	Total time (h/week)	7.07	.25	14.33	1.16	75	54.32	<.001
Physical activity	Leisure score index	58.2	5.8	33.1	7.1	75	18.84	<.001
BRIEF-A	MI T-Score	52.3	1.4	61.0	2.0	75	9.8	.002
	MI rank	60.3	4.0	78.6	3.1	75	10.17	.001
BRIEF-A	GEC T-score	53.3	1.3	61.4	2.1	75	8.28	.004
	GEC rank	62.7	3.9	77.6	3.3	75	7.38	.007
	BRI T-score	54.0	1.3	60.3	2.2	75	3.7	.054
	BRI rank	64.5	3.7	74.2	4.2	75	3.6	.058
STROOP Time (s)	Incongruent colors	34.7	1.9	39.6	1.8	75	5.8	.016
MEMORY	Recognition	27.1	1.4	23.9	.6	72	7.6	.006
STROOP time (s)	Percent retention	90.4	2.6	79.3	3.7	73	5.74	.017
SMALL LEGO®	Total space	848.3	8.2	882.8	13.9	75	5.28	.022
	Total time (s)	198.1	11.8	242.0	20.6	75	3.95	.047
SMALL LEGO® MEGA Bloks	Building errors	1.5	.3	2.9	.5	75	5.00	.026
	Total errors	3.0	.4	5.0	.7	75	4.6	.032
	Building errors	2.5	.4	4.3	.6	75	5.52	.019
	Total errors	4.7	.5	7.4	1.0	75	4.73	.03
SNAP	Shape errors	1.9	.2	2.7	.3	73	4.11	.043
	Total errors	5.1	.6	6.6	.6	73	3.7	.054

Results were significant at the p < .05 level. df = 1

The remaining tests were not statistically significant between groups (p > .05, data not shown)



Table 3 Pearson's r correlations between time spent in sedentary behaviors and measures of cognitive ability

	Nap	TV	Hobby	Car	Church
BRI Tscore	.13	.25ª	.27ª	.23ª	.11
BRI rank	.11	.23a	.21	.21	.11
MI Tscore	.16	.24a	.21	.14	.11
MI rank	.25a	.24a	.26a	.11	.10
GEC Tscore	.16	.26a	.26a	.20	.12
GEC rank	.22	.25a	$.26^{a}$.16	.12
Stroop time	.21	.30 ^a	.30 ^b	.01	.17
Memory percent recognition	30^{b}	22	19	11	19
Lego total space	.42 ^b	.27a	.25 ^a	.09	.23
Lego total time	.34 ^b	.27a	.46 ^b	03	.09
Lego building errors	.27 ^b	.17	.45 ^b	09	.16
Lego small total errors	.38 ^b	.23a	.43 ^b	.12	.29a

^aCorrelation is statistically significant at the .05 level (2-tailed)

Results

No difference in age was found between the less sedentary (mean age: 76.5 ± 1.4 years) and more sedentary (mean age: 72.0 ± 1.5 years) individuals using a two-tailed t test (p = .5).

Group differences

As seen in Table 2, participants who self-reported as less sedentary also self-reported superior EF on 3 measures of the BRIEF-A when compared to adults who were more sedentary. In addition, less sedentary adults out-performed more sedentary adults on some of the performance-based EF measures. They took less time to complete the incongruent trials on the Stroop test (validated Stroop test), and made fewer shape and total errors on the Snap test. Furthermore, less sedentary adults had better memory according to the memory recognition total score and the percent retention score than participants who were more sedentary. We also

Table 4 Associations of cognitive ability with ST, PA, and age

	Age (B (95% CI)	Leisure score Index	Total sedentary time
BRI T-score	.087 (18, .35)	05 (11, .01)	.005 (.002, .009)
BRI rank	.23 (38, .84)	09 (23, .05)	.008 (.000, .017)
MI T-score	.26 (002, .52)	02 (08, .04)	.006 (.002, .009)
MI rank	.78 (.2, 1.3)	001 (14, .13)	.01 (.004, .02)
GEC T-score	.19 (07, .44)	03 (88, .03)	.006 (.003, .009)
GEC rank	.61 (.04, 1.2)	04(17,.1)	.01 (.003, .018)
Stroop time	.62 (.37, .88)	009 (07, .05)	.005 (.002, .009)
Memory percent recognition	16 (35, .03)	.02 (03, .06)	001 (003, .002)
Lego total space	.59(-2.8,4)	09(87,.69)	.01 (034, .06)
Lego total time	3.6 (1.9, 5.3)	09(48,.3)	.006 (02, .03)
Lego building errors	.15 (.08, .22)	01 (03, .004)	.000 (001, .001)
Lego small total errors	.25 (.18, .32)	001 (02, .01)	.000 (001, .001)

Unstandardized beta coefficients (95% CI)

Significant predictors are bolded

Table 5 Associations of cognitive abilities with ST, PA, and age split between participants who are less active, and those who are more active

	Less active older ad	ults	More active older adults		
	Age (B (95% CI)	Sedentary time	Age	Sedentary time	
BRI T-score	08 (5, .34)	.005 (.001, .01)	.4 (.02, .78)	.001 (009, .01)	
BRI rank	27 (-11, .57)	.007 (002, .02)	1.02 (01, 2.04)	.002 (02, .03)	
MI T-score	.007 (37, .38)	.005 (.001, .004)	.63 (.25, 1.01)	.001 (009, .01)	
MI rank	19 (54, .91)	.009 (.001, .017)	1.6 (.55, 2.6)	.002 (025, .028)	
GEC T-score	04 (42, .34)	.006 (.001, .01)	.55 (.19, .91)	.001 (008, .01)	
GEC rank	06 (67, .79)	.009 (.001, .017)	1.4 (.41, 2.4)	.000 (026, .026)	
Stroop time	.29 (.001, .58)	.004 (.001, .008)	1.1 (.61, 1.5)	.004 (008, .02)	

Unstandardized beta coefficients (95% CI)

Significant predictors are bolded



^bCorrelation is statistically significant at the .01 level (2-tailed)

found that the less sedentary participants made fewer mistakes in both the small and the large block building tasks.

Correlation analysis

Several significant correlations were found between time spent in different sedentary behaviors and scores on tests of cognitive ability (see Table 3 for details). In short, we found that napping, watching TV, and performing a hobby negatively associated with cognitive abilities (EF and memory).

Regression analyses

Results of the linear regression analyses are presented in Table 4. Total ST was significantly associated with several measures of EF after adjusting for age, and physical activity.

The results of the linear regression stratified by the Leisure Score Index are summarized in Table 5. For those participants who engaged in less physical activity, ST predicted scores in the metacognition index (a subscale of the BRIEF) and the global executive composite (GEC). However, in the more active older adults, age was the only significant predictor of EFs.

Discussion

The main finding of this study was that individuals who self-reported less ST had superior EF and memory in both paper- and performance-based measures. Specifically, individuals who were classified as less sedentary had a lower score on the MI and GEC components of the BRIEF-A, took less time to complete the incongruent color trials on the Stroop test, took less time to complete the LEGO® task, made fewer building and total errors on both the small and mega block tasks, and made fewer shape errors on the snap test. They also scored higher on the memory test, as they had better memory recognition and percent retention than their more sedentary peers. Importantly, we found no difference in age between groups, which suggests that our results were not simply due to age-related differences in EF. We further discovered that not all of the SBs in the LASA questionnaire were associated with cognitive abilities. Overall, TV viewing, napping, and performing a hobby were associated with worse EF abilities.

While many studies have investigated the relationship between PA and EF [5, 11–14, 35, 36], there has been less research conducted on how ST is related to EF. Of the research completed on this relationship, there have been conflicting results, which have been attributed to the amount of heterogeneity in the measures of cognitive ability used [19]. Thus, the main objective of this study was to conduct a thorough and rigorous assessment of cognitive

abilities. Both paper-based and performance-based measures showed similar negative associations with sedentary time. Our results are in line with other studies that have documented increased ST leads to worse EF [37, 38], and memory [38]. For example, Kivipelto and colleagues recruited older adults (65–79) and had them self-report PA and ST, and then assessed their memory using the minimental state examination (for description see: [39]). They found that the older adults who were more sedentary were 2.07 times more likely to develop dementia over a 21-year span. The results from our present study add to this body of evidence that like PA, ST impacts cognitive abilities in older adulthood.

In the present study, we found that watching TV, napping, and performing a hobby while seated were all associated with worse cognitive abilities measured by both the paper- and performance-based tests. When we compared all SB measured, watching TV related to most measures of EF as well as to memory. Specifically, TV viewing time correlated with worse self-reported EF on the BRIEF-A (as a higher score is indicative of poorer EF) and worse performance on the Stroop and small LEGO® building tasks (see Table 5). These results are in line with other studies that have suggested some SBs may have more deleterious consequences than others [40, 41], and specifically that TV viewing is the most detrimental to health and cognitive ability [37]. Kesse-Guyot et al. had participants selfreport the time they spent watching TV, using a computer, reading, and engaging in PA. The participants were also required to complete six EF tests. The authors reported a negative association between TV viewing and EF skills. This association was not present in any of the other SBs, in fact there was a positive relationship between the time spent using a computer and verbal memory performance and EF skills. These results suggest that not all SBs have adverse effects on cognitive abilities. The finding that time spent doing a hobby was associated with worse EF was somewhat surprising to us, as previous research has shown that hobbies such as computer use [37], sewing, knitting, and crocheting [42], and engaging in crossword puzzles [43] all benefit EF in older adults. We speculate that the wording on the LASA questionnaire might have influenced our results. For this questionnaire when the participants are required to estimate the amount of time they spend performing a hobby, the two examples given are doing a puzzle and knitting. These may not apply to many participants or may confuse them. Another factor that could have impacted this correlation is that the LASA-SB questionnaire does not require participants to list which hobbies they specifically perform, so future studies should consider adding this information. The fact that TV watching, napping, and hobbies have been previously shown to be most



strongly related to device-measured sedentary time [24] adds support to our findings.

The results of our linear regressions indicate that ST is a significant predictor of several measures of EF (all measures of the BRIEF and Stroop time), while PA does not predict any EF measure. Previous research has suggested that PA and SB have separate impacts on health [44, 45]; our findings indicate that these activities also play independent roles in EF. In fact, for every added minute of ST, the participants' scores on the MI and GEC rank (subscales of the BRIEF) were increased by .01. This means that an added hour of sitting results in an increase of .6 to a participants score. This increase is the equivalent of a participant changing their response (on a question on the BRIEF) from sometimes to always. On the BRIEF, a higher score indicates poor EF ability. Therefore, the positive unstandardized beta coefficients indicate an increase in ST results in worse EF. It is important to find ways to strengthen EFs especially in the older adult population who may already be experiencing age-related declines in cognitive abilities [5]. The results from our study suggest that decreasing ST is a statistically viable option to increase EF abilities in this population. This could ultimately play a role in maintaining independence into older adulthood [8]. Future studies should investigate at what point behavioral changes in EF become apparent following a reduction of ST (i.e., how much of a reduction of ST is necessary to see clinical changes in EF).

Previous research has shown that the effects of sedentary time on cardiometabolic health and mortality are most pronounced among people with low physical activity [21, 22, 46] Our results suggest a similar relationship for cognitive ability, with ST having detrimental associations with cognitive abilities only among those classified as less active (LSI < 24). We found that ST was a significant predictor of the BRI, MI, and GEC t-scores, as well as the MI and GEC rank, and the Stroop time among less physically active older adults. This can be interpreted as the participants with high sedentary time and low physical activity levels having worse behavioral regulation (BRI), self-managing and monitoring (MI), behavioral inhibition (Stroop) and overall worse EF (GEC) the more time they spend participating in sedentary activities. Among more active participants, only age was a significant predictor of cognitive ability, with no significant associations observed with sedentary time. These findings suggest that promoting physical activity remains an important population health priority for older adults, and also that reducing sedentary time among older people who have low levels of physical activity may also be an important goal.

An important strength of this study is the thorough and rigorous assessment of cognitive ability (EF and memory). The limitations of this study include a reliance on self-reported physical activity and sedentary time, although we did use validated questionnaires. Although we confirmed

that participants had not been diagnosed with cognitive impairment, we did not collect other medical history or the use of pharmaceuticals in this population; medication or other undisclosed conditions could have potentially influenced the results. The cross-sectional study design is also a limitation since we cannot rule out reverse causality; it is possible that individuals with better cognitive abilities are less likely to engage in long periods of passive sedentary behaviors like TV. Furthermore, previous research has identified a robust bi-directional relationship between PA and EF, where changes in EF can promote PA participation, and increased PA can improve or sustain EF ability later in life [47]. In addition, we had no information regarding their participation in SB across their lifespan, which could have impacted our findings regarding EF and memory. It is possible that lower ST in childhood or early adulthood influence cognitive development and preserve EF and memory later in life. These are important questions to consider for future research.

To conclude, older adults who engage in less ST outperform more sedentary peers on a variety of EF and memory tasks, and time spent in different types of SB (TV watching, reading, listening to music, etc.) appeared to correlate differently with cognition. We also demonstrated that the negative associations of ST with cognitive abilities are significant primarily among older adults who are less physically active. As the population ages, strategies to preserve cognitive abilities become increasingly important; initiatives that promote physical activity and reduce passive sedentary time are needed.

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Compliance with ethical standards

Conflict of interest statement On behalf of all authors, the corresponding author states that there is no conflict of interest.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Statement of human and animal rights All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional research committee (University of Lethbridge Human Subject Research Committee) and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent Informed consent was obtained from all individual participants included in the study.



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