



Physical activity and sedentary time: associations with fatigue, pain, and depressive symptoms over 4 years post-treatment among breast cancer survivors

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Received: 16 November 2020 / Accepted: 25 July 2021 / Published online: 13 August 2021
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Abstract

Introduction Despite the recommendations for cancer survivors to engage in physical activity (PA), little is known about the effects of both PA and sedentary time (ST) on key health symptoms. This study prospectively examined the lifestyle behaviors of moderate-to-vigorous PA (MVPA) and ST as predictors of depressive symptoms, pain, and fatigue in breast cancer survivors using longitudinal data from early post-treatment to 4-year survivorship.

Methods Breast cancer survivors ($n = 199$, mean(SD) age = 55.0(11.0) years) self-reported depressive symptoms, pain, and fatigue, and wore an accelerometer to measure MVPA and ST every 3 months during the first year (times 1 to 5) and 2 and 4 years (times 6 and 8) post-cancer treatment. Linear mixed models were adjusted for personal (e.g., age, BMI, education) and cancer (e.g., stage, time since treatment) variables.

Results MVPA and ST were independent predictors of depressive symptoms, but not fatigue, and only ST was associated with pain over 4 years post-treatment. Higher levels of MVPA were associated with lower scores of depressive symptoms (β (95%CI): -0.062 ($-0.092, -0.031$) $p < .001$), whereas higher levels of ST were associated with higher scores of depressive symptoms (β (95%CI): 0.023 ($0.017, 0.028$) $p < .001$). Higher levels of ST were associated with increased pain level over time (β (95%CI): 0.017 ($0.007, 0.027$) $p = .001$).

Conclusions Rehabilitation interventions should aim to both increase MVPA and reduce ST to promote health and well-being among breast cancer survivors, in particular during the early post-treatment period.

Keywords Physical activity · Sedentary time · Accelerometer · Pain · Fatigue · Depressive symptoms · Breast cancer survivors · Longitudinal study

Introduction

Breast cancer remains the highest incident cancer diagnosis in Canadian females [1]. Valuable progress in breast cancer survival has been achieved (5-year survival rate of 88% in Canada) as a result of advances in prevention, screening, early detection, and combined modality treatment [1]. As survival rates increase, a rise in symptom burden secondary to breast cancer survivorship is common [2]. Side effects can develop as early as immediately after cancer diagnosis, usually becoming more severe during cancer treatment and can continue for months and even years [3]. Fatigue, pain, and depressive symptoms are among the most prevalent long-term breast cancer effects that undermine physical and psychosocial functioning [2].

Breast cancer survivors report severe chronic pain [4], debilitating fatigue [5], and depressive symptoms [6, 7] that

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independently and together interfere with physical and psychological functioning, quality of life, distress, and many comorbidities long after treatments have ended [5–10]. As such, it is important to identify strategies to reduce the experience of these symptoms.

Physical activity (PA) is a safe, feasible, and effective strategy that could be targeted to prevent and reduce late-effect symptoms of pain [11], fatigue [12, 13], and depression [14] among breast cancer survivors. The American College of Sports Medicine published the first recommendations in 2010 suggesting that cancer patients and survivors should (i) avoid physical inactivity and (ii) aim for 150 min of moderate-to-vigorous PA (MVPA) per week [15]. Recently, updated evidence-based guidelines suggest that 90 min per week of MVPA may be valuable to health outcomes [16]. Specifically, MVPA has been associated with a significant reduction in cancer-related fatigue in breast cancer survivors [13]. Higher levels of MVPA [17] and even light intensity PA [18] are associated with fewer depressive symptoms and breast cancer survivors who were meeting MVPA guidelines were less likely to report above-average pain [19].

In addition to PA, sedentary time (ST) (i.e., resting activity < 1.5 METS in a sitting or reclined position) [20] is an independent risk factor of cancer mortality and morbidity [21]. Reducing ST time in breast cancer survivors may be a more feasible behavioral target to reduce symptoms of pain, fatigue, and depression, when compared to attempts at increasing MVPA [22]. ST has been associated with poorer quality of life [23], decreased wellbeing, and increased fatigue duration [24] in breast cancer patients and survivors. Little is known about the relationships and interactions among MVPA and ST on pain, fatigue, and depression symptoms over time among breast cancer survivors.

The purpose of this study was to prospectively examine the association between lifestyle behaviors of MVPA and ST and depressive symptoms, fatigue, and pain in early post-treatment breast cancer survivors using long-term follow-up over 4 years. The target sample of early post-treatment women following breast cancer diagnosis helps us understand the associations among MVPA, ST, and the common effects of depression, fatigue, and pain during a cancer phase that is rarely explored. Addressing limitations of self-reported behaviors, MVPA and ST were device-measured. It was hypothesized that an increase in ST, in addition to a decrease in MVPA level, would be associated with more symptoms of depression, fatigue, and pain.

Methods

Participants and procedures

Women who had completed primary treatment (i.e., surgery, radiotherapy, chemotherapy) for stages I to III breast cancer

were invited to participate in a longitudinal study investigating the natural developmental changes in lifestyle behaviors. Survivors were recruited through advertisements and oncologist referrals from various local medical clinics and hospitals in Montreal (Canada). Interested survivors were asked to contact the research team by phone to obtain additional details on the study. Screening for eligibility was conducted using the following inclusion criteria: (i) at least 18 years of age, (ii) recently (i.e., 0–20 weeks) post-primary treatment (i.e., surgery, chemotherapy, radiation therapy) for stages I to III breast cancer, (iii) treated for a first cancer diagnosis, (iv) report no health concerns that may inhibit participation in PA, and (v) provide written informed consent in either English or French. Involvement in this study included completing self-report questionnaires every 3 months for 5 times over the first 12 months of the study (times 1 to 5) and once a year thereafter respectively 24, 36, 48, and 60 months after study inception (times 6 to 9). Participants wore an accelerometer to measure PA and ST for 7 consecutive days at data collection times 1 to 5, 6, and 8. The study methods are reported in more detail elsewhere [25]. This study was approved by appropriate hospital and university research ethics committees. All participants provided written informed consent.

Measures

Lifestyle behaviors (MVPA and ST) were measured using Actigraph GT3X accelerometers (Actigraph, Pensacola, FL) every 3 months during the first year (times 1 to 5) and 2 and 4 years (times 6 and 8) post-cancer treatment. During each data collection cycle used in the present study, breast cancer survivors were asked to wear the accelerometer on their hip during waking hours over 7 days, except when participating in water activities (e.g., bathing/showering, pool activities). Movement data were recorded every minute and the number of daily minutes of light (100 to 1951 counts/min), moderate (1952–5724 counts/min), and vigorous (> 5725 counts/min) PA was calculated using established cut points [26]. MVPA was a sum score of weekly minutes in MVPA. ST was analyzed as < 100 counts/minute, adjusted for non-wear time operationalized as at least 60 min of consecutive zeros [27]. Data were included in the analyses if there were no extreme counts (> 20,000) and if data were available for at least 500 min on 4 or more days. The lower limit of wear time compared to established criteria of 600 min was consistent with participants' daily diary records for time awake [25]. For the current study, MVPA and ST variables reflect the average percent of the day spent in MVPA and ST (e.g., time in MVPA and ST accounting for time the accelerometer was worn) using the following formula: Total = [Day 1(Total time in MVPA /Total time accelerometer worn) + Day 2(Total time in MVPA /Total

time accelerometer worn) + ...Day 7 (Total time in MVPA /Total time accelerometer worn)]/7.

Depressive symptoms were assessed using the 10-item Center for Epidemiologic Studies Depression Scale (CES-D) [28]. Participants were asked to rate how often they felt each of the 10 items state (e.g., I felt depressed; I felt fearful) on a 4-point Likert scale (*rarely or none of the time* (0), *some or little of the time* (1), *occasionally or a moderate amount of time* (2); and *most or all of the time* (3)). The global depressive symptoms score was calculated by taking the average of these 10 items at each of the seven data collections. Similar to previous reports in HIV-positive people [28], our findings support the internal consistency of the score across data collections; Cronbach's alpha coefficients range $\alpha = 0.82$ – 0.86 .

Fatigue was measured using the Brief Fatigue Inventory (BFI) [29] which provides an assessment of the fatigue severity, amount of interference with function caused by fatigue, and the presence of factors that worsen fatigue, such as pain and medications. Three items ask participants to rate the severity of their fatigue (e.g., weariness, tiredness) by circling the one number that best describes their fatigue RIGHT NOW (item 1), USUALLY in the past 24 h (item 2), and their WORST fatigue in the past 24 h (item 3), ranging from *no fatigue* (0) to *as bad as you can imagine* (10). Six items assess the amount that fatigue has interfered with different aspects of the participant's life during the past 24 h: general activities, mood, walking ability, normal work, relations with others, enjoyment of life. Each item is scored on a 0–10 continuum, ranging from *does not interfere* (0) to *completely interferes* (10). A global score was calculated by taking the average of the 9 items. Cronbach's alpha coefficients for internal consistency range from $\alpha = 0.94$ to 0.95 across data collections. Fatigue was assessed using the BFI starting at time 2 (3 months after study inception) and then at all time points thereafter.

Pain was assessed asking participants to report whether (yes/no) they experienced any of 6 acute pain symptoms during the day for 2 nonconsecutive days in each week of data collection: stomach pain, back pain, pain in arms, legs or joints (knees, hips, etc.), pain or problems during sexual intercourse, headaches, and chest pain [30]. A score (0–6) is obtained by summing the number of acute pain symptoms experienced during this period, with higher score representing higher level of pain and averaging across the 2 days per data collection. Many of these symptoms have been shown to be prevalent among breast cancer survivors and were drawn from the Primary Care Evaluation of Mental Disorders screening questionnaire (PRIME MD) [31].

Covariates Participants were asked to self-report their age, highest education level attained, income, marital status, breast cancer stage, and time since treatment was received. A trained lab technician measured and recorded baseline

height and weight, which were used to calculate body mass index (BMI; i.e., weight in kilograms divided by height in meters squared).

Analysis

Preliminary analyses comprised descriptive statistics to assess distributions, identify outliers, and compute proportions, means, and standard deviations. Linear mixed effects modeling (LMM) was used to account for repeated measures. Separate models were estimated for each health outcome (i.e., depressive symptoms, fatigue, pain). The final estimated models included fixed effects for MVPA and ST, a time variable, and were adjusted for covariates. The models also included random intercepts and slopes, to allow participants to differ at baseline and also over time (i.e., women can follow different trajectories over time), as well as an “exchangeable” one-parameter correlation matrix. Models were compared using Akaike information criterion (AIC) goodness of fit index [32]. The software R 3.4.2 was used as well as the package *nlme*, Version 3.1–137. A significance level of 5% was used. In sensitivity analyses, we considered models in which covariances varied over time with no meaningful difference observed.

Results

Participants include 199 women breast cancer survivors (mean (SD) age = 55.0 (11.0) years, 85% White/Caucasian) recruited at baseline with stage I ($n = 83$), II ($n = 78$), or III ($n = 38$) breast cancer ($n = 2$; did not indicate). On average, participants had a household income of \$102,041 (SD = 185,217), a BMI of 26.06 (SD = 6.15) kg/m², and were 3.5 months (SD = 2.3) post-surgery, chemotherapy, and/or radiation. Most participants reported having a university degree or higher education ($n = 101$; 50.7%) and being married ($n = 128$; 64.3%) (see Table 1 for sample descriptive statistics). A total of 199 participants provide data at study inception (time 1); in times 2, 3, 4, 5, 6, and 8, n (% of the initial sample) 186 (93.5%), 181 (91.0%), 180 (90.4%), 180 (90.4%), 100 (50.3%), and 85 (42.7%) participants provide data. For the main analysis, only observations with complete data for all variable of interest (questionnaire and accelerometer data) were included in the analysis: a total of 835 observations were included in the analyses for depression symptoms, 826 observations were included in the analyses for pain, and 671 observations were included in the analyses exploring fatigue as the outcome. There was no difference between participants at time 8 (60 months) ($n = 85$) and those who were lost to follow-up ($n = 114$) over a 4-year and seven data collection points time-frame for MVPA (mean(SD) = 1.79(1.32) vs. 2.01(1.49),

Table 1 Characteristics of participants ($n = 199$)

| | Range | n (%) or mean (SD) or median |
|---|----------------|--------------------------------|
| Age (y), mean (SD) | 28–79 | 55.0 (11.0) |
| Race — Caucasian, n (%) | - | 169 (84.9) |
| Income, mean (SD) | 9000–2,000,000 | 102,041 (185,217) ^a |
| BMI (kg/m^2), mean (SD) | 18.1–50.2 | 26.3 (5.7) |
| Education — university level, n (%) | | |
| < High school | - | 11 (5.5) |
| High school diploma | - | 30 (15.1) |
| Some post-secondary | - | 18 (9.0) |
| College/technical diploma/certificate | - | 39 (19.6) |
| University degree | - | 55 (27.6) |
| Postgraduate degree | - | 46 (23.1) |
| Marital status | | |
| Single | - | 27 (13.6) |
| Married/common-law | - | 128 (64.6) |
| Separated | - | 5 (2.5) |
| Divorced | - | 28 (14.2) |
| Widowed | - | 11 (5.5) |
| Breast cancer stage, n (%) | | |
| I | - | 83 (41.7) |
| II | - | 78 (39.2) |
| III | - | 38 (19.1) |
| Treatments received, n (%) | | |
| Lymph or axillary node dissection | - | 116 (58.3) |
| Lumpectomy | - | 119 (59.8) |
| Single mastectomy | - | 56 (28.1) |
| Double mastectomy | - | 34 (17.1) |
| Chemotherapy | - | 128 (64.3) |
| Radiotherapy | - | 176 (88.4) |
| Hormonal therapy | - | 101 (50.8) |
| Time since treatment (months), mean (SD) | 0–8 | 3.5 (2.3) |

BMI, body mass index; SD, standard deviation; y, years

^aInformation not declared for 37 participants

$p = 0.286$), depression symptoms (mean(SD) = 1.75(0.49) vs. 1.73(0.55), $p = 0.779$), and pain (mean(SD) = 1.26(1.10) vs. 1.25(1.08), $p = 0.988$) at time 1 (0 month) and fatigue (mean(SD) = 3.45(2.18) vs. 3.13(2.36), $p = 0.342$) at time 2 (3 months) (fatigue was not assessed at time 1; this is a 3-month difference between time 1 and time 2). Participants who remain involve in the study at time 8 (60 months) report lower ST at time 1 (0 month) compared to those lost to follow-up (mean(SD) = 79.13(5.74) vs. 77.06(5.49), $p = 0.011$).

Descriptive statistics of percent of day spent in MVPA and ST, depressive symptoms, fatigue, and pain scores across all data collections are reported in Table 2. Across the seven data collections, mean accelerometer wear times were 812.6 (SD = 123.9) to 841.1 (SD = 94.1) min/day, and the median number of days worn per week was seven. Average percent of day spent in MVPA remained relatively stable over the first-year post-treatment (ranged from 1.81%

(SD = 1.37) to 1.98% (1.52)) and increased at 24 months post-treatment (3.04%, SD = 2.63) and then remained stable at 48 months post-treatment (2.98%, SD = 2.72). Average percent of day spent in ST ranged from 77.75% (SD = 5.84) to 78.55% (SD = 5.92) during the first-year post-treatment, then decreased to 64.01% (SD = 9.45) at 24 months and 65.92% (SD = 11.23) at 48 months post treatment. Average scores for depressive symptoms, fatigue, and pain all decreased over time during the 4 years post-treatment.

In linear mixed effects models, MVPA and ST were independent predictors of depressive symptoms, but not fatigue, and only ST was associated with pain over 4 years post-treatment, after adjustment for potential confounders. Higher levels of MVPA were associated with lower scores of depressive symptoms (β (95%CI): -0.062 (-0.092, -0.031) $p < 0.001$), whereas higher levels of ST were associated with higher scores of depressive symptoms (β (95%CI):

Table 2 Mean scores for lifestyles and health outcomes at each data collection ($n = 199$)

| | Data collection cycle (months since study inception) | | | | | | |
|---------------------------|--|----------------------|----------------------|----------------------|-----------------------|-----------------------|-----------------------|
| | Time 1 (0 month) | Time 2 (3 months) | Time 3 (6 months) | Time 4 (9 months) | Time 5 (12 months) | Time 6 (24 months) | Time 8 (48 months) |
| Scores, mean (SD) | | | | | | | |
| MVPA, % of day | 1.88 (1.39) | 1.98 (1.52) | 1.92 (1.54) | 1.81 (1.37) | 1.71 (1.38) | 3.04 (2.63) | 2.98 (2.72) |
| ST, % of day | 78.24 (5.72) | 77.75 (5.84) | 77.86 (5.77) | 78.25 (6.00) | 78.55 (5.92) | 64.01 (9.45) | 65.92 (11.23) |
| Pain (0–6) | 1.26 (1.09) | 1.23 (1.15) | 1.27 (1.24) | 1.25 (1.22) | 1.11 (1.12) | 0.17 (0.18) | 0.20 (0.20) |
| Fatigue (0–10) | - | 3.3 (2.26) | 3.02 (2.12) | 3.15 (2.30) | 3.13 (2.44) | 3.21 (2.29) | 2.74 (2.31) |
| Depressive symptoms (0–3) | 1.74 (0.51) | 1.77 (0.57) | 1.65 (0.47) | 1.71 (0.54) | 1.68 (0.53) | 0.66 (0.49) | 0.53 (0.35) |

MVPA, moderate-to-vigorous intensity physical activity; ST, sedentary time; SD, standard deviation; y, years

0.023 (0.017, 0.028) $p < 0.001$). Higher levels of ST were associated with increased pain level over time (β (95%CI): 0.017 (0.007, 0.027) $p = 0.001$). No statistically significant association was found for MVPA in the pain model (β (95%CI): -0.053 ($-0.113, 0.007$) $p > 0.05$). The interaction between MVPA and ST was statistically non-significant. Detailed results are presented in Table 3.

Discussion

This study shows that both MVPA and ST were independently associated with fewer depressive symptoms over time. ST, but not MVPA, was associated with higher pain levels. Neither MVPA nor ST were associated with fatigue over time. This study innovates by including both device-measured MVPA and ST in the same model and by combining the use of multiple time points during the first year following treatment (5 time-points every 3 months) with a 24-month and a 48-month follow-up.

The link between MVPA and depressive symptoms is well established in the literature, both during and after treatment [17, 18]. Similarly, we found in the current study that higher levels of MVPA were associated with lower depressive symptoms. Moreover, our results show

that the association is maintained even when including time points up to 48 months after treatment completion. The results of the current study expand on previous cross-sectional findings reported by Trinh et al. [22] suggesting that higher level of ST is associated with more depressive symptoms over time. However, Phillips et al. [24] report no cross-sectional association between ST and depression in breast cancer survivors. Based on the current study results, observing that depressive symptoms diminish over time allows us to capture the variation in the severity of symptoms while also exploring the association with ST and MVPA.

Contrary to previous studies [12, 13], our results did not show an association between MVPA or ST and fatigue. This difference could be explained by including both MVPA and ST in the analysis model. Current literature contains mixed results regarding the association between ST and fatigue. Phillips et al. [24] found that ST was positively associated with fatigue duration, but not with fatigue interference or severity whereas George et al. (2013) found no association between ST and fatigue 3.5 years post-diagnosis in breast cancer survivors [24, 33]. These inconsistencies may be due to fatigue measurement [12]. Moreover, fatigue measurement in longitudinal studies can be altered by the patient experience factors such as the beliefs and attitudes that

Table 3 Beta coefficients and 95% confidence intervals of the association between lifestyles, time, and health outcomes ($n = 199$)

| | Depressive symptoms β (95% IC) | Pain β (95% IC) | Fatigue β (95% IC) |
|------|--|--|------------------------------|
| Time | -0.025 ($-0.029, -0.022$) | -0.025 ($-0.033, -0.018$) | -0.012 ($-0.029, 0.004$) |
| MVPA | -0.062 ($-0.092, -0.031$) | -0.053 ($-0.113, 0.007$) | -0.134 ($-0.028, 0.014$) |
| ST | 0.023 ($0.017, 0.028$) | 0.017 ($0.007, 0.027$) | 0.005 ($-0.018, 0.028$) |

Models adjusted for age, BMI, highest education level, income, marital status, breast cancer stage, and time since treatment

Bold indicates statistically significant result at $p < .05$

patients have about PA as well as their evolving perception of fatigue over time [34].

In the current study, MVPA was not statistically significantly associated with pain; however, the magnitude of the estimate and the upper confidence interval was close to zero suggesting that there might be an association that we were not able to detect due to the relatively small sample size. Nonetheless, increased ST was associated with increased pain. Similarly, previous study found that ST was positively associated with pain; however, the association only appears to be present when MVPA levels are low [22]. Although several studies have shown that PA interventions reduce pain in people with cancer [14, 16], the longitudinal effects of MVPA on pain are less well established. Forsythe et al. (2013) found a negative association between self-reported MVPA and pain only in women who continued to be active 5 to 10 years after diagnosis [19]. In contrast, Alfano et al. (2007) found that higher levels of PA 39 months after diagnosis were associated with increased breast pain [35]. Pain levels were potentially not severe enough in our sample for an effect to be detected, suggesting a ceiling effect; it is more common to see fairly high pain symptoms in women with breast cancer [19].

Strengths and limitations

Strengths of the current study include multiple data collection immediately after cancer treatment and a 4-year follow-up for a total of seven data collections included in the current analysis. Also, MVPA and ST were device-measured which reduce information bias and misclassification [36]. Wearing an accelerometer could encourage participants to be more active and less sedentary which could be reflected in higher MVPA and lower ST levels. However, information bias due to social desirability is likely to be constant across participants and data collections and consequently is unlikely to distort the association between PA, ST, and each health outcomes assessed in the study. Limitations also include the use of self-report questionnaires for depressive symptoms, pain, and fatigue which are subject to misclassification. A multidimensional measure of pain may be more accurate to consider intensity and interference in a person's life separately. At study inception (time 1), fatigue was only assessed as one item within a list of symptoms rather than a multidimensional scale examining the impact of fatigue on daily functioning. The sequential measurement of fatigue was a study design decision to explore predictors of health outcomes in a sequential way. Consequently, fatigue was assessed using the BFI for the first time at time 2 and the main analysis for fatigue started at time 2. High loss at follow-up may result in selection bias. It is possible that participants remaining in the study who provided data at T6 and T8 (respectively 24 and 48 months after study inception) may be more active

and less sedentary compared to women who did not continue in the study. For example, Shi et al. (2020) investigated PA and ST trajectories in female breast cancer survivors and observed decreasing or stable low self-reported MVPA trajectories during the first 24 months following cancer [37]. They also found four distinct ST trajectories (high maintainer (18%), high decreaser (27%), low increaser (24%), and low maintainer (31%)), suggesting that a quarter of the participants show decreasing ST over time while the other three quarters show increasing or stable ST [37]. The device-measured increasing PA and decreasing ST level observed at 24 and 48 months in the current study need to be replicated.

Although the association between MVPA, ST, and depressive symptoms was found over time, the observational design of the current study and the modeling strategy do not allow inferences of causality. Moreover, the current analyses are exploring the general association between MVPA, ST, and each of depressive symptoms, fatigue, and pain over a 4-year follow-up. However, it is possible that these associations differ throughout the survivorship (early vs. long-term survivorship). This study is underpowered to test this proposition, and future work is needed. Finally, because the study is limited to women with breast cancer, it is not possible to generalize the results of this study to other cancer populations.

Conclusion

This is the first study to demonstrate that lower ST, in addition to higher MVPA, is associated to lower depressive symptoms and that ST is associated with lower fatigue over a 4-year follow-up. The study shows the importance of these associations over time, not only during early post-treatment when there is a heightened emphasis on care. These findings support international guidelines suggesting targeting physical inactivity and reduce ST among cancer patients to improve physical and psychological outcomes. Our findings support that PA promotion as part of routine care for breast cancer patients and survivors could improve both physical and mental health functioning. Rehabilitation interventions should aim to both increase PA and reduce ST to promote health among breast cancer survivors, both during the early post-treatment period and on the long-term survivorship. Recommendations for cancer patients and survivors should target small changes in lifestyle habits, which are known to be associated not only with the uptake of PA, but also with its maintenance over time [38]. Developing specific interventions for people with cancer that focus on reducing ST could be less of a challenge for people with cancer than increasing MVPA and therefore make it possible to initiate a change in the lifestyle behaviors of this population.

Author contribution ID conceptualized the objectives, conducted all analyses, drafted, and revised the manuscript. CS designed the main study and critically reviewed the manuscript. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

Funding This research was supported by a Canadian Institutes of Health Research grant (#186128).

Data availability The data that supports the findings of this study are available on request from the corresponding author.

Code availability The R code that supports the findings of this study are available on request from the corresponding author.

Declarations

Ethics 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. Ethical approval was obtained from the University of Toronto (REB# 28180).

Consent to participate All participants provided written informed consent.

Consent for publication Not applicable.

Conflict of interest The authors declare no competing interests.

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