Intensity-specific effect of physical activity on urinary levels of 8-hydroxydeoxyguanosine in middle-aged Japanese

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Physical activity (PA) is recommended to both promote and maintain health and prevent cancer by improving the body's DNA repair system, which is considered a mechanism of cancer prevention. However, associations between PA and urinary levels of 8-hydroxydeoxyguanosine (8-OH-dG), which reflects DNA damage, are unclear. This cross-sectional study included 2370 men and 4052 women aged 45–74 years enrolled between 2010 and 2012. Habitual PA was assessed by single-axis accelerometer and urinary 8-OH-dG levels by automated HPLC. Multiple linear regression analysis was used to examine the relationship between log-transformed urinary 8-OH-dG and total PA (TPA) and PA of moderate/vigorous intensity (MVPA; ≥3 metabolic equivalents), with adjustment for age, body mass index, energy intake, alcohol consumption, smoking status, daily coffee drinking, menopause status (in women), and TPA (for MVPA). On multivariate adjustment, urinary 8-OH-dG levels were inversely correlated with TPA ($\beta = -0.020$, $P < 0.01$) in women, and this correlation was not changed by PA intensity. Conversely, urinary 8-OH-dG levels were inversely correlated with MVPA ($\beta = -0.022$, $P < 0.05$) in men, although the correlation with TPA was non-significant. This inverse correlation was clearer in current smokers than in never or former smokers, although the interaction between smoking status and MVPA on urinary 8-OH-dG levels was non-significant. In conclusion, greater TPA in women and greater MVPA in men were correlated with reduction in urinary 8-OH-dG, suggesting sex-specific effects of MVPA and TPA on protection from oxidative DNA damage. Increasing PA may mediate reduction in oxidative stress.

Recent epidemiological studies have reported that physical activity (PA) reduces the risk of many types of cancers such as colon cancer, breast cancer, and lung cancer, among others.[1–4] Although the biological mechanisms underlying the cancer-preventive effects of PA remain unclear, several have been suggested:[5–7] (i) reducing levels of endogenous sex hormones, such as estrogen, progesterone, testosterone, and androgens, and of glucose and insulin through weight loss;[5,7,8] (ii) reducing levels of insulin-like growth factors and their increasing binding proteins;[5,7–9] (iii) enhancing immune function and reducing chronic inflammation;[5,7] and (iv) increasing gut motility and pulmonary ventilation and levels of vitamin D.[10,11]

In addition to the above, adaptive response to oxidative stress through increased PA may also be a mechanism of cancer prevention.[5–9,12–16] Findings from experimental studies have suggested that moderate-intensity exercise enhances activities of certain antioxidant enzymes, such as superoxide dismutase, glutathione peroxidase, catalase, glutathione

reductase, and the DNA repair system, subsequently ameliorating oxidative stress,[14–16] although exhausting exercise may conversely increase oxidative stress and damage DNA.[17,18] However, few epidemiological studies have examined the association between PA and oxidative stress by intensity and duration,[19] and results from observation studies are inconsistent, mainly due to limitations in PA measurement.[4,20]

Here, to examine the effect of TPA and intensity-specific PA on urinary levels of 8-OH-dG, the most extensively studied marker of cellular oxidative stress relevant to carcinogenesis, we investigated the association between urinary levels of 8-OH-dG and objective PA assessed by accelerometer in a middle-aged Japanese population.

Materials and Methods

Study subjects. The Japan Multi-institutional Collaborative Cohort Study in the Saga region (designated the Saga J-MICC

Study) was carried out in 2005–2007 in Saga (Japan).[21] We invited 61 447 residents aged 40–69 years by mail to participate in the baseline survey, 12 078 of whom ultimately agreed to participate. After excluding participants who left the area ($n = 423$), died ($n = 163$), and withdrew from the study ($n = 9$) during follow-up, 11 483 people were invited by mail to participate in a face-to-face second survey between 2010 and 2012, 5 years after the baseline survey. A total of 8454 subjects participated in the second survey at age 45–74 years,[22] and the current study was undertaken in these second survey participants.

Assessment of habitual PA. A waist-worn single-axis accelerometer (Lifecorder; Suzuken, Nagoya, Japan), a valid and reliable objective PA measure,[23] was used for habitual PA assessment. The accelerometer is designed to measure acceleration in the vertical direction, collecting activity data every 4 s, as well as total step counts. Methods of data collection and estimation of PA are described in detail elsewhere.[24] Briefly, the study subjects were instructed to firmly attach the accelerometer to a belt on the waist just above the midline of the thigh (right or left), during all waking hours except when bathing or swimming, for 10 continuous days. The device was returned by mail, and data were processed in Microsoft Excel (Microsoft, Redmond, VA, USA). To eliminate bias from any potential initial changes in PA occurring with the use of accelerometers, data from the first 3 days were excluded from analyses. In addition, only data from the most recent 7 days during which the accelerometer was worn for ≥8 h/day were used for analyses.

Accelerometer wear time was calculated based on an acceleration intensity of ≥0.5 (for example, posture changes), while acceleration intensities of 1–9 were used to discriminate light-intensity PA (<3 METs; acceleration intensities, 1–3), moderate-intensity PA (3–6 METs; acceleration intensities, 4–6), and VPA (>6 METs; acceleration intensities, 7–9), which were calculated from the corresponding METs multiplied by the time (h) spent at the corresponding intensity levels (MET h).[23] The physical activity level was calculated as the total energy expenditure divided by the basal metabolic rate (BMR), which was calculated with the following equation:

$$BMR\ (kcal) = Kb \times BSA \times t \times 1/10\,000,$$

where Kb is the standard value of basal metabolism per BSA per time in the Japanese population (kcal/m²/h)[25] and was set according to each subject's sex and age. Body surface area was calculated with the following equation:[26]

$$BSA\ (cm^2) = weight^{0.444}(kg) \times height^{0.633}(cm) \times 88.83.$$

Questionnaire and anthropometric measurements. A self-administered questionnaire that included items on age, gender, lifestyle-related factors, medical history, and medication status[21] was sent to participants prior to the study. Participants were asked to bring their completed questionnaires to the study site. On the survey day, a research nurse or nutritionist asked participants about any missing or inconsistent answers.[22] Details regarding methods of assessment, including dietary assessment, have been reported elsewhere.[27] Anthropometric characteristics (height and weight) were measured to the nearest 0.1 cm and 0.1 kg, respectively. Body mass index was calculated as the weight (kg) divided by the square of the height (m).

Urine sampling and laboratory assay. Participants were asked to provide an 8-mL urine sample taken immediately upon waking. We collected 8405 urine samples from 8454 participants. After using approximately 3 mL to measure density, pH, and levels of protein, bilirubin, ketone, occult blood, and sugar, the remaining urine was transferred to Eppendorf tubes (1.5 mL × 3) and stored at −80°C until needed. Levels of urinary 8-OH-dG and creatinine were measured at the Department of Environmental Oncology, Institute of Industrial Ecological Sciences, University of Occupational and Environmental Health and OHG Institute Co., Ltd. (Kitakyushu, Japan) by an established method using an HPLC system.[28] The accuracy of the measurement, which was estimated based on the recovery of an added 8-OH-dG standard, was reported to be 90–8%, and the coefficient of variation was reported to be ±7% using this method.[29]

Statistical analysis. We excluded from analysis 2032 participants with one or any of the following: (i) urinary 8-OH-dG was

Table 1. Characteristics of study subjects in the second survey of the Japan Multi-institutional Collaborative Cohort Study in the Saga region, 2010–2012

Characteristics	Men ($n = 2370$)	Women ($n = 4052$)	P-value†
Age, years	60.7 [8.1]	60.2 [8.2]	0.01
BMI, kg/m²	23.6 [2.9]	22.2 [3.1]	<0.01
Smoking status (%)			
Never	690 (29.1)	3625 (89.5)	<0.01
Former	1089 (46.0)	238 (5.9)	
Current	591 (24.9)	187 (4.7)	
1–19 cigarettes/day	237 (10.0)	140 (3.5)	
20–39 cigarettes/day	329 (13.9)	47 (1.2)	
≥40 cigarettes/day	25 (1.0)	0 (0.0)	
Alcohol consumption (%)			
Never	466 (19.7)	2434 (60.1)	<0.01
Former	49 (2.1)	31 (0.8)	
Current	1855 (78.3)	1586 (39.1)	
0.1–22.9 g ethanol/day	882 (37.2)	1434 (35.4)	
23.0–45.9 g ethanol/day	526 (22.2)	111 (2.7)	
≥46.0 g ethanol/day	447 (18.9)	41 (1.0)	
Daily coffee drinking (%)	1301 (54.9)	2320 (57.3)	0.07
Daily green tea drinking (%)	1835 (77.4)	3400 (83.9)	<0.01
Daily green or yellow vegetable intake (%)	416 (17.6)	1403 (34.6)	<0.01
Daily fruit intake (%)	240 (10.1)	872 (21.5)	<0.01
Total energy intake, kcal/day	1902 [345]	1502 [235]	<0.01
Vitamin C supplement use (%)	176 (7.4)	427 (10.5)	<0.01
Postmenopausal (%)		3277 (80.9)	–
Steps, count/day	7957 [3658]	7241 [2933]	<0.01
PAL	1.45 [0.11]	1.45 [0.09]	0.40
Total PA, MET h/day	3.92 [1.98]	3.49 [1.59]	<0.01
Light intensity (<3 METs)	2.33 [1.00]	2.18 [0.81]	<0.01
Moderate intensity (3–6 METs)	1.39 [1.30]	1.10 [0.88]	<0.01
Vigorous intensity (>6 METs)	0.21 [0.60]	0.21 [0.53]	0.73
Moderate to vigorous intensity (≥3 METs)	1.59 [1.51]	1.31 [1.14]	<0.01
Urinary 8-OHdG,‡ ng/mg creatinine	3.70 [1.60]	4.10 [1.70]	<0.01

†P-values for sex difference based on t-test for continuous variables and χ²-test for categorical variables. ‡Geometric means [geometric standard deviation]. Data are mean [standard deviation] or number (percentage). –, not applicable; 8-OHdG, 8-hydroxydeoxyguanosine; BMI, body mass index; MET, metabolic equivalent; PA, physical activity; PAL, physical activity level.

Table 2. Characteristics of study subjects in the second survey of the Japan Multi-institutional Collaborative Cohort Study in the Saga region, 2010–2012, according to quartiles (Q) of accelerometer-determined total physical activity

	Men (n = 2370)					Women (n = 4052)				
	Q1	Q2	Q3	Q4	P for trend†	Q1	Q2	Q3	Q4	P for trend†
Total PA, MET h/day	1.88	3.05	4.18	6.58		1.78	2.82	3.76	5.61	
Age, years	62.6	60.1	60.1	60.2	<0.01	62.6	60.0	59.5	58.6	<0.01
BMI, kg/m²	23.9	23.6	23.4	23.4	<0.01	22.6	22.1	22.1	21.8	<0.01
Current smoker, %	29.2	28.5	22.0	20.1	<0.01	6.1	4.4	3.7	4.2	0.19
Current drinker, %	74.0	78.6	80.9	79.6	<0.01	33.5	39.0	40.9	43.3	<0.01
Daily coffee drinking, %	51.5	57.2	55.5	55.5	0.28	50.4	59.0	59.7	59.9	<0.01
Daily green tea drinking, %	76.5	80.3	76.4	76.5	0.30	84.8	84.1	82.5	84.2	0.55
Daily green or yellow vegetable intake, %	17.1	16.5	19.1	17.6	0.69	33.1	37.4	34.4	33.7	0.17
Daily fruit intake, %	10.1	8.9	10.8	10.6	0.71	22.1	21.3	21.9	20.7	0.87
Total energy intake, kcal/day	1846	1884	1929	1950	<0.01	1479	1495	1513	1522	<0.01
Vitamin C supplement use, %	6.4	8.1	8.3	6.9	0.56	10.1	9.5	11.5	11.2	0.43
Postmenopausal, %						86.7	79.3	79.7	77.9	<0.01
PA classified by intensity, MET h/day										
Light (<3 METs)	1.43	2.10	2.55	3.22	<0.01	1.35	1.98	2.41	3.00	<0.01
Moderate (3–6 METs)	0.41	0.86	1.47	2.82	<0.01	0.38	0.73	1.17	2.10	<0.01
Vigorous (>6 METs)	0.04	0.09	0.16	0.54	<0.01	0.06	0.11	0.18	0.50	<0.01
Moderate to vigorous (≥3 METs)	0.45	0.96	1.63	3.36	<0.01	0.44	0.84	1.35	2.61	<0.01
Urinary 8-OHdG, ng/mg creatinine‡	3.76	3.76	3.61	3.59	0.03	4.36	4.11	4.08	3.85	<0.01

†P-value for linear trends across quartiles are based on linear regression analysis for continuous variables and Mantel test for current smoking, drinking, and menopausal status. ‡Geometric means. Data are means. BMI, body mass index; MET, metabolic equivalent; 8-OHdG, 8-hydroxy-deoxyguanosine; PA, physical activity.

not measured (n = 49); (ii) insufficient data on PA (n = 294) data; and (iii) history of diseases possibly associated with change in PA, such as cancer (n = 703), angina pectoris (n = 241), myocardial infarction (n = 103), cerebral infarction (n = 148), cerebral hemorrhage (n = 41), subarachnoid hemorrhage (n = 41), diabetes mellitus (n = 692), liver disease (n = 118), and renal failure (n = 19). After exclusion, data for 2370 men and 4052 women aged 45–74 years were retained for analysis. Among these participants, data on alcohol consumption (1 woman) or smoking status (2 women) were missing.

For continuous variables, normality was checked before statistical analysis. Urinary 8-OH-dG level (ng/mL) was divided by urinary creatinine level (mg/mL) for correction of its variation in spot samples,[30] and then log-transformed. To compare characteristics of participants by sex, we used t-tests for continuous variables and χ^2-tests for categorical variables. Subjects were divided into quartiles of PA, and linear trends across quartiles were examined based on linear regression analysis for continuous variables and the Mantel test for current smoking, drinking, daily coffee drinking, daily green tea drinking, daily vegetable intake, daily fruit intake, and menopausal status (in women). To examine correlations between log-transformed 8-OH-dG and baseline characteristics, Spearman's rank correlation coefficients were calculated. Adjusted geometric means and their 95% confidence intervals of 8-OH-dG according to quartiles of TPA, MVPA, and VPA were evaluated by least-square general linear regression, and linear trends were assessed based on the statistical significance of the regression coefficient of an ordinal variable for each PA measurement. Adjustments were made for age (continuous), BMI (continuous), smoking status (never, former, or current smoker of 1–19, 20–39, or ≥40 cigarettes/day), alcohol consumption (never, former, or current drinker consuming 0.1–22.9, 23.0–45.9, or ≥46.0 g ethanol/day), energy intake (continuous) in

Table 3. Spearman's rank correlation coefficients between log-transformed urinary 8-hydroxydeoxyguanosine (ng/mg creatinine) and basic characteristics of participants in the second survey of the Japan Multi-institutional Collaborative Cohort Study in the Saga region, according to sex

Characteristics	Men (n = 2370)		Women (n = 4052)	
	r_s	P-value	r_s	P-value
Age, years	0.07	<0.01	0.33	<0.01
BMI, kg/m²	−0.09	<0.01	−0.01	0.51
Smoking status†	0.11	<0.01	0.02	0.18
Alcohol consumption, g ethanol/day	0.01	0.78	−0.05	<0.01
Daily coffee drinking, %	0.00	0.85	−0.10	<0.01
Daily green tea drinking, %	−0.01	0.73	0.07	<0.01
Daily green or yellow vegetable intake, %	0.01	0.51	0.02	0.23
Daily fruit intake, %	0.03	0.10	0.09	<0.01
Total energy intake, kcal/day	−0.02	0.27	0.03	0.10
Vitamin C supplement use, %	0.02	0.26	0.02	0.12
Menopause status (premenopause, postmenopause)			0.41	<0.01
TPA, MET h/day	−0.03	0.11	−0.09	<0.01
PA classified by intensity, MET h/day				
Light (<3 METs)	0.01	0.50	−0.07	<0.01
Moderate (3–6 METs)	−0.04	0.05	−0.05	<0.01
Vigorous (>6 METs)	−0.04	0.04	−0.08	<0.01
Moderate to vigorous (≥3 METs)	−0.05	0.01	−0.08	<0.01

†Smoking status: never, former, or current smoker of 1-19, 20-39, or ≥40 cigarettes/day. BMI, body mass index; MET, metabolic equivalent; PA, physical activity; TPA, total physical activity.

men and women, and daily coffee drinking (yes or no) and menopausal status in women. To clarify the intensity-specific effects of MVPA and VPA, which are independent of TPA, we divided either MVPA or VPA by TPA and made indices of MVPA/TPA and VPA/TPA. Adjusted geometric means and their 95% confidence intervals of 8-OH-dG according to quartiles of MVPA/TPA and VPA/TPA were then calculated with adjustment for possible confounding factors and TPA. We also carried out stratified analyses according to baseline characteristics and examined interactions.

All analyses were undertaken using the SAS statistical software package (version 9.3 for Windows; SAS Institute, Cary, NC, USA). A P-value of <0.05 was considered statistically significant.

Ethical considerations. The study protocol of the overall J-MICC Study was approved by the Ethics Committee at Nagoya University Graduate School of Medicine (Nagoya, Japan);[31] the Saga J-MICC Study was approved by the Ethics Committees of both Saga University Faculty of Medicine (Saga, Japan) and Nagoya University Graduate School of Medicine. Our study was carried out in accordance with the ethical guidelines for epidemiological research set by the Ministry of Education, Culture, Sports, Science and Technology and the Ministry of Health, Labour and Welfare of Japan.

Results

Basic characteristics of study subjects by sex are shown in Table 1. Means of age, BMI, total energy intake, step counts, physical activity levels of TPA, light-intensity PA, moderate-intensity PA, and MVPA were significantly higher in men than in women ($P < 0.01$), as were proportions of current smokers and drinkers. In contrast, proportions of daily coffee drinking, daily green tea drinking, daily green or yellow vegetable intake, daily fruit intake, and vitamin C supplement uses, and geometric mean of urinary 8-OH-dG were significantly higher in women than in men.

Characteristics of study subjects according to quartiles of accelerometer-determined TPA are shown in Table 2. Higher levels of TPA were correlated with younger age, lower BMI, higher proportion of current drinkers, and higher total energy intake in both men and women, lower proportion of current smokers in men, and higher proportion of daily coffee drinkers and lower proportion of postmenopausal status in women. Higher levels of TPA tended to reflect higher levels of PA at all intensities and were correlated with lower levels of urinary 8-OH-dG.

Spearman's rank correlation coefficients between urinary log-transformed urinary 8-OH-dG (ng/mg creatinine) and basic characteristics are presented in Table 3 according to sex. Age had a significant positive correlation with urinary 8-OH-dG levels in both sexes, as did smoking in men and daily green tea intake, daily fruit intake, and postmenopausal status in women. In contrast, a significant inverse correlation with 8-OH-dG was seen for BMI in men, and alcohol drinking and daily coffee drinking in women. Total PA had a significant inverse correlation with 8-OH-dG in women, but no such correlation was noted in men. Furthermore, in women, all intensities of PA, such as light, moderate, vigorous, and moderate to vigorous PA had a significant inverse correlation with 8-OH-dG, whereas in men, all intensities of PA, except light intensity, had a significant inverse correlation with 8-OH-dG.

The adjusted geometric means of urinary 8-OH-dG according to accelerometer-determined PA quartiles are shown in

Table 4. Adjusted geometric means and their 95% confidence intervals of urinary 8-hydroxydeoxyguanosine (ng/mg creatinine) according to accelerometer-determined total physical activity (TPA), moderate to vigorous intensity physical activity (MVPA), vigorous intensity physical activity (VPA), and VPA/TPA quartiles (Q)

	Men (n = 2370)						Women (n = 4052)					
	Q1	Q2	Q3	Q4	β†	P for trend‡	Q1	Q2	Q3	Q4	β†	P for trend‡
TPA§	3.72 (3.58–3.86)	3.75 (3.61–3.89)	3.63 (3.50–3.77)	3.62 (3.49–3.76)	−0.012	0.17	4.18 (4.06–4.31)	4.16 (4.04–4.29)	4.09 (3.98–4.21)	3.95 (3.84–4.07)	−0.019	<0.01
TPA¶	3.71 (3.57–3.85)	3.74 (3.61–3.88)	3.62 (3.49–3.76)	3.65 (3.51–3.79)	−0.008	0.34	4.18 (4.06–4.31)	4.15 (4.03–4.28)	4.10 (3.98–4.22)	3.98 (3.86–4.07)	−0.018	0.01
TPA††	3.66 (3.52–3.81)	3.73 (3.59–3.87)	3.64 (3.51–3.78)	3.69 (3.54–3.84)	0.000	0.98	4.17 (4.05–4.30)	4.16 (4.04–4.28)	4.09 (3.98–4.21)	3.96 (3.84–4.08)	−0.017	0.01
PA classified by different intensity												
MVPA (≥3 METs)§	3.82 (3.68–3.96)	3.69 (3.55–3.82)	3.65 (3.52–3.79)	3.56 (3.43–3.70)	−0.022	0.01	4.21 (4.09–4.34)	4.17 (4.05–4.29)	4.04 (3.92–4.16)	3.97 (3.85–4.08)	−0.021	<0.01
MVPA/TPA‡‡	3.83 (3.68–3.99)	3.69 (3.56–3.83)	3.59 (3.46–3.73)	3.60 (3.46–3.75)	−0.022	0.03	4.11 (3.98–4.24)	4.15 (4.03–4.27)	4.05 (3.93–4.17)	4.08 (3.95–4.21)	−0.005	0.51
VPA (>6 METs)§	3.82 (3.68–3.97)	3.68 (3.54–3.81)	3.62 (3.49–3.76)	3.60 (3.47–3.74)	−0.019	0.03	4.15 (4.02–4.27)	4.07 (3.95–4.19)	4.11 (3.99–4.23)	4.06 (3.95–4.19)	−0.005	0.48
VPA/TPA‡‡	3.74 (3.60–3.88)	3.74 (3.60–3.88)	3.60 (3.47–3.73)	3.64 (3.50–3.78)	−0.012	0.21	4.12 (4.00–4.25)	4.04 (3.92–4.16)	4.10 (3.98–4.22)	4.12 (4.00–4.25)	0.002	0.83

†Regression coefficient. ‡Based on multiple linear regression analysis. §Adjusted for age, body mass index, smoking status, alcohol drinking status, total energy intake in men and women, and coffee drinking status and menopausal status in women. ¶Adjusted for age, body mass index, smoking status, alcohol drinking status, total energy intake, MVPA/TPA in men and women, and coffee drinking status and menopausal status in women. ††Adjusted for age, body mass index, smoking status, alcohol drinking status, total energy intake, VPA/TPA in men and women, and coffee drinking status and menopausal status in women. ‡‡Adjusted for age, body mass index, smoking status, alcohol drinking status, total energy intake, TPA in men and women, and coffee drinking status and menopausal status in women. CI, confidence interval; MET, metabolic equivalent; PA, physical activity.

Table 4. The inverse correlation with TPA in women was significant, even after adjustment for age, BMI, smoking status, alcohol drinking status, total energy intake, daily coffee drinking status, menopausal status, and MVPA/TPA or VPA/TPA. This correlation was stronger in premenopausal women ($\beta = -0.044$, $P = 0.02$) than in postmenopausal women ($\beta = -0.013$, P trend $= 0.06$), although interaction was not significant (P interaction $= 0.07$) (Fig. S1). In women, MVPA had a significant inverse correlation with 8-OH-dG levels, but MVPA/TPA, VPA, and VPA/TPA had no correlation. In contrast, in men, urinary 8-OH-dG was inversely correlated with MVPA, MVPA/TPA, and VPA, although the correlation with TPA was not significant.

Stratified analysis by smoking status showed that urinary 8-OH-dG levels were higher in current smokers than in former and non-smokers at all MVPA levels other than the highest MVPA quartile (Fig. 1). The inverse correlation between MVPA level and urinary 8-OH-dG was clear in current smokers ($\beta = -0.036$, P trend $= 0.04$), although interaction between smoking status and MVPA levels on urinary 8-OH-dG was non-significant (P interaction $= 0.44$).

Discussion

In our population-based study, we observed an inverse correlation between objective PA and urinary levels of 8-OH-dG, which is a marker of cellular oxidative stress. Our results support the hypothesis that habitual PA may reduce oxidative stress. Perhaps of greatest note of our finding was that greater TPA in women and greater MVPA in men were independently correlated with reduced levels of urinary 8-OH-dG. To our knowledge, no previous report has investigated and objectively measured the intensity-specific effects of PA on oxidative stress, and only one study reported a significant negative correlation between MVPA and urinary levels of 8-OH-dG in men, with PA assessed using a self-reported questionnaire.[19] We hope that our findings will support the notion that increasing PA can contribute to reduced oxidative stress.

Intense, exhaustive exercise has been reported to increase oxidative stress and subsequent DNA damage through increased levels of ROS.[14,17,18] However, when undertaken at a moderate intensity, exercise can induce production of antioxidant enzymes such as superoxide dismutase, glutathione peroxidase, catalase, and glutathione reductase.[14–16] In addition, PA might increase the activity of human 8-oxoguanine DNA glycosylase, which plays a major role in the base repair pathway by removing 8-oxoguanine base lesions generated by ROS.[14,32] Radak *et al.*[14] suggested that regular exercise-induced handling of ROS through redox signaling, including the above-mentioned effects on antioxidants and oxidative damage repair systems, significantly contributes to the health-promoting effects of regular exercise. Of note, the effects of PA resembled the characteristics of hormesis, as exercise must attain a certain level of intensity to exert its beneficial effects.[8,14–16]

Greater TPA in women and greater MVPA in men were correlated with reduced levels of urinary 8-OH-dG in the present study. We suspect that the gender difference in fitness level might have influenced this gender-specific relationship. Although low-level exercise loading may be sufficient to induce adaptation to oxidative stress, including production of antioxidant enzymes and increased human 8-oxoguanine DNA glycosylase activity among individuals with low physical fitness, well-trained individuals require higher levels of exercise
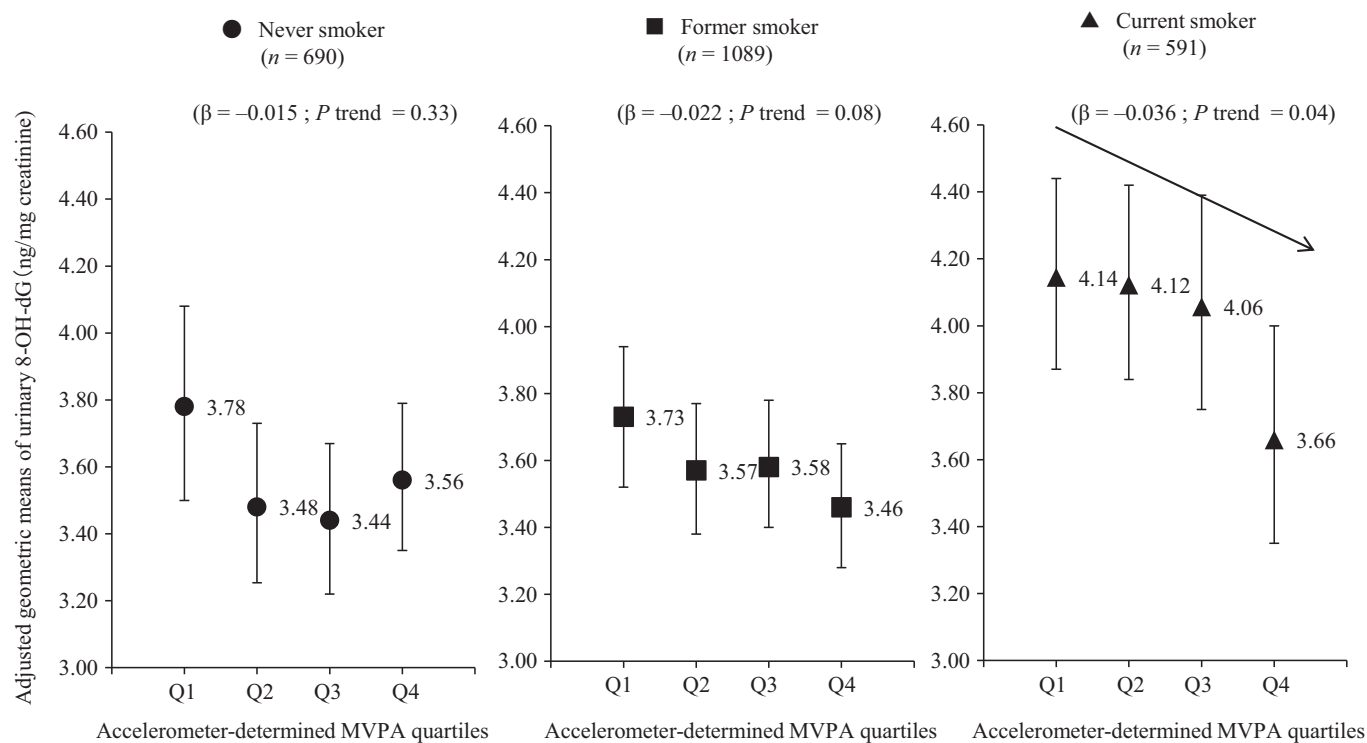
Fig. 1. Age, body mass index, smoking status, alcohol drinking status, and total energy intake adjusted geometric means of urinary 8-hydroxy-deoxyguanosine (8-OH-dG) (ng/mg creatinine) according to accelerometer-determined moderate to vigorous intensity physical activity (MVPA; ≥3 metabolic equivalents) quartiles (Q) by smoking status in men.

intensity to achieve the same results. Because women tend to be less physically fit than men,[33,34] generation of antioxidant enzymes against oxidative stress might be induced by lower intensity of PA in women than in men. In contrast, men might need MVPA for such enzyme induction to occur, due to their tendency to have a higher fitness level than women.

Similar to previous studies,[35–37] older age, lower BMI, and being a smoker had positive correlations with urinary levels of 8-OH-dG, whereas daily coffee drinking had a negative correlation in the present study. In addition, age, BMI, smoking status, alcohol drinking status, and coffee drinking status in women were correlated with PA and were therefore considered confounding factors influencing the correlation between PA and urinary levels of 8-OH-dG. Among these factors, smoking status had a particularly large influence on the above correlation. Although urinary levels of 8-OH-dG in current smokers were higher than in former and non-smokers, they decreased with increasing MVPA to a greater degree in current smokers than in former and non-smokers. This result concurs with an epidemiological finding that PA reduces cancer risk and mortality among those with marked tobacco exposure.[38] Our results therefore suggest that men who have a smoking habit and are inactive might experience greater oxidative damage to DNA and be at a higher risk of developing cancer than those with more active lifestyles who do not smoke.

This study has several advantages over previous studies with regard to evaluating PA and oxidative stress. First, most epidemiological studies have assessed PA by questionnaires using various types of activity categories, such as leisure-time activity, exercise, sports, and occupational activities, and neglected to collect details on intensity and duration of PA. Perhaps for this reason, previous cross-sectional studies have reported inconsistent results regarding associations between habitual exercise and oxidative stress.[35,36] Second, the sample size in the present study was relatively large, facilitating evaluation of the effect of intensity-specific PA on 8-OH-dG levels. Sample sizes of previous studies[35–37] evaluating 8-OH-dG levels have been <900, approximately one-tenth of our sample size. Finally, the accuracy of the HPLC method used to assess urinary 8-OH-dG levels in our study was higher than that of an enzyme-linked immunosorbent assay, which may cross-react with other structure-related compounds.[35]

However, several limitations to the present study also warrant mention. First, the cross-sectional nature of our study limits our ability to determine causation, even though we excluded participants who had a history of diseases potentially associated with changes in PA, including cancer. Second, our method of measuring PA was unable to capture cycling, swimming, and load-bearing activities, potentially causing non-differential misclassification of PA which might reduce estimations for the association. In addition, the accelerometer was unable to distinguish intensity of ≥8.3 METs. We were therefore unable to examine the correlation between an exhausting level of PA and 8-OH-dG concentration. Third, physical activity was measured only for 10 days. Physical activity varies according to seasons,[39] which might be a source of measurement error. Although the present study found lower levels of physical activity during winter (Table S1), further adjustment for season did not change the results. Finally, antioxidant enzyme activity and endogenous antioxidant levels as well as fitness level, which might influence the correlation, were not measured. Confirming our hypothesis on gender difference in adaptation for oxidative stress will require further studies.

In conclusion, high TPA in women and high MVPA in men were related to reduced levels of 8-OH-dG, suggesting a sex-specific effect of PA for protection from oxidative DNA damage. This finding supports the notion that increasing habitual physical activity in daily life is linked to reduction of oxidative stress.

Abbreviations

BMI	body mass index
BSA	body surface area
J-MICC	Japan Multi-institutional Collaborative Cohort
MET	metabolic equivalents
8-OH-dG	8-hydroxydeoxyguanosine
MVPA	moderate/vigorous intensity physical activity
PA	physical activity
ROS	reactive oxygen species
TPA	total physical activity
VPA	vigorous intensity physical activity

Disclosure Statement

The authors have no conflict of interest.

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Supporting Information

Additional Supporting Information may be found online in the supporting information tab for this article:

Fig. S1. Age, body mass index, smoking status, alcohol drinking status, total energy intake, daily coffee drinking status, and menopausal status adjusted geometric means of urinary 8-hydroxydeoxyguanosine (8-OH-dG) (ng/mg creatinine) according to accelerometer-determined total physical activity (TPA) quartiles by menopausal status in women.

Table S1. Physical activity and urinary 8-hydroxydeoxyguanosine (8-OhdG) (ng/mg creatinine) levels according to season.