

Sex difference in fluid balance responses during prolonged exercise

T. M. H. Eijsvogels¹, R. R. Scholten^{1,2}, N. T. L. van Duijnhoven¹, D. H. J. Thijssen^{1,3}, M. T. E. Hopman¹

¹Department of Physiology, Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands, ²Department of Obstetrics and Gynecology, Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands, ³Research Institute for Sport and Exercise Sciences, Liverpool John Moores University, Liverpool, UK

Corresponding author: Dr M. T. E. Hopman, Department of Physiology (143), Radboud University Nijmegen Medical Centre, PO Box 9101, 6500 HB Nijmegen, The Netherlands. Tel: (+31) (0)24 36 13650, Fax: (+31) (0)24 354 0535, E-mail: m.hopman@fysiol.umcn.nl

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Maintaining a proper fluid balance is important during exercise as athletes are prone to develop dehydration during exercise. Although several factors may regulate the fluid balance, little is known about the role of sex during prolonged moderate-intensity exercise. Therefore, we compared body mass changes and fluid balance parameters in men vs women in a large heterogeneous group of participants during prolonged exercise. Ninety-eight volunteers walked 30–50 km at a self-selected pace. Exercise duration (8 h, 32 min) and intensity (69% HR_{max}) were comparable between groups. Men demonstrated a significantly larger change in body mass than women (−1.6% vs −0.9%, respectively, $P < 0.001$) and a

higher incidence of dehydration (defined as $\geq 2\%$ body mass loss) compared with women (34% vs 12%, respectively, odds ratio = 4.2, 95% CI = 1.1–16.7). Changes in blood sodium levels were significantly different between men (+1.5 mmol/L) and women (−0.4 mmol/L), while 27% of the men vs 0% of the women showed postexercise hyponatremia (sodium levels ≥ 145 mmol/L). Moreover, men demonstrated a significantly lower fluid intake (2.9 mL/kg/h) and higher fluid loss (5.0 mL/kg/h) compared with women (3.7 and 4.8 mL/kg/h, respectively). Taken together, our data suggest that men and women demonstrate different changes in fluid balance in response to a similar bout of exercise.

Imbalance between fluid intake and fluid loss during prolonged exercise may increase the risk for development of dehydration. Previous laboratory studies showed that substantial fluid/body mass losses were related to an impaired exercise performance (Sawka, 1992; Barr, 1999) and to a larger increase in core body temperature (Montain & Coyle, 1992; Sawka, 1992). Accordingly, dehydration may enhance the development of hyperthermia, heat exhaustion, and heat stroke (Carter et al., 2005; Armstrong et al., 2007). Warm and humid conditions, high-intensity exercise, and/or prolonged exercise increase the risk of a mismatch between fluid intake and fluid loss and, thereby, the potential development of dehydration (Rehrer, 2001; Chevront et al., 2003).

Sex may impact fluid loss and fluid intake. Indeed, men have higher sweat rates that might lead to more fluid loss during exercise as compared with women (Sawka et al., 1983; Bar-Or, 1998). Previous studies also showed that men have higher plasma sodium levels and a higher prevalence of hyponatremia than women after prolonged exercise (Chorley et al., 2007; Eijsvogels et al., 2008), which suggest a larger fluid loss in men. In contrast, it is also reported that women have an increased

risk for overdrinking, which could lead to exercise-associated hyponatremia (Hew-Butler et al., 2008a). While these observations suggest that men are more prone to dehydration than women, little is known about a potential sex difference during prolonged, moderate-intensity exercise.

The purpose of this study, therefore, was to examine markers of dehydration during prolonged moderate-intensity exercise in men vs women in a large heterogeneous group of participants. Markers of dehydration include body mass changes, a $\geq 2\%$ decrease in body mass after finishing (Chevront et al., 2003; Sawka et al., 2007), sodium levels, and plasma volume changes after exercise. We hypothesize that men have a higher incidence of dehydration and will develop larger changes in markers of (de)hydration (e.g. sodium levels and plasma volume changes) than women. Furthermore, we aimed to identify factors (subject and race characteristics) that may contribute to the potential sex differences.

Methods

Subjects

Ninety-nine participants (21–82 years, 57 men and 42 women) volunteered to participate in this study (Table 1). Subjects with

Conflicts: None.

Table 1. Demographic characteristics and health status for men ($n = 56$) and women ($n = 42$)

	Men	Women	<i>P</i> value
Demographic characteristics			
Age (year)	61 ± 14	56 ± 15	0.92
Height (cm)	180 ± 7	165 ± 7	<0.001
Weight (kg)	83.9 ± 13.5	65.5 ± 10.3	<0.001
Body mass index (kg/m ²)	26.0 ± 3.3	24.0 ± 3.2	0.004
Body fat (%)	25 ± 6	35 ± 5	<0.001
Waist circumference (cm)	96 ± 10	82 ± 9	<0.001
Health status			
Physical activity (h/week)	3.1 ± 5.1	3.3 ± 3.4	0.86
≥5 times/week ≥30 min exercise (%)	79%	85%	0.48
Mean arterial pressure (mm Hg)	104 ± 11	104 ± 11	0.13
Use of prescribed medicines			
Diuretics	23 (41%)	11 (26%)	0.13
Antihypertensive drugs	6 (11%)	2 (5%)	0.29
Statins	16 (29%)	3 (7%)	0.008
Statin	11 (20%)	3 (7%)	0.08
Other (e.g. painkillers, asthma, antirheumatics)	3 (5%)	5 (12%)	0.24
Pathology			
Hypertension	35 (63%)	23 (55%)	0.44
Cardiovascular diseases	14 (25%)	7 (17%)	0.32
Hypercholesterolemia	16 (29%)	7 (17%)	0.17
Hypercholesterolemia	16 (23%)	5 (12%)	0.15
Other (e.g. osteoporosis, skin disease, asthma)	21 (38%)	17 (41%)	0.77

P value refers to an unpaired *t*-test or chi-square test between men and women.

chronic (inflammatory) bowel problems were excluded from participation as a contraindication for the use of the telemetric temperature sensor. The Medical Ethical Committee of the Radboud University Nijmegen Medical Centre approved the study and all subjects gave their written informed consent prior to participation. This study was conducted in line with the Declaration of Helsinki.

Experimental design

Baseline measurements were conducted under controlled conditions. Immediately before the race, we measured body mass, heart rate, and core body temperature. Subsequently, subjects walked 30 km (men: 37%/women: 31%), 40 km (men: 34%/women: 40%), or 50 km (men: 29%/women: 29%) at a self-selected pace. During exercise, we assessed heart rate and core body temperature every 5 km, and all subjects registered their fluid intake using a diary. Immediately after finishing, all baseline measurements were repeated.

Measurements

Subject characteristics

At baseline, body mass (Seca 888 scale, Hamburg, Germany) and height were measured. Subsequently, a four-point skinfold thickness measurement (biceps, triceps, sub-scapular, supra-iliac) was obtained in order to calculate the lean body mass (Durnin & Womersley, 1974). Waist circumference was measured midway between the lower rib margin and iliac crest. Thereafter, resting heart rate and blood pressure were measured twice using an automated sphygmomanometer (M5-1 intellisense, Omron Healthcare, Hoofddorp, the Netherlands) after 5-min seated rest. Finally, all subjects completed a questionnaire about their physical activity and health status. Women were defined as postmenopausal when the last menstruation in absence of hormonal treatment occurred at least 1 year before the start of this experiment.

Fluid balance

Before the start of exercise, and directly after finishing, body mass was measured. The relative change in body mass (in %) between

both measurements was calculated. Dehydration was defined as a body mass loss of 2% or more, which is a frequently applied definition for dehydration in field studies (Sawka et al., 2007; Sawka & Noakes, 2007). Furthermore, all subjects received written and individual oral instructions concerning the registration of their fluid intake. Subjects were allowed to drink *ad libitum*, while they registered the time and amount (standard-sized cups, bottles, etc.) of their individual fluid intake from 12 h preceding the start until the end of the experiment.

Blood analysis

After 5 min of rest in an upright position, 2 mL of venous blood was drawn at baseline and directly after finishing in order to determine plasma levels of sodium, hematocrit (Rapidpoint® 400, Siemens Healthcare Diagnostics Inc., Tarrytown, New York, USA) and hemoglobin (HemoCue AB, Ängelholm, Sweden). Hyponatremia and hypernatremia were defined as a plasma sodium concentration of ≤135 mmol/L and ≥145 mmol/L, respectively (Adroge & Madias, 2000; Hew-Butler et al., 2008a). Relative changes in plasma volume (%) were calculated from changes in blood hematocrit and hemoglobin concentrations according Dill and Costill's equation (Dill & Costill 1974): $\Delta PV (\%) = 100 \times [(Hb_{\text{baseline}}/Hb_{\text{post-exercise}}) \times (1 - Hct_{\text{post-exercise}} \times 0.01)] / (1 - Hct_{\text{baseline}} \times 0.01) - 100$.

Urine specific gravity

Subjects were asked to provide a 5-mL urine sample at baseline as well as directly after finishing. The samples were immediately analyzed to determine the urine specific gravity (Clinitek Status® Analyzer; Siemens Healthcare Diagnostics).

Core body temperature

Core body temperature was determined using a portable telemetry system (CorTemp™ system, HQ Inc., Palmetto, Florida, USA), which has been demonstrated to be safe and reliable (Gant et al., 2006; Byrne & Lim, 2007). Participants ingested an individually

calibrated telemetric temperature sensor the evening preceding the experiment, in order to avoid any interaction with fluid ingestion (Wilkinson et al., 2008). Prior to the start, core body temperature of each individual participant was measured using an external recorder. Baseline core body temperature was determined as the average of three consecutive measurements. Subsequently, core body temperature was similarly determined at every 5 km during the experiment. The highest value of these measurements was presented as maximum core body temperature.

Exercise intensity

Heart rate was measured simultaneously with core body temperature (i.e. every 5 km point, three consecutive measurements), using a 2-channel ECG chest band system (Polar Electro Oy, Kempele, Finland). Mean heart rate during exercise was calculated as the average heart rate, excluding the values derived directly before the start and after the finish. Exercise intensity was calculated by dividing the mean heart rate during exercise by the maximal predicted heart rate ($208 - 0.7 \times \text{age}$) (Tanaka et al., 2001).

Ambient conditions

Throughout the experiment, dry bulb, wet bulb, and globe temperatures were measured every 30 min using a portable climate monitoring device (Davis Instruments Inc., Hayward, California, USA) positioned at the start/finish area. The wet bulb globe temperature index (WBGT) was calculated using the formula: $\text{WBGT} = 0.1 (T_{\text{dry bulb}}) + 0.7 (T_{\text{wet bulb}}) + 0.2 (T_{\text{globe}})$ (Armstrong et al., 2007).

Statistics

All values are presented as means with standard deviation, unless indicated otherwise. Statistical analyses were performed using Statistical Package for Social Sciences 16.0 (SPSS, Chicago, Illinois, USA). The level of statistical significance was set at $P < 0.05$. The normality of the data distribution was examined by the Kolmogorov–Smirnov test. To compare differences between men and women or pre- and postmenopausal women, an unpaired *t*-test was used for continuous variables. To assess differences in the incidence of hypernatremia and high urine specific gravities between the groups, a Pearson's chi-square test was applied. A multivariable logistic regression analyses was used to model the relation between sex and the incidence of dehydration as indicated by $\geq 2\%$ body mass loss. Crude odds ratios (crORs) were obtained by entering the incidence of $\geq 2\%$ body mass loss as dependent variable and sex (categorized) as independent variable. Differences in anthropometrical data, health status, exercise characteristics and hydration status, between men and women, were tested for their potential confounding effects on the incidence of dehydration as indicated by $\geq 2\%$ body mass loss. To evaluate the confounding effect size of these covariates, we added one factor at a time to our regression model based on sex. When the odds ratio (OR) changed with more than 10% compared with the crOR after introducing a potential confounder, the confounding effect was considered large enough and therefore we retained this factor in our final model for the estimation of the incidence of dehydration based on sex (adjusted OR). All ORs are presented with their 95% confidence intervals (CIs). To identify factors that contribute to the development of $\geq 2\%$ body mass loss, a backward stepwise linear regression analysis was used. We have included sex, age, body mass index (BMI), exercise duration, exercise intensity, and fluid intake as potential determinants of the presence of dehydration. To exclude the possibility that baseline differences in body mass between men and women may impact the presence of $\geq 2\%$ body

mass loss, we additionally performed a Pearson correlation and a backward logistic regression analysis to study the contribution of body mass to our outcomes. A two-way repeated measures analysis of variance was used to test the changes in plasma sodium concentration over time between men and women.

Results

One male subject withdrew from participation after baseline measurements and was therefore excluded from analysis. We found no sex difference regarding age, physical activity level, mean arterial pressure, and the presence of hypertension, cardiovascular diseases, and hypercholesterolemia (Table 1). Apart from antihypertensive drugs, no difference in the use of medication between both groups was present (Table 1). Men showed a higher BMI, height, weight, and waist circumference, but lower fat percentage compared with women (Table 1). To correct for the anthropometric differences between men and women, BMI and the use of antihypertensive drugs were included in the multivariate logistic regression analysis.

Exercise characteristics

Relative humidity, dry bulb, and wet bulb temperature were 67%, 14.2°C, and 12.2°C, respectively, in the early morning and 78%, 20.5°C, and 16.7°C, respectively, in the afternoon. WBGT during the experiment varied between 13.2°C and 20.6°C. Duration of the exercise bout was comparable between both groups (Table 2). Subjects performed exercise at $69 \pm 11\%$ of their predicted maximal heart rate, which was similar between men and women (Table 2). While baseline and maximal core body temperature were significantly higher in women than in men, the increase in core body temperature was comparable between both groups (Table 2).

Body mass losses

A significant decrease in body mass was observed in both groups after finishing, which was significantly larger in men than in women (Fig. 1). While 25% of our population demonstrated $\geq 2\%$ body mass loss, men more often exceeded this threshold than women (34% vs 12%, respectively, crOR = 3.8, CI = 1.3–11.3, Fig. 2). After correction for BMI, antihypertensive drugs, speed, maximum core body temperature and fluid intake, the relation between sex and $\geq 2\%$ body mass loss remained significant (adjusted OR = 4.2, CI = 1.1–16.7). Indeed, a backward logistic regression analysis revealed that only sex (OR = 2.7, CI = 0.9–8.5) and fluid intake (OR = 0.6, CI = 0.4–0.9) significantly contributed to the presence of $\geq 2\%$ body mass loss. In addition, we found no correlation between baseline body mass and body mass changes after prolonged exercise within men ($P = 0.79$, $r = -0.04$) and women ($P = 0.86$, $r = 0.03$). This suggests

Table 2. Exercise characteristics and hydration status for men ($n = 56$) and women ($n = 42$)

	Men	Women	<i>P</i> value
Exercise characteristics			
Exercise duration (hh : mm)	8:15 ± 1:39	8:54 ± 1:38	0.054
Speed (km/h)	4.8 ± 0.6	4.5 ± 0.7	0.043
Baseline core body temperature (°C)	37.2 ± 0.4	37.4 ± 0.4	0.025
Maximum core body temperature (°C)	38.1 ± 0.3	38.3 ± 0.4	0.018
Increase in core body temperature (°C)	1.0 ± 0.4	0.9 ± 0.4	0.59
Exercise intensity (%)	68 ± 11	70 ± 10	0.28
Hydration status			
Fluid intake (liter)	2.1 ± 1.2	2.2 ± 0.8	0.80
Water (%)	57%	52%	
Sports drink (%)	11%	16%	
Other (%)	32%	32%	
Fluid intake/h/kg (mL)	2.9 ± 1.1	3.7 ± 1.3	0.002
Body mass change (kg)	-1.4 ± 0.9	-0.6 ± 0.5	<0.001
Body mass change (%)	-1.6 ± 1.0	-0.9 ± 0.8	<0.001
Dehydration; >2% body mass loss (%)	19 (34%)	5 (12%)	0.01
Calculated plasma volume change (%)	-1.5 ± 9.0	1.4 ± 7.4	0.09
Baseline urine specific gravity ≥1.030 g/mL	15 (27%)	14 (33%)	0.48
Finish urine specific gravity ≥1.030 g/mL	25 (45%)	11 (26%)	0.063
Baseline sodium concentration (mmol/liter)	141.7 ± 2.0	141.3 ± 1.4	0.24
Finish sodium concentration (mmol/liter)	143.2 ± 2.6	140.9 ± 1.9	<0.001
Baseline hypernatremia (%)	2 (4%)	0 (0%)	0.22
Finish hypernatremia (%)	15 (27%)	0 (0%)	<0.001

P value refers to an unpaired *t*-test or chi-square test between men and women.

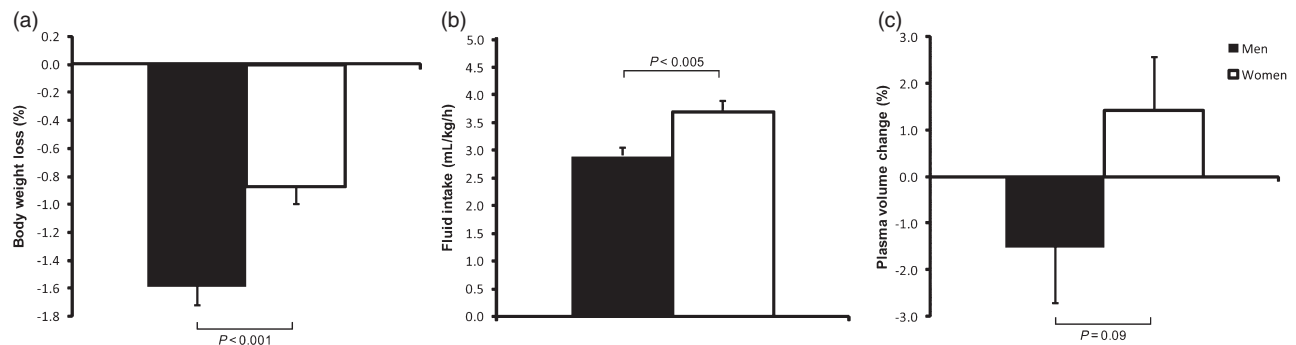


Fig. 1. Fluid balance parameters in men ($n = 56$) and women ($n = 42$). Men demonstrated a significantly larger body weight loss (a) and higher fluid intake (b) compared with women, while also a trend in plasma volume changes (c) among both groups was observed. Data are presented as mean ± SE.

that differences in body mass between men and women do not explain the sex differences in the presence of ≥2% body mass loss.

Fluid balance

The average fluid intake preceding the start of the experiment was 1.2 ± 0.6 liter in men and 1.0 ± 0.6 liter in women and did not statistically differ between the groups ($P = 0.21$). Also, the total amount and type of fluid intake during exercise was not different between groups (Table 2). However, when corrected for body mass and exercise duration, men demonstrated a significantly lower fluid intake during exercise than women (Fig. 1). In addition, men showed a decrease in postexercise plasma volume (-1.5%), while women demonstrated an increase

(1.4%, Fig. 1). Moreover, men demonstrated a higher incidence of postexercise urine specific gravity levels ≥1.030 g/mL compared with women (Table 2). Comparison between premenopausal and postmenopausal women revealed no differences for fluid intake, body mass changes, or urine specific gravity levels (Table 3).

Sodium levels

While men demonstrated an increase in plasma sodium levels, a decrease was observed in women (Fig. 3), leading to higher postexercise blood sodium levels in men (Table 2). In addition, the incidence of hypernatremia (sodium level ≥145 mmol/L) was significantly higher in men compared with women (27% vs 0%, respectively, $P < 0.001$, Fig. 4). The exercise-induced change in

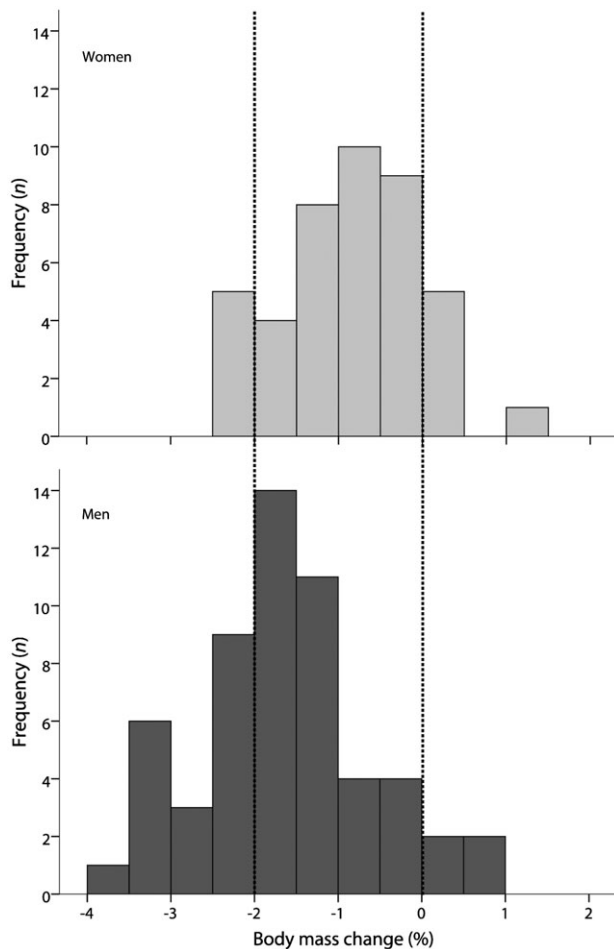


Fig. 2. Frequency distribution of body mass change between start and finish in men ($n = 56$) and women ($n = 42$). The area between the two dashed lines represents euhydration, while a body mass change of $\geq 0\%$ or $\leq -2\%$ represents overhydration and dehydration, respectively. The incidence of dehydration was significantly higher ($P < 0.01$, OR = 4.2, CI = 1.1–16.7) in men (34%) compared with women (12%).

plasma sodium levels was not different between premenopausal (-0.9 ± 1.9 mmol/L) and postmenopausal women (-0.3 ± 2.0 mmol/L, *t*-test; $P = 0.41$).

Discussion

This study examined the impact of sex on fluid balance responses during prolonged exercise in a large group of men and women. A clear difference between the sexes was observed, with a significantly lower fluid intake and larger loss of body mass in men. Moreover, a sex difference is further supported by the larger increase in plasma sodium levels and higher incidence of hypernatremia in men. Although none of our subjects reported clinical signs or health problems, the findings of this study indicate that important differences are present in the hydration status between men and women during prolonged exercise.

Given the prolonged duration (8 h, 30 min) and intensity (69% of their maximum predicted heart rate) of the exercise bout, our subjects are susceptible to disturbances in their fluid balance (Cheuvront et al., 2003; Sawka, Burke 2007). While the relation between fluid disturbances and the duration or intensity of exercise is widely acknowledged (Murray, 1996; Casa et al., 2000; Rehrer, 2001; Cheuvront et al., 2003), we focused on dehydration after prolonged moderate-intensity exercise. In our study, the average body mass loss was 1.6% in men and 0.9% in women. The magnitude of this response is in line with a previous study that used models to predict body mass changes after prolonged running (Montain et al., 2006). Moreover, we found that 25% of the participants lost more than 2% of their body mass, which is frequently used as a marker for dehydration. It is important to notice that this high incidence was observed during mild to moderate ambient conditions, and ergo is likely to increase when the same type of exercise is performed under more strenuous ambient conditions.

The principal finding of our study is that men demonstrated a larger body mass loss compared with women during prolonged exercise. Decreases in body mass after exercise relates, at least partly, to water loss (Baker et al., 2009). Therefore, men might be more prone to develop dehydration during exercise than women. This observation raises the question on what the potential mechanisms might be that contributes to this sex difference. Although longer duration and high-intensity exercise predisposes subjects to develop dehydration (Rolls & Phillips, 1990; McConell et al., 1997; Cheuvront et al., 2003), men and women performed exercise at a similar relative intensity and exercise duration in our study. Another potential explanation relates to the amount and type of fluid intake during exercise. Although there was no difference in the type of fluid intake (e.g. water, sports drink, or other), men demonstrated a 22% lower rate of fluid intake than women (2.9 mL/h/kg vs 3.7 mL/h/kg). This observation is in parallel with a previous laboratory study (Baker et al., 2005). Alternatively, sex differences may influence the thirst stimulus in the hypothalamus. Because advanced age attenuates the thirst stimulus (Rolls & Phillips, 1990; Mack et al., 1994), we matched age between the various groups in our study. Moreover, increased core body temperature influences the perception of thirst (Takamata et al., 1995). Interestingly, women demonstrated a slightly, but significantly higher maximum core body temperature during exercise than men (38.3°C vs 38.1°C), which may partially contribute to their higher fluid intake. Nonetheless, even after correction for fluid intake in our statistical model, men demonstrated a higher risk for dehydration compared with women. This makes it unlikely that fluid intake alone explains the sex difference in the occurrence of dehydration.

Combining fluid intake with the absolute body mass change, we were able to roughly estimate the fluid loss (Casa et al., 2000). Men demonstrated a significantly

Table 3. Hydration status in premenopausal ($n = 12$) and postmenopausal ($n = 30$) women during prolonged walking

	Premenopausal	Postmenopausal	<i>P</i> value
Hydration status			
Fluid intake (liter)	2.2 ± 0.9	2.2 ± 0.8	0.79
Fluid intake/h/kg (mL)	3.4 ± 1.5	3.8 ± 1.2	0.42
Body mass change (kg)	-0.4 ± 0.6	-0.7 ± 0.5	0.25
Body mass change (%)	-0.6 ± 0.8	-1.0 ± 0.8	0.18
Dehydration (%)	0 (0%)	5 (17%)	- ^a
Calculated plasma volume change (%)	-0.2 ± 6.3	2.0 ± 7.9	0.39
Urine specific gravity ≥1.030 g/mL	1 (8%)	10 (33%)	0.096
Baseline sodium concentration (mmol/liter)	140.8 ± 1.0	141.5 ± 1.5	0.13
Finish sodium concentration (mmol/liter)	139.9 ± 1.8	141.2 ± 1.8	0.036
Hypernatremia (%)	0 (0%)	0 (0%)	- ^a

^aThe *P* value could not be calculated as (one of) the groups had an incidence of 0%.

P value refers to an unpaired *t*-test or chi-square test between premenopausal and postmenopausal women.

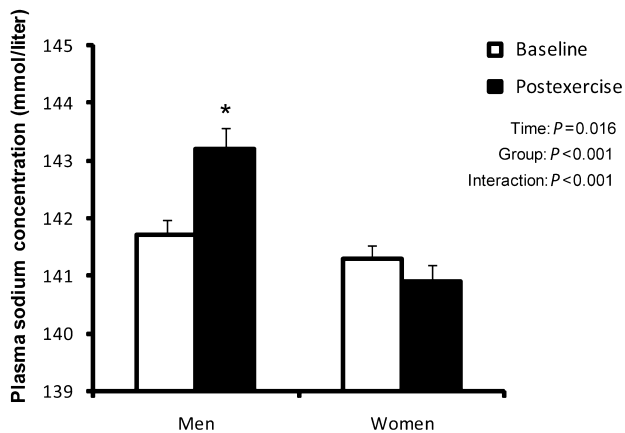


Fig. 3. Plasma sodium concentration in men ($n = 56$) and women ($n = 42$) at baseline and directly postexercise. Two-way repeated measures analysis of variance revealed a significant time, group, and interaction effect. Data are presented as mean ± SE.

greater rate of fluid loss than women during exercise (424 ± 162 mL/h vs 311 ± 93 mL/h, $P < 0.001$). Although this approach does not correct for respiratory and gastrointestinal fluid losses, previous studies showed that metabolic water production in the muscles compensates the respiratory fluid losses (Sawka & Young, 2005; Chevront et al., 2007), while gastrointestinal losses are normally negligible (i.e. 100 mL/day) (Kavouras, 2002). Therefore, fluid loss during exercise is assumed to be predominantly attributed to sweating (Chorley et al., 2007; Sawka, Burke 2007), and subordinately to urinary excretion. The underlying mechanism for the larger sweat loss in men compared with women in our study may relate to differences in sweat gland threshold and/or sweat rate. The threshold for sweating relates to an absolute core body temperature (Nadel et al., 1971). Several studies found that men start sweating at a lower core body temperature threshold than women (Fox et al., 1969; Cunningham et al., 1978;

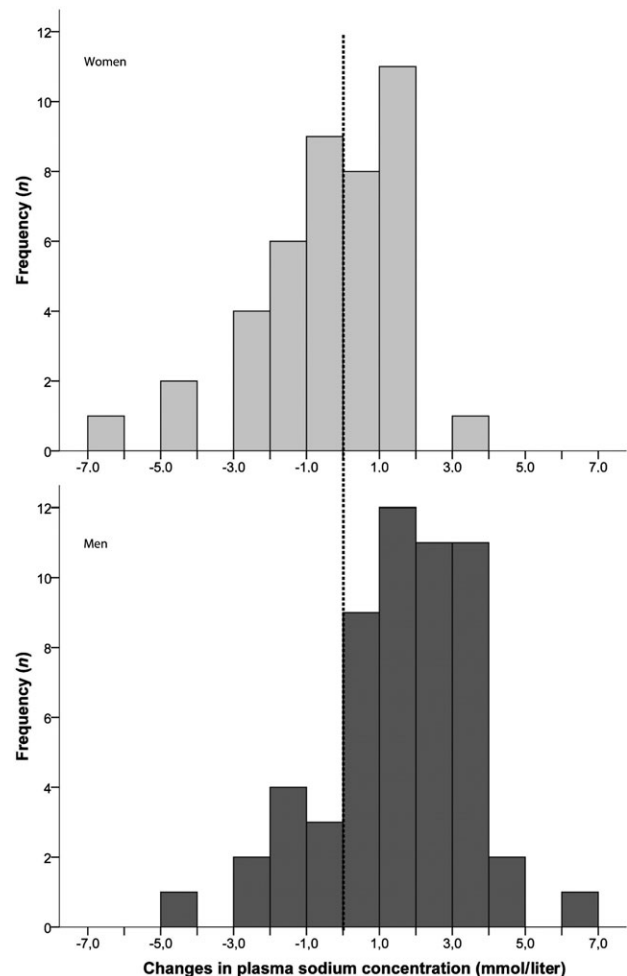


Fig. 4. Frequency distribution of the changes in plasma sodium concentration in men ($n = 56$) and women ($n = 42$) after prolonged moderate-intensity exercise. The dashed line represents no change in plasma sodium concentration, meaning that baseline and postexercise plasma sodium levels were similar. This figure also illustrates the sex difference: that is, men demonstrate an increase in plasma sodium levels, while women show a slight decrease ($P < 0.001$).

Frye & Kamon, 1981; Lopez et al., 1994). However, it was suggested that morphometric differences, rather than sex itself, relate to the difference in sweating between men and women (Havenith & van Middendorp 1990). Indeed, another study reported no sex difference in the sweat gland threshold, despite a higher sweat rate in men (Anderson et al., 1995). Therefore, also a greater sweating rate may contribute to the larger fluid loss in men (Sawka et al., 1983; Bar-Or, 1998). A recent study provided further insight into the underlying mechanisms and found an elevated cholinergic sensitivity of the sweat glands in male subjects, a finding which was independent of physical fitness level (Madeira et al., 2010). This higher sensitivity may relate to a higher sweat rate in men compared with women. Collectively, (sex-related) differences in sweat gland threshold and/or sweat gland sensitivity are likely to contribute to the higher sweat loss during exercise in men compared with women.

The antidiuretic hormone arginine vasopressin (AVP) regulates fluid balance by increasing renal water reabsorption and thereby reducing urinary output (and thus preventing dehydration). Sex hormones modulate the synthesis and osmotic regulation of AVP (Sar & Stumpf, 1977, 1980; Stachenfeld et al., 1998). At rest, men have a higher baseline plasma AVP level and a greater sensitivity to changes in plasma osmolality than women (Claybaugh et al., 2000; Stachenfeld et al., 2001). Nonetheless, no differences in urinary output between men and women were found after osmotic stimulation (Stachenfeld et al., 2001). Also, exercise represents a stimulus that induces a change in AVP levels to maintain fluid balance (Hew-Butler et al., 2008b). Possibly, an attenuated exercise-induced increase in AVP in men may contribute to their larger fluid losses.

The differences between men and women may also relate to the menstrual cycle, especially because the menstrual cycle alters body fluid regulation (Stachenfeld, 2008). In our study, ~70% of the female population was postmenopausal. Nonetheless, fluid intake and changes in body mass, plasma volume, and plasma sodium were not different between pre- and postmenopausal women. Therefore, the sex-related differences in our study unlikely relate to the menstrual cycle.

Limitations

Dehydration in our study was assessed by body mass changes before and after exercise. Although this

approach is a frequently and popular method, especially during field studies (Sawka & Montain, 2000; Chevront et al., 2003; Maughan, 2003; Almond et al., 2005), some limitations must be considered. A recent study showed that using body mass changes in men and women is a reliable and accurate method to assess total body water changes after prolonged running (Baker et al., 2009). Nonetheless, this measure does not account for substrate oxidation and metabolic water production during prolonged exercise (Maughan et al., 2007; King et al., 2008). Consequently, body mass loss during exercise is not solely because of fluid loss, as also discussed in response to the findings of Baker et al. (Baker, 2010; Nolte & Noakes, 2010). Therefore, one may question the validity of the subjective 2% cut-off value for the definition of dehydration. Nonetheless, we believe that our methods are valid to examine the primary aim of the study, especially because we also included other markers of hydration (i.e. sodium concentration and plasma volume changes) that reinforce the sex differences in hydration status.

Perspectives

We found that men demonstrated larger decreases in body mass and a higher incidence of dehydration after prolonged exercise than women. This difference was reinforced by backward linear regression analysis that revealed that sex and fluid intake were the only parameters that relate to $\geq 2\%$ body mass loss during prolonged walking. These findings suggest that the control of the fluid balance is regulated differently in men and women during exercise. Although sex differences in fluid intake and sweat rate can partially explain our results, these parameters do not fully account for the observed sex differences. Our findings suggest that sex must be considered when providing fluid replacement advices and/or guidelines.

Key words: Fluid balance, hyponatremia, walking exercise, electrolyte balance, endurance exercise.

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