Eur J Appl Physiol (1996) 72: 468–477

ORIGINAL ARTICLE

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Fluid balance and renal response following dehydrating exercise in well-trained men and women

Accepted: 1 August 1995

Abstract We examined the recovery of plasma volume, plasma osmolality, renal water and sodium handling and fluid-regulating hormones to dehydrating exercise in well-trained women and compared them to men. Ten male and eight female athletes cycled at anaerobic threshold at an ambient temperature of 32°C until dehydration by 3% of their body mass (m_b). After exercise, they drank water equal to 1% m_b and rested for 240 min. Plasma renin activity (PRA), serum aldosterone $[ALDO]_s$, plasma arginine vasopressin $[AVP]_{pl}$, norepinephrine concentrations and plasma osmolality (Osm_{pl}) were determined at baseline, end of exercise, 30, 60, 120 and 240 min postexercise. Urine was collected at baseline, end of exercise, 60, 120 and 240 min postexercise. Renal free water and sodium handling were assessed. The recovery of OSM_{pl} and plasma volume occurred within the first 60 min of recovery and at similar rates between the groups. However, women had lower PRA at the end of exercise ($P = 0.05$), an earlier recovery of $[ALDO]_s$, and a slower $[AVP]_{pl}$ recovery. Overall fluid balance was similar between the men and women, as were the early recovery of renal free water clearance (C_{H_2O}). During the last 120 min of recovery C_{H_2O} was more negative (greater water reabsorption) and fractional sodium excretion was increased in the women compared to the men. Despite small differences in sodium and water reabsorption following dehydration, it appears from other study that recovery from dehydrating exercise in well-trained men and women is remarkably similar.

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Key words Women · Fluid-regulating hormones · Heat stress · Dehydration · Exercise recovery

Introduction

Most studies describing fluid balance during recovery from dehydrating exercise have been conducted on men (Costill et al. 1974; Nose et al. 1988a, b, c). However, it has been reported that women have different responses to exercise in the heat, having for example lower sweat rates for similar core temperatures (Frye and Kamon 1983; Morimoto et al. 1967; Paolone et al. 1978; Sawka et al. 1983; Shapiro et al. 1980; Weinman et al. 1967; Wyndham et al. 1967). Earlier investigations have failed to find differences between men and women to be enhanced due to dehydration (Sawka et al. 1983) and others have noted no sex differences in the major fluid-regulating hormones in response to exercise in the heat (Fransconi et al. 1985). These findings would argue against sex differences in the fluid regulatory and renal responses to exercise in stressful environments, but these investigations did not allow for differences due to the menstrual cycle. Because thermoregulatory and hormonal responses have been shown to be influenced by the phase of the menstrual cycle (De Souza et al. 1989; Stephenson et al. 1989) and by estrogen (Forsling et al. 1981), variable thermoregulatory responses in the women may have hidden sex differences occurring in fluid balance.

Even though some attention has been given to the thermoregulatory responses of women during exercise in stressful environments, the fluid balance, volume-regulating hormones and kidney function have not been evaluated in women during the period following exercise. Therefore, the aim of the present investigation was to document the recovery of plasma volume, plasma osmolality, fluid-regulating hormones and renal function during the period following dehydrating

exercise in well-trained women and men. Based on earlier experiments demonstrating lower sweating during exercise in the heat in women and the known effects of oestrogen on water retention, we hypothesized that following dehydrating exercise renal water and sodium retention would be greater in women despite lower or comparable concentrations of fluid-regulating hormones.

Methods

Subjects

The subjects were well-conditioned male ($n = 10$) and female ($n = 8$) cyclists and bi-athletes, who had no known medical problems and were taking no medications. The women had a history of consistent recurrence of menstruation at intervals of 23–33 days. To reduce effects due to phase of the menstrual cycle, all the women were tested during the first 8 days of their menstrual cycles (De Souza et al. 1989; Kolka and Stephenson 1989). The subjects were informed of the risks associated with the study and gave their written consent. The protocol was approved by the Human Investigations Committee at Lenox Hill Hospital.

Protocol

Day 1. maximal oxygen consumption assessment

Training status and ventilatory threshold (VT) were assessed with a progressive maximal oxygen consumption stress test on a cycle ergometer. The subjects performed an incremental test to exhaustion on a Monark 818 cycle ergometer (Monark AB, Varberg, Sweden). The subjects pedaled at 80 or 90 rpm and exercise intensities were increased by 25–30 W every minute (Wasserman 1984). Oxygen consumption and carbon dioxide production were continuously measured with 20 s averaging by a Sensor Medics 2900 metabolic cart (Sensor Medics Corp, Yorba Linda, Calif.) previously calibrated with standardized gases. Heart rate was monitored continuously with three leads by a Sensor Medics Horizon ECG (Sensor Medics Corp).

VT determination

The VT was determined by visual inspection of graphs of ventilatory equivalent for oxygen ($\dot{V}E_{O_2}$) and carbon dioxide ($\dot{V}E_{CO_2}$) by the methods of Wasserman (1984) from the $\dot{V}O_{2peak}$ test. The point where ($\dot{V}E_{CO_2}$) reached a nadir and then showed a trend upwards while $\dot{V}E_{CO_2}$ maintained a plateau or declined was defined as VT.

Pretest status

To ensure sodium balance on days prior to the experimental protocol, for 2 days prior to dehydration testing, the subjects ingested 2-g sodium chloride day^{-1} in addition to their normal energy and sodium intake. On the day prior to the fluid balance study, 24-h urine was collected and sodium balance determined from 24-h sodium excretion. All the subjects excreted more than 2 g of sodium over the 24-h collection period [5.8 (SEM 1.0) $g \cdot 24\ h^{-1}$] indicating adequate sodium balance. The subjects were weighed on two earlier visits to the laboratory to establish their baseline body mass. The women were weighed during their follicular phase. The subjects refrained from exercise for 24 h prior to the experiment.

Day 2: dehydration test

On the day of the dehydration tests, the subjects arrived at the laboratory between 0600–0900 hours having eaten a light breakfast. The subjects refrained from any caffeinated or alcoholic beverages for at least 12 h prior to the study. The subjects were instructed to drink 5 $ml \cdot kg^{-1}$ of water at home and hydration status was verified by baseline urine osmolality (< 500 $mosmol \cdot kg\ H_2O^{-1}$). One (male) subject did not meet this criterion and was asked to return on a separate day for the test. Upon arriving at the laboratory, the subjects voided urine and an 18-gauge Med teflon intravenous cannula was inserted into a forearm vein and connected to a 500-ml solution of heparinized normal (0.9% NaCl) saline. Following the intravenous catheter insertion, the subject rested for 1 h in a semi-recumbent position. The subject remained in this position while a baseline blood sample was drawn, after which the subject moved to the cycle ergometer. For baseline and subsequent sampling, the line was flushed with approximately 3–4 ml saline (1.5 ml dead space) and 20–22 ml blood drawn for analysis. During the baseline period heart rate was monitored by electrocardiogram and blood pressure by auscultation of the brachial artery with an automated blood pressure cuff (Colin, STBP-780) was measured at the end of the rest period. Cuff size to arm circumference ratio was maintained within limits recommended by the American Heart Association. Urine was collected at the end of the baseline period.

The temperature in the laboratory was maintained at 32°C, with a relative humidity of less than 40% for all dehydration tests. The subjects exercised on the Monark cycle ergometer for 25 min periods with 5 min rests until they had lost 3% of their baseline body mass (Francesconi et al. 1985; Sawka et al. 1983, 1985). During the 5-min rest periods, the subjects were towel dried and weighed in dry clothing to monitor loss of body mass. All exercise was performed at the oxygen uptake ($\dot{V}O_2$) corresponding to VT (about 60% of maximal oxygen consumption). During exercise, $\dot{V}O_2$ was monitored 15 min into each exercise bout to verify the exercise intensity. When 3% loss of body mass was achieved, the subject returned to the ergometer, pedaled for 15 min, after which blood was drawn through the indwelling catheter while the subject was still pedaling. Following the blood sampling, the subject dismounted, voided urine and assumed a semi-recumbent position. The subjects then consumed an amount of water to match 1% of their initial body mass within the first 30 min of recovery. Blood was sampled at 30, 60, 120 and 240 min and urine collected at 60, 120 and 240 min following exercise. Heart rate was continuously monitored with a Polar Accurex heart rate monitor (Polar USA, Inc., Stamford, Conn.) and recorded every 60 min. Blood pressure was measured and recorded every 60 min manually during exercise and with the automated cuff during recovery. Resting metabolic rate was determined with the dilution method (Sensor Medics 2900, Sensor Medics Corp.) at baseline, 60, 120 and 240 min of recovery.

Blood and urine analysis

Blood was distributed into tubes without anticoagulant for serum (serum separation tube, Bectin/Dickinson, Rutherford, N.J.), heparinized tubes with ethylene diaminotetra-acetic acid (EDTA) for plasma and hematocrit, and tubes with potassium oxalate and sodium fluoride (glycolytic inhibition tube, Bectin/Dickinson), for lactate analysis. Blood samples for catecholamines were placed in tubes with EDTA and glutathione. Blood samples were kept on ice following sampling and analyzed immediately for lactate concentration. Blood in the tube without anticoagulant was centrifuged at

3000 rpm for 10 min. One of the tubes of heparinized blood was used for determining hematocrit and hemoglobin concentration and another for plasma osmolality (Osm_{pl}). The remainder of blood in heparinized tubes was centrifuged and plasma removed with a glass pipette and stored at −70°C until analysis for plasma renin activity (PRA), and arginine vasopressin (AVP) and catecholamines concentrations. Serum potassium, sodium, creatinine and glucose concentrations were determined using a Technicon SMAC II system. Serum aldosterone concentration ($[ALDO]_s$ was determined using the solid phase antibody method (Coat-A-Count, Diagnostic Product Corp, Los Angeles, Calif.). Blood lactate concentration $[La^-]_b$ was analyzed with a Yellow Springs Instruments (Yellow Springs, Ohio) lactate analyzer and catecholamine concentrations were analyzed by high performance liquid chromatography with electrochemical detection (Waters, Milford, Md.). The samples were run in duplicate according the methods of Shoup and Keefe (1980) with alumina extraction. Intra-assay coefficients of variation were 3.4% for norepinephrine (NE) and 8.5% for epinephrine (EPI). PRA was analyzed in duplicate with a competitive-binding method with single antibody (New England Nuclear, Billerica, Md.) according to the methods of Yallow and Berson (1971). Intra-assay coefficient of variation for PRA was 7.3%. Plasma arginine vasopressin concentration ($[AVP]_{pl}$) was measured via standard radio-immuno assay methods (Incstar Corp, Stillwater, Minn). Intra-assay coefficient of variation for $[AVP]_{pl}$ was 4.4%. Plasma (Osm_{pl}) and urine (Osm_u) osmolality were determined using the freezing-point depression method. Urine was frozen at −70°C until analysis. Analysis of urinary sodium, potassium, and creatinine concentrations was made with a Beckman Astra eight Chemical Analyzer (Beckman, Los Angeles, Calif.).

Statistics

Recovery responses to the dehydration challenge were compared using a two-way (sex × time) analysis of variance for repeated measures. Where baseline differences between men and women were evident, the baseline value was entered into the analysis as a covariate (ANCOVA). To stabilize the variance, log transformations were performed on the PRA, $[AVP]_{pl}$ and $[ALDO]_s$ data, but in subsequent tables and figures the means and standard errors from the raw data are reported. Differences between the sexes and with time were accepted at $P < 0.05$. The Bonferroni t procedure was used for a priori multiple comparisons (Kirk 1982). Power and sample size were calculated based on effect sizes reported in the literature. An estimated sample size of eight subjects per group at an α level of 0.05 yielded a power level exceeding 0.80 for all variables. Statistical analyses were done with the BMDP Statistical Software Package (Berkely, Calif.).

Calculations

Percentage change in plasma volume (%Δ PV) was calculated with changes in hematocrit and hemoglobin (Dill and Costill 1974). Renal function was assessed by estimating glomerular filtration rate (GFR) with creatinine clearance and the quantification of renal water excretion was assessed through the calculation of osmolar and free water clearances (Pitts 1974). Because of the differences in body size between men and women, all renal function parameters were normalized for body size by dividing by body surface area (BSA) · 1.73^{-1} (Pitts 1974). Fluid loss during exercise was determined by subtracting urine and sweat loss and adding fluid intake and is presented cumulatively over the recovery period. Fractional excretion of Na^+(FE_{Na^+}) was calculated by the following equation (Takamata et al. 1994):

$$FE_{Na^+} = 100 \cdot (U_v \cdot [Na^+]_u / GFR \cdot [Na^+]_f)$$

$$[Na^+]_f = DF \cdot [Na^+]_s$$

where f is glomerular filtrate, u is urine and s is serum, U_v is urine flow rate, $[Na^+]_s$ is serum sodium concentration and DF is the Donnan factor for cations (i.e. 0.95; Koeppen 1990).

Results

Subject characteristics

The women were significantly shorter, weighed less and had significantly higher percentages of body fat than the men (Table 1). The men and women were both well-trained athletes but the men had significantly higher $\dot{V}O_{2peak}$. Both groups attained their VT at a similar $\dot{V}O_2$ and % $\dot{V}O_{2peak}$ indicating the exercise was performed at a similar relative intensity.

Baseline

There were no significant baseline sex differences in Osm_{pl}, $[Na^+]_s$, $[K^+]_s$, PRA, $[ALD]_s$ or plasma norepinephrine $[NE]_P$, but $[AVP]_{pl}$ was significantly, lower in the women (Table 2, Figs. 1, 2a,b). Baseline Osm_u, U_v, renal osmolar (C_{osm}), free water (C_{H_2O}) clearance, sodium ($U_{Na^+}V$) and potassium ($U_{K^+}V$) excretion were not significantly different between men and women (Table 3, Fig. 3). Baseline Osm_u: Osm_{pl} ratio was 1.17 (SEM 0.19), and 0.84 (SEM 0.27), (NS) in the men and women respectively.

Exercise responses

The men required 129.6 (SEM 14.3) min (range 60–190) and the women 121.3 (SEM 10.0) min (range 90–160) of exercise to lose 3% of their body mass. End exercise $\dot{V}O_2$ was 35.72 (SEM 1.72) $ml \cdot kg^{-1} \cdot min^{-1}$ for the men and 34.82 (SEM 1.38) $ml \cdot kg^{-1} \cdot min^{-1}$ for the

Table 1 Characteristics of subects. $\dot{V}O_{2peak}$ Peak oxygen consumption, VT ventilatory threshold, FFM fat free mass, $\dot{V}O_2$ oxygen uptake

	Men ($n = 10$)		Women ($n = 8$)	
	mean	SEM	mean	SEM
Age (years)	28.6	1.6	31.1	1.5
Body mass (kg)	72.0	2.6*	60.2	1.8
Height (cm)	178.5	1.9*	166.3	1.4
Body fat (%)	10.8	0 8*	14.7	2.6
$\dot{V}O_{2peak}$, ($l \cdot min^{-1}$)	4.682	0 191*	3.521	0.157
$\dot{V}O_{2peak}$, ($ml \cdot kg^{-1}$ body mass $\cdot min^{-1}$)	65.0	1.7*	58.7	2.3
$\dot{V}O_{2peak}$, ($ml \cdot kg^{-1}$ FFM $\cdot min^{-1}$)	74.0	1.5	68 5	2.3
$\dot{V}O_2$ at VT, ($ml \cdot kg^{-1}$ body mass $\cdot min^{-1}$)	39.3	1.5	36 8	2.8
% $\dot{V}O_{2peak}$ at VT	60.4	1.6	63.1	1.7

*Men different to women, $P < 0.05$

Table 2 Concentrations of serum sodium ($[Na^+]_s$) and pottassium ($[K^+]_s$) in men and women under baseline conditions, after dehydration and at 30, 60, 120 and 240 min of recovery

	Rest		End exercise		30-min recovery		60-min recovery		120-min recovery		240-min recovery	
	mean	SEM	mean	SEM	mean	SEM	mean	SEM	mean	SEM	mean	SEM
$[Na^+]_s$, (mmol l^{-1})												
Men	138.7	0.7	141.4	0 7	140.4	1.0	138.1	0.9	139.2	0.9	139.0	0.7
Women	138.8	0.7	143.7	0.7	140.4	1.0	140.4	1.3	139.4	0.5	140.0	0.7
$[K^+]_s$, (mmol l^{-1})												
Men	4.11	0.09	4.86	0.09	4.35	0.08	4.41	0.06	4.33	0.07*	4.17	0.06*
Women	4.05	0.09	5.03	0.08	4.39	0.08	4.33	0.13	4.14	0.07	4.06	0.07

*Men different to women, $P = 0.08$

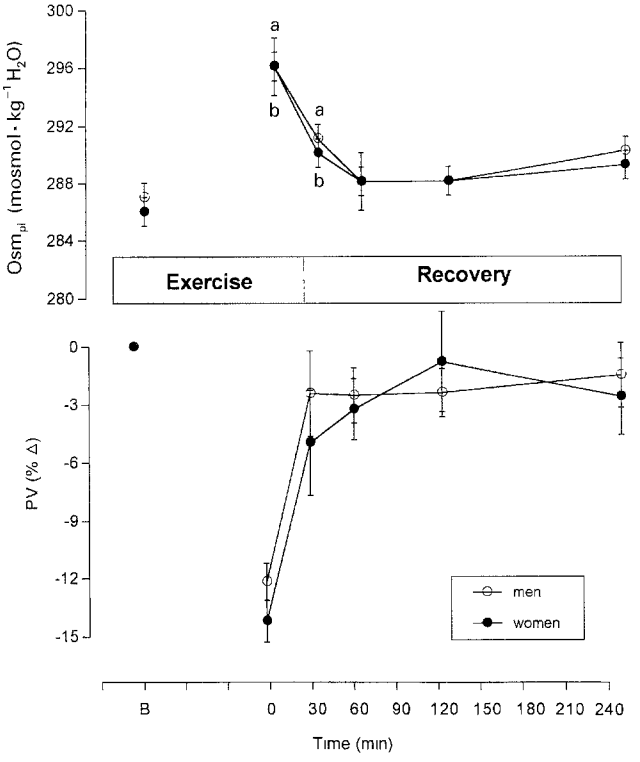

Fig. 1 Plasma osmolality (Osm_{pl}) and percentage change from baseline in plasma volume ($\%\Delta PV$) for men and women at the baseline, end of exercise and at 30, 60, 120 and 180 min of recovery. Mean and SEM. Exercise time varied from 60 to 190 min. [a]Women different from baseline, $P < 0.05$. [b]Men different from baseline, $P < 0.05$

women. End of exercise heart rates were 164 (SEM 3) beats·min^{-1} and 156 (SEM 4) beats·min^{-1} (NS), for the men and women, respectively. The dehydration and exercise resulted in a %ΔPV of − 10.19 (SEM 0.96)% and − 12.21 (SEM 0.96) % and Osm_{pl} increased significantly to 295 (SEM 1.38) mosmol·kg·H_2O^{-1} and 296 (SEM 0.86) mosmol·kg·H_2O^{-1} in the men and women, respectively (Fig. 1). Blood lactate concentration was 1.0 (SEM 0.1) and 1.0 (SEM 0.1) mmol·l^{-1} for the men and women, respectively. There were no differences between men and women in $[K^+]_s$ or $[Na^+]_s$ following exercise (Table 2). Exercise resulted in significant increase in $[AVP]_{pl}$, $[ALDO]_{pl}$, PRA and $[NE]_{pl}$ in both the men and women (Fig. 2a,b). At the end of exercise, the men had significantly higher PRA than the women (Fig. 2b). None of the other hormones showed any sex differences at the end of the exercise, although $[NE]_{pl}$ was 30% higher in the men (Fig. 2b, NS). There were no significant sex differences at the end of exercise for GFR, osm_u, C_{osm}, C_{H_2O}, Creatinine clearance (C_{Cr}), $U_{Na^+}V$ or $U_{K^+}V$ (Table 3, Fig. 3). The GFR data should be interpreted with caution because low urine volume (i.e. < 100 ml) can lead to errors in the calculation of C_{Cr} due to incomplete bladder emptying. At the end of exercise serum concentration of albumin was increased from 40.4 (SEM 6.6) g·L^{-1} and 37.5 (SEM 10.1) g·L^{-1} to 49.2 (SEM 6.3) g·L^{-1} and 46.8 (SEM 1.1) g·L^{-1}, in the men and women, respectively ($P < 0.05$). Two of the men and one of the women were unable to give a urine sample at the end of exercise. They were excluded from subsequent analysis for renal function variables.

Recovery

Blood variables

The %ΔPV showed only minor differences from baseline by 30 min postexercise, and Osm_{pl} had returned to baseline levels by 60 min following exercise in the men and women (Fig. 1). Men had higher $[ALDO]_s$ during recovery at 60 and 120 min postexercise compared to the women (Fig. 2a) and their $[ALDO]_s$ was significantly greater than baseline through 120 min of recovery. In contrast, the women restored baseline levels of $[ALDO]_s$ by 60 min following exercise. Despite the higher end exercise PRA in the men, there were no further sex differences in PRA at any recovery time and no difference in the rate of recovery. There were no sex differences in $[NE]_{pl}$ at any time during recovery which followed a similar recovery pattern to PRA, remaining above baseline through 30 min of recovery in the men only (Fig. 2b). Although there were no sex differences in $[AVP]_{pl}$ at the end of exercise or at any time during

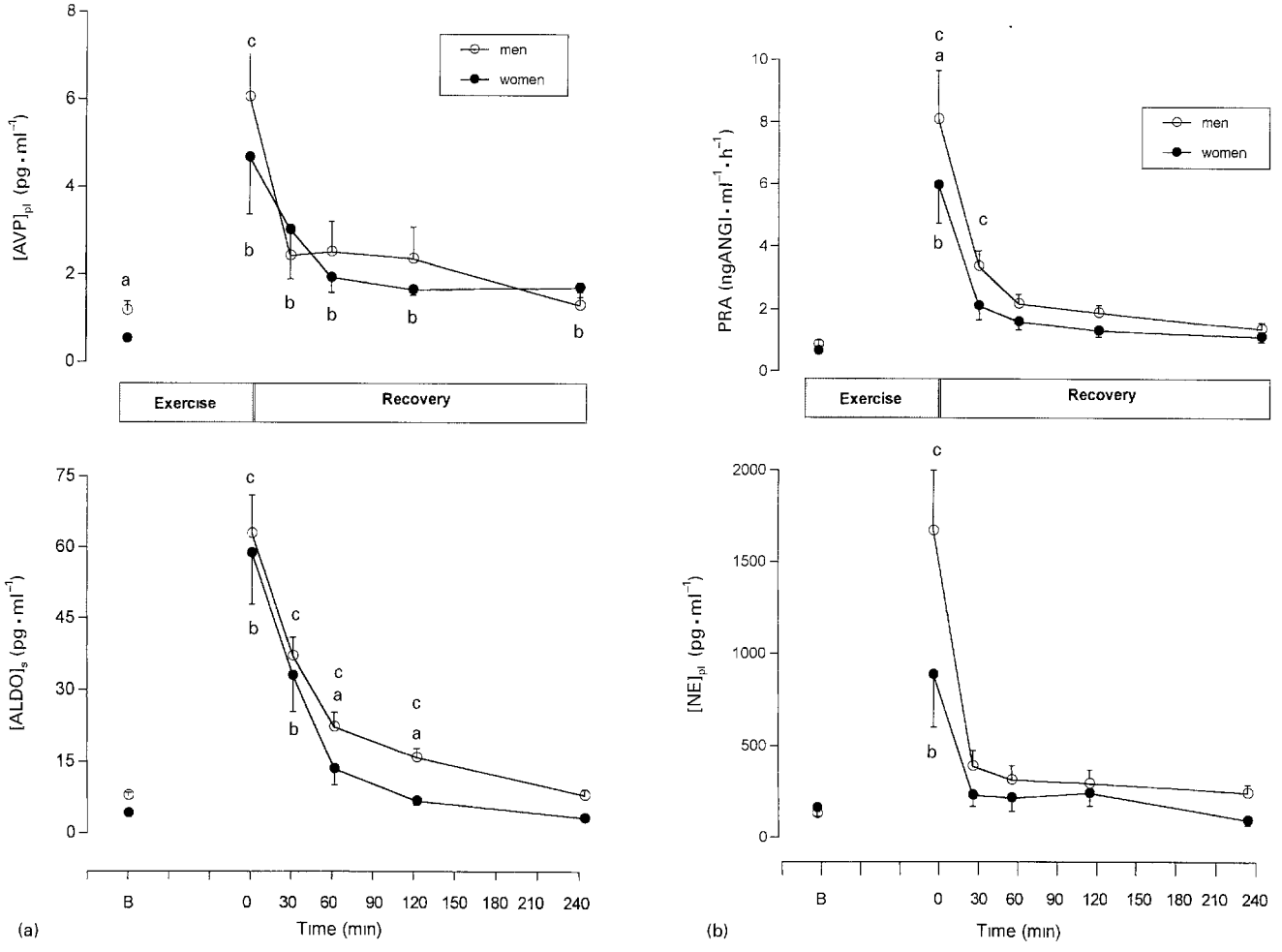

Fig. 2a Concentrations of plasma arginine vasopressin ($[AVP_{pl}]$), serum aldosterone ($[ALDO]_s$) Mean and SEM. Exercise time varied from 60 to 190 min. [a]Men different from women. $P < 0.05$. [b]Women different from baseline. $P < 0.05$. [c]Men different from baseline, $P < 0.05$. **b** Plasma renin activity (*PRA*) and concentration of plasma norepinephrine ($[NE]_{pl}$) at the baseline, end of exercise and at 30, 60, 120 and 180 min of recovery. Mean and SEM. Exercise time varied from 60 to 190 min. [a]Men different from women, $P < 0.05$. [b]Women different from baseline, $P < 0.05$. [c]Men different from baseline. $P < 0.05$. AngiotensinI (ANGI)

recovery (Fig. 2a), $[AVP]_{pl}$ was greater than baseline throughout the 240 min recovery period in the women, but returned to baseline in the men by 60 min postexercise. There were no differences in the $[Na^+]_s$ or $[K^+]_s$, until 120 and 240 min of recovery when $[K^+]_s$ was higher in the men. Serum concentration of albumin remained significantly elevated in both the men and the women through 120 min of recovery [43.1 (SEM 0.8) $g \cdot L^{-1}$ and 40.1 (SEM 1.4) $g \cdot L^{-1}$] and through 240 min of recovery in the men only [43.1 (SEM 0.9) $g \cdot L^{-1}$ and 38.7 (SEM 10.0) $g \cdot L^{-1}$, for the men and women, respectively].

Renal water and sodium handling

There was no effect of sex on Osm_u or Osm_u: Osm_{pl} but C_{H_2O} was significantly more negative (greater water reabsorption) and FE_{Na^+} increased (lower sodium reabsorption) in the women at 240 min of recovery (Table 3, Fig. 3, $P < 0.05$). The $U_{K^+}V$ was also greater at the end of the recovery period in the women (Table 3, $P < 0.05$). C_{H_2O} was significantly lower than baseline only in the women at the end of recovery ($P < 0.05$).

Discussion

This investigation is the first to document the responses of the fluid-regulating hormones during the period following dehydrating exercise in endurance trained women and compare them to the responses of endurance trained men. The major finding was that in equally trained men and women recovery from exercise was similar for most blood and renal fluid and osmoregulatory parameters following dehydrating exercise in the heat. As expected, $[AVP]_{pl}$, $[ALDO]_s$ and PRA

Table 3 Renal function in men and women under baseline conditions, after dehydration and at 120 and 240 min of recovery. *GFR* Glomerular filtration rate, U_v urine excretion rate, C_{osm} renal osmolar clearance, C_{H_2O} renal free water clearance, $U_{Na+}V$ urine sodium excretion, $U_{K+}V$ urine potassium excretion are expressed as millimoles per litre. *GFR*, U_v, C_{osm}, C_{H_2O}, $U_{Na+}V$ and $U_{K+}V$ were corrected for body surface area · 1.73^{-1}

	Baseline		End exercise		120-min recovery		240-min recovery	
	mean	SEM	mean	SEM	mean	SEM	mean	SEM
GFR (ml · min^{-1})								
Men	123.8	21.5	68 8	14.3	119.9	18.7	136.8	22.6
Women	125 5	14.1	84.7	18.7	118.1	26.9	151.2	10.8
U_v (ml l^{-1})								
Men	2.74	0.71	0.86	0.35	1.12	0.28	0.62	0.10[b]
Women	3.27	0.93	0.76	0.23[b]	0.41	0.07[b]	0.99	0.17
C_{osm} (ml l^{-1})								
Men	2.90	0.40	1.18	0.23	2.62	0.40[a]	1.66	0 34[a]
Women	2.02	0.52	1.15	0.37	1.25	0.23	3.13	0 55
C_{H2O} (ml l)								
Men	− 0.11	0.53	− 0.21	0.32	− 1.39	0.26	− 1.03	0.21[a]
Women	1.32	0.70	− 0.39	0.25	− 0.86	0.25	− 2.18	0.37[b]
$U_{Na+}V$ (mmol l^{-1})								
Men	11.3	2.5	4.3	2.6	7.6	0.8	10.3	2.5
Women	9.2	3.2	3.0	1.3	3.6	1.7	13.5	3.3
$U_{K+}V$ (mmol · l^{-1})								
Men	4.8	1.1	5.4	1.4	8.4	1.4	6.9	1.2[a]
Women	5.9	1.5	6.1	1.6	6.7	2.1	14.5	3.2
U_{osm}/Osm_{pl}	1.17	0.19	1.58	0.31	2.61	0.23[b]	3.04	0.15[b]
	0.84	0.27	1.59	0.21	2.83	0.22[b]	3 13	0.36[b]

[a]Men different to women
[b]Different to baseline, $P < 0.05$

were all increased at the end of exercise (Opstad et al. 1985; Wade 1984), but PRA was significantly lower in the women. There were no sex differences in the recovery of Osm_{pl}, but $[AVP]_{pl}$ recovery was slower and $[ALDO]_s$ recovery more rapid in the women compared to the men. The altered recovery of these hormones in the women was associated with decreased sodium reabsorption (greater FE_{Na^+}) but increased water reabsorption (negative C_{H_2O}) late in recovery. There were no sex differences in PRA or $[NE]_{pl}$ during recovery and no differences between the men and women in overall fluid balance during exercise or recovery.

PV and Osm_{pl}

There was a rapid recovery of Osm_{pl} and PV in both the men and women. The subjects had lost 3% of their body mass through exercise after which they drank 1% of their body mass in water. However, greater than 96% of the PV reduction from exercise was replenished during the first 30 min of recovery. The PV recovery was due to the absorption of the ingested water into the vascular space and/or the reabsorption of fluid into the vasculature from the extravascular compartment (Nose et al. 1988b). Recovery of PV has been shown to be mediated by a number of conditions that favor absorption at this time. The increased osmotic pressure due to high $[Na^+]_{pl}$ (Nose et al. 1988b); increased oncotic pressure due to the elevated serum albumin concentration throughout most of recovery (Senay et al. 1980; Gillen et al. 1991); and Starling forces also favor absorption during recovery because of a postexercise fall in hydrostatic pressure inside the vasculature (Morimoto 1990). Therefore, PV was *selectively* restored at the expense of intracellular and interstitial water replenishment. Further restitution of fluid to these compartments did not occur because our protocol limited drinking.

Even though our protocol limited fluid intake, it is unlikely that these compartments would have been restored within 4 h even with *ad libitum* drinking (Greenleaf 1992; Nose et al. 1988c). The selective restoration of PV results in the suppression of the fluid regulating hormones and their fluid retention effects on the kidney. In addition, it has been shown that with dehydration, $[AVP]_{pl}$ is reduced immediately following drinking (Geelen et al. 1984). Because Osm_{pl} and PV the primary regulators of $[AVP]_{pl}$, are unchanged at this point, the reduction in $[AVP]_{pl}$ is probably due to oropharyngeal factors. The fall in $[AVP]_{pl}$ not only reduces fluid retention, but has been shown to be accompanied by thirst inhibition (Thompson et al. 1987), limiting fluid intake and restoration of all body fluid compartments.

Despite the early recovery of PV and Osm_{pl} in men and women, the return of $[AVP]_{pl}$ to baseline was slower in the women. This sustained increase in $[AVP]_{pl}$ may be related to the levels of endogenous estrogen in the women and is consistent with the findings of Forsling et al. (1982) in that the administration of estrogen in postmenopausal women has led to increases in $[AVP]_{pl}$. The concentrations of estrogen in the plasma achieved by Forsling et al. (1982) were similar to those seen in the follicular phase of the

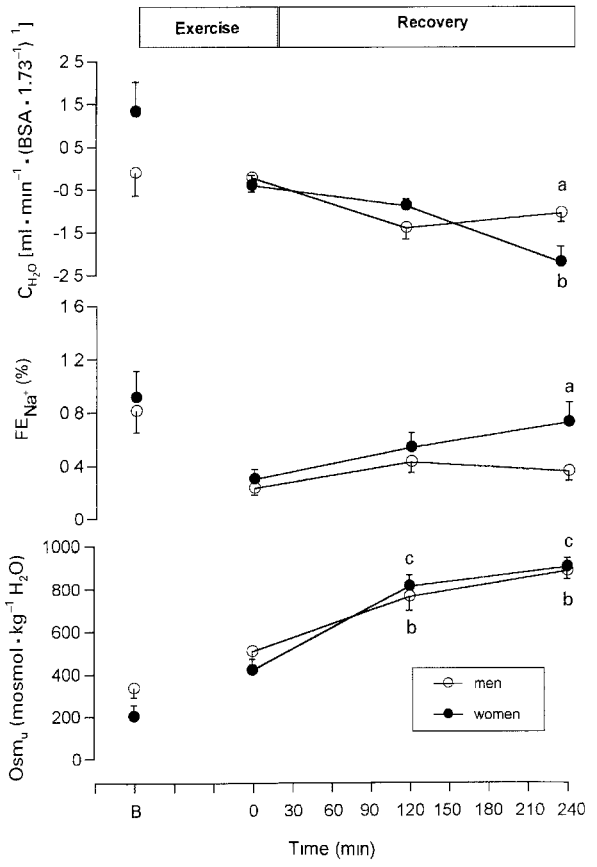

Fig. 3 Free water clearance (C_{H_2O}), fractional excretion of sodium (FE_{Na^+}) and urine osmolality (Osm_u) at the baseline, end of exercise and at 30, 60, 120 and 180 min of recovery in men ($n = 8$) and women ($n = 7$). Means and SEM. C_{H_2O} is corrected for body surface area (BSA)·1.73^{-1} to adjust for differences in body size. Exercise time varied from 60 to 190 min. Means and SEM. [a]Men different than women, $P < 0.05$. [b]Women different from baseline, $P < 0.05$. [c]Men different from baseline, $P < 0.05$

menstrual cycle (72 pmol·l^{-1} to 313 pmol·l^{-1}). There is also evidence that exogenous estrogen leads to increased water retention through its effects on $[AVP]_{pl}$ in both postmenopausal (Aitken et al. 1974) and young women (Bland et al. 1974), and that the $[AVP]_{pl}$ response to increased Osm_{pl} is greater during pregnancy (Davison et al. 1984). These effects on AVP suggest the greater endogenous concentrations of oestrogen may be a mediator of the women's sustained elevations of $[AVP]_{pl}$ and water retention in the present investigation.

The women's faster recovery of $[ALDO]_s$ and increased sodium excretion may also have been due to oestrogen effects on the regulation of body water and sodium. A recent study has found that the increase in $[ALDO]_s$ normally seen during exercise was suppressed in women (age 55 years) taking oestrogen (Tankersley et al. 1992). Postexercise 24-h urine volume, osmolality, or sodium concentration were unaffected by the lower $[ALDO]_s$, which has suggested increased tubular sensitivity to $[ALDO]_s$ in women on oestrogen therapy (Tankersley et al. 1992). In the present investigation, the lower sodium excretion in the women is consistent with their faster recovery of $[ALDO]_s$ indicating similar tubular sensitivity to aldosterone between men and women.

Another finding in this study was the lower exercise PRA in women compared to men. The attenuated PRA response in women is most likely due to their lower concentrations of plasma catecholamines because $[NE]_{pl}$ and PRA have been shown to be closely related during exercise (Kotchen et al. 1971). Although the sex differences in $[NE]_{pl}$ failed to attain statistical significance, the $[NE]_{pl}$ was 30% lower in the women compared to the men at the end of exercise. The increased $[NE]_{pl}$ may have been due to small differences in exercise intensity between the men and women. Even though the men and women were exercising at VT, $\dot{V}O_{2peak}$ was greater in the men so they were exercising at a 30% higher external work rate (212 W compared to 170 W). Both $[NE]_{pl}$ and PRA have been shown to increase in a curvilinear manner above 50% of $\dot{V}O_{2peak}$ (Convertino et al. 1981; Kotchen et al. 1971) so even a small increase in the relative exercise intensity may have caused large differences in the concentrations of these substances. However, $[la^-]_b$ were identical in the men and women (1.0 mmol·l^{-1}) and PRA has been shown to be related to the lactate concentration (Gleim et al. 1984) and VT (Stachenfeld et al. 1995). The ventilatory and $[la^-]_b$ measurements indicate that while intensity was slightly higher in the men, it is unlikely that this difference represented a great enough increase to induce large increases in PRA.

Another possible explanation for the increased PRA in the men is increased body temperature. While we did not measure core temperature, it is unlikely that core temperature would have been greater in the men than the women. Sawka et al. (1983) have found slightly higher rectal temperatures in women during exercise in hot-wet and hot-dry environments, but the women were less trained and there was no evaluation of the phase of the menstrual cycle. When training status is similar between men and women, and the women are tested during the follicular phase, no sex differences in core temperature have been found during submaximal exercise in the heat (Kolka et al. 1987). In addition, the differences in PRA are not explained by differences in ΔPV (i.e. through renal baroreceptors) because these changes were similar [− 10.19 (SEM 0.96)% and − 12.21 (SEM 0.96)%, for the men and women, respectively] and there were no differences in mean arterial pressure at the end of exercise [90 (SEM 4) versus 88 (SEM 4) mm Hg (12.0 SEM 0.53 and 11.73 SEM 0.53 KPa), for the men and women, respectively].

Renal water and osmoregulation

Despite the restoration of pre-exercise PV and Osm_{pl}, the subjects were still actively retaining water

throughout recovery. Renal free C_{H_2O} was negative, and Osm_u increased relative to Osm_{pl} (Osm_u:Osm_{pl}) throughout the 240-min recovery period. The responses of most renal fluid and osmoregulatory variables were similar between the men and the women until the last 120 min of recovery. By the end of recovery, C_{H_2O} was more negative in the women, indicating greater water reabsorption at this time. The increased rate of water reabsorption in the women was possibly related their elevated $[AVP]_{pl}$ above baseline. While overall fluid balance was virtually identical between the men and women, during the latter 120 min of recovery, C_{H_2O} was reduced in the women, which was consistent with their elevated $[AVP]_{pl}$.

Atrial natriuretic peptide (ANP) was not measured in this study and is a key regulator of water and sodium balance. Increases in ANP have been demonstrated following acute (Tanaka et al. 1986) and long-term exercise (Nose et al. 1994). During short-term exercise, this increase has been attributed directly to atrial stretch (Tanaka et al. 1986), but during long-term exercise factors such as catecholamines, heart rate and core temperature may be more important for ANP release. Nose et al. (1994) have demonstrated that changes in ANP were related to changes in blood volume during long-term exercise. Blood volume was not measured in our subjects, but is typically higher in men compared to women. The similar %ΔPV would suggest greater absolute changes in blood volume in the men. This, and the greater increases in $[NE]_{pl}$ may have led to a greater release of ANP and may explain the men's lower water retention and more rapid $[AVP]_{pl}$ recovery. This explanation is weakened, however, by the men's slower recovery of $[ALDO]_s$, which has been shown to be reduced at high levels of ANP (Shenker 1989).

The greater water retention in the women late in recovery may have been related to relatively greater shifts of blood flow away from the renal and splanchnic regions. Despite the similar %ΔPV, $[NE]_{pl}$ and PRA were lower in the women at the end of exercise suggesting different degrees of sympathetic nervous stimulation. Afferent sympathetic signals ascend to the central nervous system that modulate sympathetic efferents originating in the liver, indicating the presence of baroreceptors in the splanchnic region. However the physiological significance of these receptors has not been established. Furthermore, $[NE]_{pl}$ and PRA were no different between the men and women at the end of recovery when differences in water retention were found.

Baseline differences

Even though the differences were not significant, the women had higher baseline U_V, $U_{Na^+}V$ and C_{H_2O}. The baseline Osm_u: Osm_{pl} [1.17 (SEM 0.19) and 0.84 (SEM 0.27) in the men and women, respectively] suggests that the women were excreting dilute urine and may have been slightly more hydrated than the men, thus explaining their lower baseline $[AVP]_{pl}$. These differences did not seem to have an influence on exercise performance because there were no sex differences in the exercise time required to lose 3% of their body mass. However, given earlier work that demonstrated lower sweating rates for similar core temperatures in women, the men should have lost water faster during exercise (Frye and Kamon 1983; Morimoto et al. 1967; Shapiro et al. 1980; Weinman et al. 1967; Wyndham et al. 1967). It is conceivable that the baseline lower hydration level in the men led to an attenuation of their sweat rate which resulted in similar sweat rates between the men and women. This explanation is unlikely because these differences between men and women have been found only in hot-humid environments, whereas the relative humidity in our laboratory was less than 40%. In addition, the similarity of the early recovery responses and more negative C_{H_2O} later in recovery suggest that these baseline differences had little influence on the recovery of most renal fluid and osmoregulatory responses to dehydration in these athletes.

Menstrual cycle phase

We chose to test the women during the follicular phase of their menstrual cycles because most physiological and clinical research in women is conducted during this phase. However, had we conducted the tests during the luteal phase, differences between the men and women may have been enhanced. Lower resting PV and greater fluid shift out of the vascular space during exercise have been noted in response to exercise during the luteal phase (Stephenson and Kolka 1988; Stephenson et al. 1989). Increased $[ALDO]_s$ (De Souza et al. 1989), PRA and angiotension concentrations (Stephenson et al. 1989) in response to exercise have also been noted during the luteal phase. All of these hormones result in the retention of fluid in the plasma, so had we tested the women during the luteal phase, higher concentrations of these hormones may have acted to increase fluid retention further in the women during recovery.

Lean body mass

The men had significantly greater lean body mass than the women, indicating greater total body water. Therefore, the 3% loss of mass was a larger percentage of total body water for the women. Despite this difference, the women's overall response to the dehydration challenge was less stressful than the men's (i.e. lower end exercise $[NE]_{pl}$ and PRA). In addition, because the men had greater lean body mass, they also had a higher

metabolic rate during recovery [319.5 (SEM 34.1) $ml \cdot min^{-1}$ and 281.1 (SEM 44.1) $ml \cdot min^{-1}$, for the men and women, respectively] perhaps leading to greater metabolic water production in the men. However, because metabolic rate was so low during the recovery period, it is unlikely that water production influenced these results.

Summary and conclusions

Following dehydrating exercise, well-trained men and women had similar responses for the early (under 120 min) part of recovery. The women had a significantly greater rate of water reabsorption during the last 120 min of the 240-min recovery period. The women also had slight $[AVP]_{pl}$ elevations relative to baseline throughout recovery which may in part explain their greater water reabsorption. In addition, the women had a more rapid recovery of $[ALDO]_s$ which was consistent with their lower sodium reabsorption. The sex differences in renal fluid and sodium regulatory function may have been due to oestrogen effects on fluid regulating hormones and water retention. Despite these small differences, the overall fluid balance during exercise and recovery were similar between the men and women.

Acknowledgements We would like to thank Beth Glace, Saeed Butt and Fatima Sherzad for their technical assistance.

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