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Preexercise sodium loading aids fluid balance and endurance for women exercising in the heat

Stacy T. Sims,[1] Nancy J. Rehrer,[1,2] Melanie L. Bell,[3] and James D. Cotter[1]

[1]*School of Physical Education,* [2]*Department of Human Nutrition, and* [3]*Preventive and Social Medicine, University of Otago, Dunedin, New Zealand*

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Sims ST, Rehrer NJ, Bell ML, Cotter JD. Preexercise sodium loading aids fluid balance and endurance for women exercising in the heat. *J Appl Physiol* 103: 534–541, 2007. First published April 26, 2007; doi:10.1152/japplphysiol.01203.2006.—This study was conducted during the high-hormone phase of both natural and oral contraceptive pill (OCP)-mediated menstrual cycles to determine whether preexercise ingestion of a concentrated sodium beverage would increase plasma volume (PV), reduce physiological strain, and aid endurance of moderately trained women cycling in warm conditions. Thirteen trained cyclists [peak O_2 uptake 52 ml·kg^{-1}·min^{-1} (SD 2), age 26 yr (SD 6), weight 60.8 kg (SD 5)] who were oral contraceptive users ($n = 6$) or not ($n = 7$) completed this double-blind, crossover experiment. Cyclists ingested a concentrated-sodium (High Na^+: 164 mmol Na^+/l) or low-sodium (Low Na^+: 10 mmol Na^+/l) beverage (10 ml/kg) before cycling to exhaustion at 70% Peak O_2 uptake in warm conditions (32°C, 50% relative humidity, air velocity 4.5 m/s). Beverage (~628 ml) was ingested in seven portions across 60 min beginning 105 min before exercise, with no additional fluid given until the end of the trial. Trials were separated by one to two menstrual cycles. High Na^+ increased PV (calculated from hematocrit and hemoglobin concentration) before exercise, whereas Low Na^+ did not [−4.4 (SD 1.1) vs. −1.9% (SD 1.3); 95% confidence interval: for the difference 5.20, 6.92; $P < 0.0001$], and it involved greater time to exhaustion [98.8 (SD 25.6) vs. 78.7 (SD 24.6) min; 95% confidence interval: 13.3, 26.8; $P < 0.0001$]. Core temperature rose more quickly with Low Na^+ [1.6°C/h (SD 0.2)] than High Na^+ [1.2°C/h (SD 0.2); $P = 0.04$]. Plasma [AVP], [Na^+] concentration, and osmolality, and urine volume, [Na^+], and osmolality decreased with sodium loading ($P < 0.05$) independent of pill usage. Thus preexercise ingestion of a concentrated sodium beverage increased PV, reduced thermoregulatory strain, and increased exercise capacity for women in the high-hormone phase of natural and oral contraceptive pill-mediated menstrual cycles, in warm conditions.

citrate; hypervolemia; hyperhydration; estradiol; progesterone

EXERCISE, PARTICULARLY long-lasting exercise in the heat, places large demands on the circulatory system. Concurrently, fluid and electrolyte losses and limited or inappropriate replacement during exercise can both compromise circulatory integrity via changes in pressure, hemoconcentration, and electrochemical gradients. Accordingly, preexercise hyperhydration with high-sodium fluids has been shown to decrease cardiovascular and thermal strain and enhance exercise capacity in untrained and trained men in the heat (1, 13–16, 40). Whether this would be the same in women is unknown. Women might be additionally compromised because of central and peripheral effects of female sex hormones and oral contraceptives, although our understanding of these effects during and after exercise is significantly lacking. Apart from recent studies by Charkoudian and colleagues (5–7, 18) investigating the effects of short-term oral contraceptive pill (OCP) habituation on vasodilation and heat responses in women, most research on female sex hormones and renal function has been conducted with monophasic OCPs or acute infusions of estrogen or progesterone, and the studies have primarily been in untrained women (2, 17, 20, 21, 31–36). Previous research (31–36) has not evaluated any chronic adaptations, nor has it addressed the moderation of hormone concentrations that occurs in athletically trained women.

The plasma volume (PV) in women is highly influenced by oestrogen and progesterone, and there is still limited research on the effects of the chronic perturbations of menstrual cycle hormones on water and sodium handling. Estrogen and progesterone can have profound effects on fluid dynamics, in particular the Starling forces that regulate fluid movement between the vascular and interstitial spaces altering PV as the concentrations of estrogen and progesterone change (20, 28, 31–36). Because fluid and electrolyte balance are critical for normal cellular function and maintaining adequate blood and PV, understanding the interactions of female sex hormones and the fluid regulatory system is crucial. PV is highest during the preovulatory phase of the menstrual cycle, when estrogen levels are increasing. However, PV falls by as much as 8% during the midluteal phase of the menstrual cycle due to fluid and protein movement into the interstitium (28, 34) from the effects of estrogen on plasma albumin and plasma colloid osmotic pressure (28). Women face additional challenges in the heat because of the perturbations of the female sex hormones and the influences of these hormones on fluid distribution, resting core temperature, and resetting of the thermoregulatory responses. For example in the high-hormone phase of either the endogenously or exogenously driven menstrual cycle, baseline core temperature is reset to a higher internal temperature; with this a concomitant increase in the threshold for sweat onset as well as a decreased sensitivity of cutaneous vasodilation (4–6, 17, 18, 22, 29, 37–39). With the reset of temperature thresholds (i.e., sweat onset, vasodilation, and decreased baroreflex sensitivity), the time to critical core temperature may be reduced. These aspects of altered thermoregulatory control, may compromise women's exercise capacity in the heat during the high-hormone phase of their menstrual cycle.

PV maintenance can be important for exercise performance, especially in the heat (1). A maintained or increased PV

Address for reprint requests and other correspondence: S. T. Sims, Sports Medicine-Human Performance Laboratory, Arrillaga Sports Center-Lower Level, Stanford Univ., Stanford, CA 94305-6150 (e-mail: stsims@stanford.edu).

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reduces the viscosity of blood, increases heat storage and transfer capacity, and increases exercising cardiac output and maximal O_2 uptake, at least in untrained individuals (15, 16, 25). The ability to offload heat, by virtue of the increased volume of circulating fluid, is also increased as circulation to the periphery is enhanced. However, fluid balance is often not achieved; indeed it is not even achievable when fluid availability or gastric emptying rates prevent the rates of ingestion and absorption from matching sweat rates (23). One method of inducing hyperhydration and hypervolemia, originally developed to help offset effects of PV loss in microgravity (3, 16), is "sodium loading." A sodium-concentrated beverage composed of sodium citrate and sodium chloride (164 mmol Na^+/l), with moderate osmolality (253 mosmol/kgH_2O), has been shown to expand PV, reduce cardiovascular strain, and aid exercise capacity of untrained and trained men in temperate and hot environments (13–16). Moreover, earlier studies (3, 12) showed that ingested saline solutions between 0.9 and 1.07% expanded PV over a 4-h postingestion time period. Frey and colleagues (12) determined that the 1.07% saline solution elicited the greatest PV expansion and urine concentration over the 4 h postingestion; however, the addition of 1% glucose did not improve the effectiveness of PV expansion, but it increased diuresis. Thus the authors concluded that a slightly hypertonic saline-only solution provided the most effective means of PV expansion.

In view of the aforementioned compromised thermoregulatory factors that women experience in the high-hormonal phase of their menstrual cycle, the first aim of this study was to determine whether a similar hypervolemia-inducing protocol would be effective for women exercising in the heat. We hypothesized that preexercise ingestion of a high-sodium solution would expand PV, increase exercise endurance, lower the rate of rise of core temperature, reduce cardiovascular strain, and reduce perceived exertion during exercise in the heat. The second aim was to determine the effects of this hyperhydration strategy as a function of habituated OCP usage. We hypothesized that sodium loading would have a hypervolemic effect and benefit women's exercise capacity in the heat. Additionally, because of the secondary effects of estrogen fluid balance, specifically that estrogen lowers the osmotic threshold for arginine vasopressin (AVP) release with a concomitant increase in sodium and fluid retention, and the greater concentration of estrogen associated with OCPs, we hypothesized that the hypervolemic effects of sodium loading would be attenuated with chronic OCP usage.

METHODS

Participants. Thirteen healthy, eumenorrheic, endurance-trained female cyclists [peak O_2 uptake ($\dot{V}O_{2\,peak}$) 52 ml·kg^{-1}·min^{-1} (SD 2); age 26 yr (SD 6), weight 60.8 kg (SD 5)], with no history of cardiovascular or renal disease, no contraindications to OCPs, and not taking other medication, were recruited. Participants were separated into natural menstrual cycle (Nat, $n = 7$) or triphasic OCP-mediated (OCP, $n = 6$) groups according to their usage status. Before participation, all participants were informed of the nature and demands of the investigation, after which they gave written informed consent. The study was approved by the Human Ethics Committee of the University of Otago. Of the 13 participants, 12 were familiar with the experimental protocols and conditions, and all had participated in previous laboratory trials. The naive participant was familiarized with the experimental testing procedures 2 wk before her first experimental session.

Preliminary testing. Before performance testing, all participants underwent an incremental cycling test to exhaustion to ensure that they met the criteria for being categorized as "well trained" [train ≥5 times/wk and have peak aerobic power ($\dot{V}O_{2\,peak}$) >50 ml O_2·kg body mass^{-1}·min^{-1}]. The $\dot{V}O_{2\,peak}$ test was conducted on a separate day, in temperate conditions (19 ± 2°C), 1–2 wk before experimental testing, and was used to set the intensity for the experimental trials. This comprised a progressive incremental test to exhaustion undertaken on a computer-controlled, precision electromagnetically braked bicycle ergometer (Velotron, Racermate, Seattle, WA) during which O_2 consumption ($\dot{V}O_2$), CO_2 production ($\dot{V}CO_2$), and heart rate (HR) were measured. After a 5- to 10-min warm-up at a self-selected intensity, the incremental test commenced at a workload of 2.25 W/kg, which increased by 40 W after each 150 s until the participant's HR reached 85% of the age-predicted maximum, after which the workload increased 25 W each 150 s until interval until the subject could no longer maintain the required power output and/or the pedaling frequency dropped below 75 revolutions/min. During the $\dot{V}O_{2\,peak}$ test, as well as during subsequently described experimental trials, HR was recorded using a HR monitor (PE 4000, Polar Electro, Kempele, Finland), and $\dot{V}CO_2$, $\dot{V}O_2$, and ventilation were measured on a breath-by-breath basis using an automated respiratory gas analysis system (Cortex Metalyser 3B, Borsdorf, Leipzig, Germany).

Experimental testing. A double-blind, placebo-controlled, crossover design was employed during the winter months of the southern hemisphere (June to September) to control for heat acclimatization. Experimental trials occurred on *days 20* or *21* for an endogenous 28-day cycle and on *days 18–20* (end of the third week of active pills/high-hormone phase) for a triphasic OCP cycle. Participants were asked to self-monitor menstrual cycle by recording the brand name and cycle of the pill or by recording basal temperature. Day of testing varied from participant to participant because of normal fluctuations in individual cycle length (30). The order of testing the two sodium loads was counterbalanced and randomized. Participants completed two cycling trials to exhaustion at 70% of their temperate-environment $\dot{V}O_2$ in the heat (32°C, 50% relative humidity, air velocity ~4.5 m/s) The performance trial was conducted following ingestion of 10 ml/kg body mass of a control, low-sodium beverage (Low Na^+; 10 mmol Na^+/l, 43 mosmol/kgH_2O) or a concentrated-sodium beverage (High Na^+; 164 mmol Na^+/l, 253 mosmol/kgH_2O) (14–16), chilled overnight to 4°C. Beverage was blind to the researcher conducting trials and to the participant. To minimize order effects, the allocation of the first beverage assignment was random for the first participant and thereafter was alternated.

Standardization. All women had regular menstrual cycles. For those women on the triphasic OCP, they had been following the same schedule of pills for at least 6 mo before testing. To verify phase of the menstrual cycle, plasma estrogen and progesterone concentrations were assessed from the baseline sample before the sodium-loading protocol was undertaken. To standardize the training effects on PV, each participant maintained a training diary of duration, mode, and intensity of activity, and they replicated this for consistency throughout the weeks preceding each trial. Participants completed a standardized 60-min exercise bout at 50% $\dot{V}O_{2\,peak}$, in the laboratory, 24 h before each testing day, with no further exercise until experimental testing. Participants refrained from consuming soy-based products (because of phytoestrogen effects) and nonsteroidal anti-inflammatory drugs throughout the experimental period. The same meal (low-moderate sodium and no alcohol, soy, or caffeine permitted) was consumed the evening before each testing session, and the participant drank 750 ml of water throughout that evening before bed. An additional 500 ml of water was consumed between breakfast and the start of the testing protocol. On the day of testing, the participant reported to the laboratory, fully hydrated, after a standardized breakfast (provided; 13 g protein, 10 g fat, 63 g carbohydrate, 1,680 kJ, 265

mg Na^+), which was completely consumed between 2.5 and 2 h before experimentation. Urinary specific gravity (USG) was measured in an initial baseline sample to verify similar hydration status in all tests.

Experimental session. On arrival at 0900, each participant voided her bladder before body weight was determined (± 10 g). The participant was then seated for catheter placement (22-gauge Teflon intravenous catheter) in the antecubital or other suitable forearm vein of the left arm. On withdrawal of the baseline samples (without stasis), the participant began 1 h of Low Na^+ or High Na^+ ingestion of 10 ml/kg body mass, given in seven equal portions, every 10 min with a 2-min walk every 20 min. Low Na^+ consisted of only of sodium chloride (0.58 g/l), whereas High Na^+ was composed only of sodium citrate (7.72 g/l) and sodium chloride (4.5 g/l), as developed by Greenleaf et al. (14–16) as AstroAde.

On complete ingestion of the beverage, the participant remained seated during which time skin thermistors were attached. Blood sampling was conducted 15 min after drinking, with urine sampling and core thermistor insertion immediately following (Fig. 1). Twenty minutes after drinking, each participant entered a climatically controlled chamber (32°C, 50% relative humidity, air velocity ~ 4.5 m/s) and commenced cycling on a cycling ergometer (Velotron, Racermate), fitted to her usual cycling position, without fluid or feedback cues of time or distance. The power output (watts) was set to elicit 70% of her temperate-environment $\dot{V}O_{2\,peak}$. Exercise stopped when the participant could no longer maintain exercise at the given intensity or when rectal temperature reached 39.5°C. HR was recorded at 1-min intervals throughout exercise. Rectal temperature was measured with a disposable thermistor (Thermistor 400, Mallinckrodt, St. Louis, MO), which was disinfected and reused within each participant. Skin temperatures were measured with insulated thermistors (Type EU, Grant Instrument, Cambridge, UK) taped below the head of the thermistor at four sites: biceps, calf, chest, and thigh, from which mean skin temperature was calculated (41). Temperatures were recorded at 1-min intervals (model 1200, Grant Instrument). $\dot{V}CO_2$, $\dot{V}O_2$, and ventilation were measured for 2 min initially at 5-min intervals until 30 min into exercise, and thereafter they were measured at 15-min intervals. Urine was collected at baseline, 15 min after drinking, immediately before exercise, and at exhaustion. Blood samples were at baseline, 15 min after drinking, 10 min into exercise, and at exhaustion. Urine samples were taken 2 min after blood sampling, except during exercise (Fig. 1).

Blood and urine analysis. All blood samples (17 ml each) were separated into aliquots for analysis of hematocrit (Hct), hemoglobin concentration ([Hb]), plasma sodium concentration ([Na^+]), plasma osmolality, plasma creatinine concentration ([Cr]), and plasma AVP concentration [AVP]. A 2-ml aliquot was analyzed immediately for [Hb] (Hemoximeter, OSM3 Radiometer, Copenhagen) and Hct in quadruplicate. Blood for Hct was drawn into capillary tubes and centrifuged for 6 min at 3,000 rpm (Hawklsey Microcentrifuge, Sussex, UK) and read using a modified microcapillary tube reader (Damon/IEC Division, Needham Heights, MA); the measurement error was ±0.25%. One of the remaining three additional aliquots of blood (5 ml each) was transferred into one tube containing heparin for plasma osmolality determination. The last two aliquots were transferred into tubes containing EDTA for determination of plasma [AVP], plasma [Cr], plasma estrogen concentration ([E_2]), and progesterone and progestins concentration ([P_4]). All three tubes were then centrifuged for 10 min at 6°C and 3,000 revolutions/min (model GS-15R Centrifuge, Beckman-Coulter, Fullerton, CA).

Plasma osmolality was measured using vapor point depression (Osmometer, model Vapro5520, Wescor, Logan, UT). Urine [Na^+] and plasma [Na^+] were measured using an indirect ion-specific electrode analyzer (Aeroset/c8000, Otago, Southern Community Laboratories, Dunedin, NZ) in triplicate. One-third of urine aliquots were frozen at −80°C until analysis for [Na^+] was conducted. USG was measured in triplicate with a hand refractometer (ATAGO, Tokyo, Japan). Because analyses for hormones and electrolytes were not performed immediately, plasma and urine were put on ice and stored at −80°C until analyses.

Plasma [AVP], [E_2], and [P_4] were analyzed at a local medical laboratory (Southern Community Laboratories) utilizing the radioimmunoassay technique. The plasma [AVP] was measured using an "in-house" antiserum with synthetic AVP (Ferring or Sigma) used for standards and preparation of ^{125}I-labeled AVP using chloramine T and purification by HPLC. Samples were extracted with acetonitrile (1:2 acetonitrile-plasma), and the supernatant was dried down before the assay. A 3-day preincubation step at 4°C is followed by a further 3 days at 4°C after the addition of the ^{125}I-AVP. Separation of bound and free hormone is by polyethylene glycol plus-globulin precipitation. The plasma [E_2] was measured via Sorin (sensitive) RIA kit (Sorin Biomedica Diagnostics, Saluggia, Italy), whereas plasma [P_4] was determined using the commercial conjugate-antibody method (ELISA kit, Assay Designs, Ann Arbor, MI) with sample concentrations determined against a standard curve. Intra- and interassay coefficients of variation, respectively, for the midrange standards were the following: for plasma [AVP], (23.2 pg/ml) 5.1 and 9.8%; for plasma [E_2] (115 pg/ml) 2.8 and 6.0%; and for plasma [P_4] (1.5 ng/ml) 3.6% and 4.8%.

Calculations. Changes in PV from baseline were estimated from changes in Hct and [Hb] using the following equation (9):

$$\%\Delta PV = 100\,\{(Hb_0/Hb_t) * [(1 - Hct_t)/(1 - Hct_0)]\} - 100$$

in which subscripts *t* and *0* denote measurements at time *t* and at baseline (−90 min), respectively. Hb is in grams per 100 milliliters, and Hct is a fraction. Hct was multiplied by 0.96 and then 0.91 to

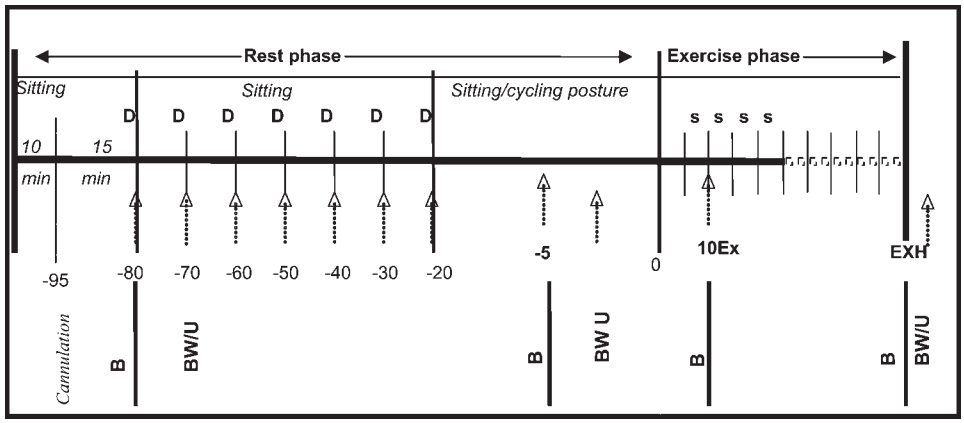

Fig. 1. Time line of experimental sessions. D, drinking; B, blood sampling; BW/U, body mass (kg) and urine sampling (volume, osmolality); S, O_2 consumption and CO_2 production for rating of perceived exertion sampling; EXH, exhaustion; 10Ex, 10 min of exercise.

correct for trapped plasma and the venous-to-whole blood Hct excess, respectively.

Statistical analysis. Mixed models were used to model each of the outcomes as a function of time and beverage. The repeated-measures nature of the study design was taken into account by using the covariance structures available in the SAS (version 9.1) procedure MIXED. Measurements were assumed to be more highly correlated within, as opposed to between, participants and also within, vs. between, treatments within a participant. Treatment differences in least squares means for each of the outcomes at each time point were calculated from the model. Because each time point was tested, a means model (entering time as a categorical variable in the model) was used, except when hypotheses were specifically about response slopes with respect to time. End time (exhaustion), E_2, and P_4 were measured only once and were tested using paired *t*-tests. Sample size was determined from a post hoc power calculation based on pilot work and the outcomes of our women's rest study; with an alpha level of 0.05 and a sample size of six participants per group, the beta level (power) was ≥0.80 for detecting effect sizes. Differences in least squares means for each of the outcomes and 95% confidence intervals (95% CI) were calculated from the model and were considered statistically significant at $P < 0.05$. Values are given as means (SD).

RESULTS

Participant compliance. To ensure participant compliance, training, diet, temperature (Nat), and pill usage (OCP) logs were collected and reviewed on the morning of the second experimental session. Additionally, hydration compliance was checked via USG. A participant with USG ≤ 1.020 was considered euhydrated. All participants completed all documentation and standardization procedures requested. Participant baseline characteristics are reported in Table 1.

Acute PV changes, plasma [Na^+], plasma osmolality, and plasma [AVP]. There was significantly greater PV expansion with the High Na^+ than with the Low Na^+ [−4.4 (SD 1.1) vs. −1.9% (SD 1.3); $P = 0.0001$; 95% CI for the difference: 5.2, 6.9; Fig. 2]. This occurred 15 min postdrinking. There was no evidence of a difference between OCP and natural (Nat) in the high-hormone phase in PV change (Δ) with either High Na^+ [Δ4.0 (SD 1.2) vs. Δ4.3 (SD 1.4) ml for OCP vs. Nat, respectively; $P = 0.23$] or Low Na^+ [Δ−2.03 (SD 1.5) vs. Δ−1.84 (SD 1.3) ml for OCP vs. Nat respectively; $P = 0.47$].

Plasma [Na^+] rose to less extent in High Na^+ than in Low Na^+ [Δ1.8 (SD 0.6) vs. Δ3.1 (SD 0.4) mmol/l; $P = 0.03$; 95% CI for the difference: 0.16, 2.44], and it rose less rapidly (slope 0.70 mmol/h; 95% CI for the difference: 1.26, 0.15; $P = 0.01$). However, no significant differences were observed between OCP and Nat groups [Δ1.9 (0.7) vs. Δ1.6 (0.8) mmol/l for High Na^+ OCP vs. Nat, respectively ($P = 0.683$); Δ3.2 (0.3) vs. Δ2.8 (0.6) mmol/l for Low Na^+ OCP vs. Nat, respectively ($P = 0.562$)]. Consistent with the plasma Na^+ responses, plasma osmolality rose to less extent in High Na^+ than in Low Na^+ (Δ10.7 vs. Δ15.5 mosmol/kgH_2O for High Na^+ vs. Low Na^+, respectively; $P < 0.0001$; 95% CI for the difference: 2.9, 6.7 mosmol/kgH_2O), and they responded similarly in OCP and Nat (Fig. 3*A*). Additionally, a positive correlation was observed between the change (from baseline) of [AVP] and plasma osmolality rise (OCP High Na^+: $r = 0.94$, Low Na^+: $r = 0.93$; Nat High Na^+: $r = 0.95$, Low Na^+: $r = 0.94$ Fig. 3*B*).

Plasma [AVP] was similar between beverages over the drinking period [1.4 (SD 0.4) vs. 1.3 (SD 3.1) pg/ml; 95% CI for the difference: −1.0, 0.5 pg/ml; $P = 0.575$], but the rise across exercise was smaller in High Na^+ than in Low Na^+ [Δ5.2 (SD 6.2) vs. 7.1 (SD 5.0) pg/ml; 95% CI for the difference: −3.0, −1.5 pg/ml; $P < 0.0001$] and occurred more slowly (slope: 2.4 vs. 3.1 pg·ml^{-1}·h^{-1}; $P = 0.001$; 95% CI: 1.2, 3.7). The OCP and Nat groups were similar with regard to plasma [AVP] responses (Fig. 4); plasma [AVP] rose more rapidly during exercise with Low Na^+ than with High Na^+, independently of OCP usage.

Exercise tolerance and thermoregulatory responses. Individual performance and temperature data are presented in Tables 2 and 3, respectively. Time to exhaustion was longer

Table 1. *Baseline subject characteristics*

	Triphasic OCP (High Hormone)	Natural 28-Day Cycle (Late Luteal)
Age, yr	24.0 (5.0)	29.0 (6.0)
Body mass, kg	61.9 (5.2)	62.7 (4.8)
Height, cm	160.6 (6.0)	165.2 (5.0)
$\dot{V}O_{2\ peak}$, ml·kg^{-1}·min^{-1}	53.1 (2.5)	51.2 (1.8)

	Beverage			
	High Na^+	Low Na^+	High Na^+	Low Na^+
Menstrual cycle, day of test	19 (2)	19 (2)	23 (2)	23 (2)
Plasma$_{[E_2]}$, pg/ml	1.4 (1.9)	1.5 (1.8)	231.2 (65.8)	238.7 (92.8)
Plasma$_{[P_4]}$, ng/ml	1.6 (0.5)	1.7 (0.5)	5.1 (1.1)	5.0 (1.2)
Plasma$_{[AVP]}$, pg/ml	1.8 (0.4)*	1.5 (0.5)*	1.2 (0.2)	1.1 (0.1)
Plasma osmolality, mosmol/kgH_2O	286.3 (3.6)	286.2 (3.3)	287.8 (3.7)	287.8 (4.2)
Urine osmolality, mosmol/kgH_2O	221.5 (65.9)	192.5 (82.8)	236.6 (57.7)	240.0 (95.7)
Plasma$_{[Na^+]}$, meq/l	141.7 (1.8)	141.0 (1.6)	142.3 (1.6)	141.8 (1.7)
Hematocrit, %	38.7 (1.6)	38.5 (1.3)	40.1 (1.7)	39.9 (1.6)
Hemoglobin, g/l	138.2 (4.0)	138.3 (3.7)	139.1 (5.5)	139.6 (6.0)
MCHC, g/100 ml	37.2 (0.6)	37.4 (0.7)	36.1 (0.8)	36.4 (0.9)
MCV, fl	92.1 (2.4)	92.0 (2.3)	91.7 (2.1)	91.9 (2.2)

Values are means (SD). OCP, oral contraceptive pill; $\dot{V}O_{2\ peak}$, peak O_2 uptake; [E_2], estradiol concentration; [P_4], progesterone and progestins concentration; [AVP], arginine vasopressin concentration; MCHC, mean corpuscular hemoglobin concentration; MCV, mean corpuscular volume; High Na^+, concentrated sodium; Low Na^+, low Na^+. *$P < 0.05$ between OCP and Natural.

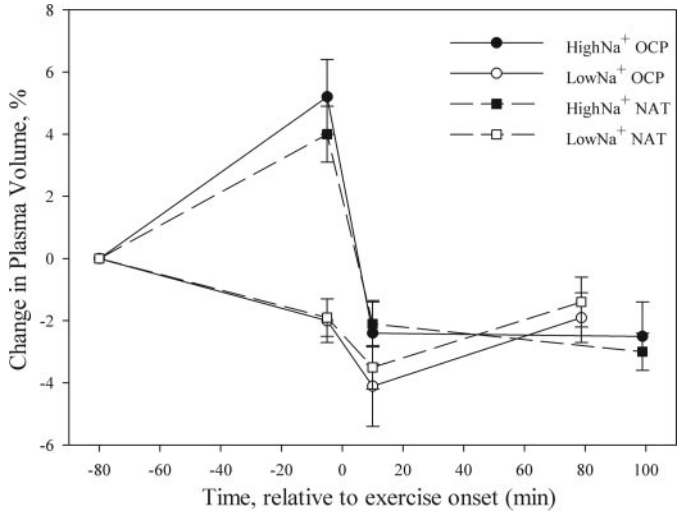

Fig. 2. Plasma volume responses at rest and during exercise after ingestion of sodium-concentrated (High Na^+) and sodium-dilute (Low Na^+) beverages. Data are means (SD) of triphasic oral contraceptive pill (OCP)-regulated and natural menstrual cycle (Nat) for both beverages.

with the High Na^+ beverage for all participants [mean: 98.8 (SD 25.6) vs. 78.7 (SD 24.6) min; 95% CI for the difference: 13.3, 26.8; $P < 0.0001$]. Time to exhaustion was similar between OCP and Nat with both High Na^+ [100.3 (SD 24.2) vs. 96.3 (SD 25.4) min; $P = 0.482$] and Low Na^+ [82.1 (SD 24.2) vs. 77.6 (SD 26.3) min; $P = 0.587$]. Independent of pill usage, all participants ceased exercise because of volitional exhaustion, not core temperature restrictions, in the High Na^+ with the longer time to exhaustion. Core temperature was higher at rest in OCP than in Nat for both beverage conditions (High Na^+: 95% CI for the difference: 0.05, 0.33, $P = 0.05$; Low Na^+: 95% CI for the difference: 0.10, 0.36, $P = 0.05$), but it rose similarly during exercise in both groups (Fig. 5). Rate of core temperature rise was greater during exercise with the Low Na^+ [1.6°C/h (SD 0.2)] than with the High Na^+ [1.2°C/h (SD 0.2); $P = 0.04$] with no significant difference between OCP and Nat. Mean skin temperature increased over time in both trials ($P < 0.00$), and it was similar between both beverages ($P = 0.85$).

Average HR was lower during exercise with High Na^+ than with Low Na^+ [156 (SD 18) vs. 165 (SD 16) beats/min, respectively; $P = 0.04$], with a slower rate of rise observed during exercise with High Na^+ than with Low Na^+ (slope 0.39 vs. 0.45 beats·min·h^{-1}, respectively; $P < 0.005$). Mean rating of perceived exertion was lower in High Na^+ than in Low Na^+ [16 (SD 2) vs. 18 (SD 1), respectively; $P = 0.05$]. There were no significant differences between OCP and Nat groups for either variable.

Fluid balance. Following ingestion of ~628 ml (SD 40) of fluid before exercise in each beverage condition, ~330 ml (SD 70) more urine was produced during the Low Na^+ trial than during the High Na^+ trial ($P = 0.03$) independent of OCP usage. Mean sweat rate as calculated from rate of body mass loss was also greater with Low Na^+ (Table 4). Urine osmolality was higher at the end of exercise with Low Na^+ than with High Na^+ (95% CI for the difference −233.06, −15.23 mosmol/kgH_2O; $P = 0.03$) at the end of exercise between trials, despite exercise being 20% longer in High Na^+ (Fig. 6).

DISCUSSION

This study is novel in that it is the first to investigate the effects of an acute sodium load on exercise tolerance and physiological strain of endurance-trained female cyclists. Moreover, the high hormone phase of both oral contraceptive pill and natural menstrual cycles was examined because this is the phase of the menstrual cycle in which women are most compromised physiologically with regard to fluid balance and thermoregulatory capacity and exercise performance in the heat (29, 30, 38). In line with our laboratory's previous findings for men running in the heat (40), sodium loading led to a modest fluid (water and sodium) retention and hypervolemia and to reduced thermoregulatory strain and greater endurance during subsequent exercise. These effects were independent of OCP usage, although women using OCPs had a slightly higher resting core temperature.

As the core body temperature rises, and increased demand for heat dissipation ensues; the PV decrease that accompanies sweat-related hypohydration may compromise work capacity

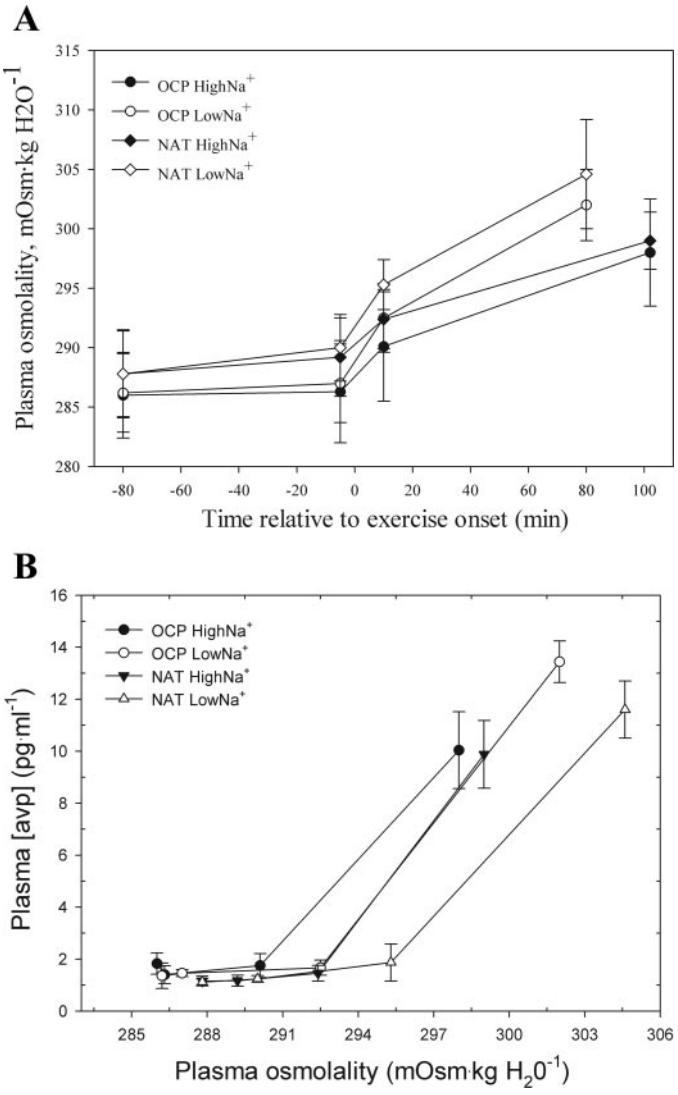

Fig. 3. *A*: change in plasma osmolality, between groups and beverages, over time. *B*: plasma arginine vasopressin concentration ([AVP]) as a function of plasma osmolality. Data are means (SD) of OCP and Nat for both beverages.

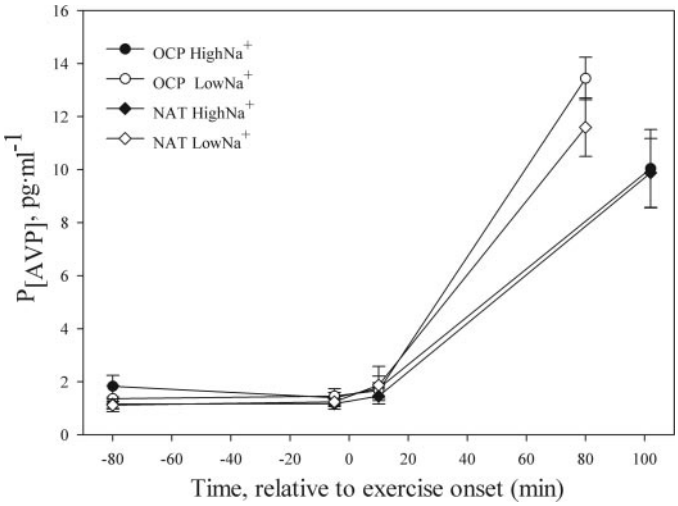

Fig. 4. Changes in [AVP] between beverages. Data are means (SD) of OCP and Nat for both beverages.

(19, 25, 27). Blood flow to the muscles must be maintained at a high level during exercise to support metabolism, but augmented blood flow to the skin is also needed to transfer heat to the body surface where it can be dissipated. Hyperthermia appears to be the critical determinant of exercise capacity in the heat with emphasis on implementing strategies to minimize the rise in core temperature during exercise (25–27). In women, reproductive hormones affect fluid balance and can affect thermoregulatory control during exercise (6, 37–39). Core temperature is elevated during the high-hormone phase of both the OCP and Nat menstrual cycles, with difference of up to 0.6°C, both at rest and during exercise in temperate environments, and this is maintained in hot environments (21, 29, 33, 37). Our data demonstrate a higher baseline core temperature in those on OCPs; however, the rate of core temperature rise was similar in both groups. This observation supports the observation of Charkoudian and colleagues (4–6, 18) that the thermogenic effect of progestins and their inhibition of cutaneous vasodilation may reset baseline core temperature upward with chronic use of oral contraceptives.

Table 2. *Individual performance times*

Participant	Watts	High Na^+ End, min	Low Na^+ End, min	ΔTime, min	Difference, %
*A**	150	117.5	80.3	+ 37.2	31.6
*B**	160	116.7	110.3	+ 6.3	5.4
*C**	155	97.2	80.0	+ 17.2	17.7
*D**	170	127.1	100.3	+ 27.2	21.4
*E**	145	58.5	40.2	+ 18.3	31.2
*F**	155	100.2	82.5	+ 17.7	17.7
H	145	55.5	46.5	+ 9.0	16.2
I	140	102.1	88.1	+ 14.0	13.7
J	145	93.2	65.3	+ 27.8	29.9
K	145	113.3	65.3	+ 48.0	42.3
L	205	109.0	93.3	+ 15.7	14.4
M	140	67.1	50.1	+ 17.1	25.5
N	155	126.9	120.5	+ 6.4	5.0
Mean	155	98.8	78.7	+ 30.7	20.9
SD	17	24.4	23.0	5.6	8.8

End, time to exhaustion; Watts, watts sustained during exercise for both trials; Δ, change. *OCP participant.

Table 3. *Individual reasons for exercise termination and core temperature at Exh*

		High Na^+		Low Na^+	
Participant	Watts	End, °C	Reason	End, °C	Reason
*A**	150	39.10	Exh	38.70	Exh
*B**	160	39.05	Exh	39.50	Rec
*C**	155	39.10	Exh	38.75	Exh
*D**	170	39.10	Exh	38.25	Exh
*E**	145	38.85	Exh	39.20	Exh
*F**	155	39.10	Exh	39.15	Exh
H	145	39.05	Exh	39.40	Exh
I	140	39.10	Exh	39.10	Exh
J	145	38.65	Exh	38.95	Exh
K	145	39.20	Exh	38.95	Exh
L	205	38.90	Exh	39.40	Exh
M	140	39.10	Exh	39.40	Exh
N	155	39.15	Exh	39.25	Exh
Mean	155	39.00		39.20	
SD	17	0.10		0.40	

Exh, volitional exhaustion; Rec, ethical restriction end point of 39.5°C. *OCP participant.

In the present study, as in the study on men (40), sodium loading was evaluated as a means of prehydration to reduce cardiac and thermoregulatory strain associated with exercise in the heat. One benefit of the sodium loading and increased fluid for redistribution is the slower rise in rectal temperature, as determined in both studies. In the men's study, the slope of rise in the High Na^+ trials was 0.32°C/h compared with 0.37°C/h in the Low Na^+ trial; with the women, it was 0.21°C/h in the High Na^+ compared with 0.29°C/h in the Low Na^+ trials. Increased body temperature increases sinoatrial node depolarization rate directly and indirectly (via catecholamines) and thus increases HR. In the men's study, HR averaged 157 beats/min during exercise in High Na^+ and 161 beats/min in Low Na^+, with the average cardiovascular drift in Low Na^+ being twice that in High Na^+(0.44 vs. 0.22 beats/min), and the time-matched final HR higher. The women demonstrated similar cardiovascular responses. The average HR during exercise in the High Na^+ for the women was significantly lower (156 vs. 165 beats/min) in the Low Na^+ and they also experienced

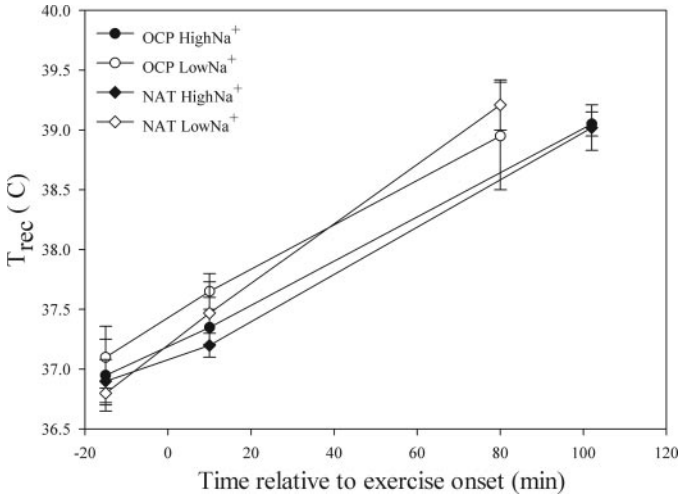

Fig. 5. Change in core temperature [rectal temperature (T_{rec})] during exercise. Data are means (SD) of OCP and Nat for both beverages.

Table 4. *Fluid balance*

	Triphasic OCP (High Hormone)		Natural 28-Day Cycle (Late Luteal)	
	High Na^+	Low Na^+	High Na^+	Low Na^+
During total experimental session				
Total fluid intake, ml	627.7 (28.2)		629.7 (45.6)	
Total urine production, ml	766.2 (212.4)*	1079.7 (230.1)	837.1 (205.5)*	1192.1 (214.2)
Body mass, kg				
Baseline (−105)	61.9 (5.2)	62.0 (4.7)	62.7 (4.8)	62.6 (4.6)
−5	62.5 (5.2)	62.6 (4.8)	63.6 (4.4)	63.3 (4.6)
End	60.2 (5.7)	60.3 (4.6)	61.3 (4.6)	61.1 (4.1)
During exercise duration				
Rate of sweat loss, l/h	1.34 (0.25)*	1.69 (0.39)	1.45 (0.31)*	1.59 (0.40)
Rate of change in body mass, %/h	0.83 (0.11)*	0.91 (0.73)	0.92 (0.17)*	1.01 (0.30)

Values are means (SD). −105 and −5, min relative to exercise onset; End, time of exhaustion, end of exercise. *$P < 0.05$ between High Na^+ and Low Na^+.

a higher time-matched final HR. With the Low Na^+, an earlier onset of performance-inhibiting hypohydration may have contributed to the cardiovascular responses. This is likely because the increased cutaneous dilation and compliance that occur with displacement of blood to the skin for cooling results in an inability to sustain adequate cardiac output, blood pressure, and perfusion to the brain (7, 13, 19, 25, 27).

Montain and Coyle (25) demonstrated that the ingestion of fluid during exercise increases skin blood flow, and therefore thermoregulatory capacity, independent of increases in the circulating blood volume. In the present investigation, no additional fluid was consumed during the exercise trials; thus the physiological strain of exercise in the heat was not attenuated by fluid intake during the exercise trials. Subsequently, the attenuation of physiological strain cannot be attributed to a PV/blood volume expansion. The interaction between thermoregulation and osmoregulation becomes an important issue here. In the present investigation and in the study on men (40), attenuation of plasma osmolality and plasma sodium was observed. Because an increase in osmolality occurs in heat-stress conditions, osmotic inhibitory input to the thermoregulatory center decreases thermoregulatory responses as well as increases osmotic AVP secretion and thirst (7). Moreover, a rise in plasma osmolality increases inhibitory input to the thermoregulatory center, thus reducing the body's ability to thermoregulate (10, 11, 19). In both the men's and women's exercise studies, a slower rate of change of plasma osmolality was observed with the High Na^+ compared with the Low Na^+ trials.

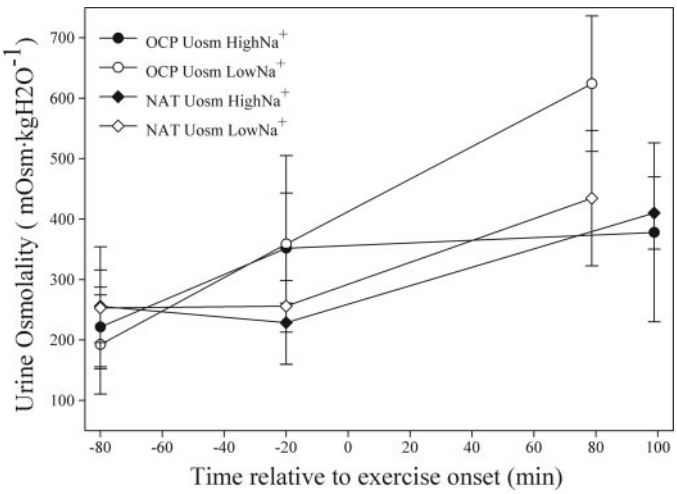

Fig. 6. Changes in urine osmolality between beverages. Data are means (SD) of OCP and Nat for both beverages.

In the present investigation, a slower AVP response to dehydrating exercise occurred in the High Na^+ trials. This attenuated response of AVP may be due to the modification of AVP sensitivity that occurs in the luteal phase, or more likely it is due to a reduced plasma osmolality across the exercise duration of High Na^+. The results of the present study agree with those of previous researchers (7, 10, 11, 19, 27) in that hyperosmolality appears to be an important factor in the regulation of AVP release, observed here with the lower rate of change of plasma sodium and osmolality concomitant with the attenuation of AVP response in High Na^+.

A greater time to exercise termination and a reduced perceived exertion indicate an increased exercise capacity with sodium loading in women during the high-hormone phase of the menstrual cycle. It appears that the increased fluid retention decreased cardiac strain and attenuated plasma osmolality and the rate of core temperature rise when no fluid was given during exhaustive cycling exercise in warm conditions. This can be attributed to the reduction of osmotic inhibitory input to the thermoregulatory system. The high-hormone phase of the menstrual cycle, whether natural or OCP regulated, is the phase in which women are most compromised for thermoregulation and fluid balance. Although exogenous female sex hormones alter cutaneous vasodilation, increase resting core temperature, and adversely affect fluid regulatory hormones, the results of the present investigation do not indicate that chronic use of OCPs further compromises women. No other differences were observed between the Nat and the OCP groups, aside from an increased resting core temperature in the OCP group, with regard to the responses to preexercise sodium loading and the subsequent increased exercise capacity. Further research is needed to determine whether these performance enhancing effects of preexercise sodium loading still occur when additional fluid is consumed during exercise.

Final conclusions. Drinking fluids with a higher sodium concentration than in regular sports drinks, before exercise, can elicit a transient hypovolemic response that is partly preserved (relative to a low-sodium drink) in exercise and is associated with improved physiological status and exercise capacity in warm conditions in trained men and women. Practical implications of these findings for female endurance athletes may be

to try to adjust their competition schedule to their menstrual cycle as well as use a preexercise sodium-loading protocol before competition in the heat. However, sporting events and competition occurs regardless of menstrual cycle phase. The results of the present studies indicate that cardiac strain decreases and exercise capacity increases in the luteal phase using sodium loading. It is, however, unclear whether cardiovascular and/or thermoregulatory mechanisms, or resultant mechanisms, are responsible for this. The performance enhancement following sodium loading found in this study was achieved with no further fluid ingestion during exercise. Rehydration behaviors in some real-life exercise situations are such that considerable hypohydration still develops, even if fluid is available during exercise.

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GRANTS

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REFERENCES

1. **Berger NJA, Campbell IT, Wilkerson DP, Jones AM.** Influence of acute plasma volume expansion on $\dot{V}O_2$ kinetics, $\dot{V}O_{2\,peak}$, and performance during high-intensity cycle exercise. *J Appl Physiol* 101: 707–714, 2006.
2. **Bisson DL, Dunster GD, O'Hare JP, Hampton D, Penney MD.** Renal sodium retention does not occur during the luteal phase of the menstrual cycle in normal women. *Br J Obstet Gynaecol* 99: 247–252, 1992.
3. **Bungo MW, Charles JB, Johnson PC.** Cardiovascular deconditioning during spaceflight and the use of saline as a countermeasure to orthostatic intolerance. *Aviat Space Environ Med* 56: 985–990, 1985.
4. **Charkoudian N, Johnson JM.** Modification of active cutaneous vasodilation by oral contraceptive hormones. *J Appl Physiol* 83: 2012–2018, 1997.
5. **Charkoudian N, Johnson JM.** Reflex control of cutaneous vasoconstrictor system is reset by exogenous female reproductive hormones. *J Appl Physiol* 87: 381–385, 1999.
6. **Charkoudian N, Stephens DP, Pirkle KC, Kosiba WA, Johnson JM.** Influence of female reproductive hormones on local thermal control of skin blood flow. *J Appl Physiol* 87: 1719–1723, 1999.
7. **Convertino VA, Keil LC, Bernauer EM, Greenleaf JE.** Plasma volume, osmolality, vasopressin, and renin activity during graded exercise in man. *J Appl Physiol* 50: 123–128, 1981.
8. **Cunningham DJ, Cabanac M.** Evidence form behavioral thermoregulatory responses of a shift in set-point temperature related to the menstrual cycle. *J Physiol (Paris)* 63: 236–238, 1971.
9. **Dill DB, Costill DL.** Calculation of percentage changes in volumes of blood, plasma, and red cells in dehydration. *J Appl Physiol* 37: 247–248, 1974.
10. **Fortney SM, Vroman NB, Beckett WS, Permutt S, LaFrance ND.** The effect of exercise hemoconcentration and hyperosmolality on exercise response. *J Appl Physiol* 65: 519–524, 1988.
11. **Fortney SM, Wenger CB, Bove JR, Nadel ER.** Effect of hyperosmolality on control of blood flow and sweating. *J Appl Physiol* 57: 1688–1695, 1984.
12. **Frey MA, Riddle J, Charles JB, Bungo MW.** Blood and urine responses to ingesting fluids of various salt and glucose concentrations. *J Clin Pharmacol* 31: 880–887, 1991.
13. **Greenleaf JE, Castle BL.** Exercise temperature regulation in man during hypohydration and hyperhydration. *J Appl Physiol* 30: 847–853, 1971.
14. **Greenleaf JE, Jackson CG, Geelen G, Keil LC, Hinghofer-Szalkay H, Whittam JH.** Plasma volume expansion with oral fluids in hypohydrated men at rest and during exercise. *Aviat Space Environ Med* 69: 837–844, 1998.
15. **Greenleaf JE, Looft-Wilson R, Wisherd JL, McKenzie MA, Jensen CD, Whittam JH.** Pre-exercise hypervolemia and cycle ergometer endurance in men. *Biol Sport* 14: 103–114, 1997.
16. **Greenleaf JE, Looft-Wilson R, Wisherd JL, Jackson CGR, Fung PP, Ertl AC, Barnes PR, Jensen CD, Whittam JH.** Hypervolemia in men from fluid ingestion at rest and during exercise. *Aviat Space Environ Med* 69: 374–386, 1998.
17. **Hiroshoren NT, Makrienko I, Edoute I, Plawner Y, Itskovitz-Eldor MM, Jacob J.** Menstrual cycle effects on the neurohumoral and autonomic nervous systems regulating the cardiovascular system. *J Clin Endocrinol Metab* 87: 1569–1575, 2002.
18. **Houghton BL, Holowatz LA, Minson CT.** Influence of progestin bioactivity on cutaneous vascular responses to passive heating. *Med Sci Sports Exerc* 37: 45–51; discussion 52, 2005.
19. **Ito T, Itoh T, Hayano T, Yamauchi K, Takamata A.** Plasma hyperosmolality augments peripheral vascular response to baroreceptor unloading during heat stress. *Am J Regul Integr Comp Physiol* 289: R432–R440, 2005.
20. **Kang AK, Duncan JA, Cattran DC, Floras JS, Lai V, Scholey JW, Miller JA.** Effect of oral contraceptives on the renin angiotensin system and renal function. *Am J Physiol Regul Integr Comp Physiol* 280: R807–R813, 2001.
21. **Kolka MA, Stephenson LA.** Effect of luteal phase elevation in core temperature on forearm blood flow during exercise. *J Appl Physiol* 82: 1079–1083, 1997.
22. **Kolka MA, Stephenson LA.** Resetting the thermoregulatory set-point by endogenous estradiol or progesterone in women. *Ann NY Acad Sci* 813: 204–206, 1997.
23. **Maughan RJ, Leiper JB, Shirreffs SM.** Factors influencing the restoration of fluid and electrolyte balance after exercise in the heat. *Br J Sports Med* 31: 175–182, 1997.
24. **Minson CT, Halliwill JR, Young TM, Joyner MJ.** Influence of the menstrual cycle on sympathetic activity, baroreflex sensitivity, and vascular transduction in young women. *Circulation* 101: 862–868, 2000.
25. **Montain SJ, Coyle EF.** Influence of graded dehydration on hyperthermia and cardiovascular drift during exercise. *J Appl Physiol* 73: 1340–1350, 1992.
26. **Moran DS, Shitzer A, Pandolf KB.** A physiological strain index to evaluate heat stress. *Am J Physiol Regul Integr Comp Physiol* 275: R129–R134, 1998.
27. **Morimoto T, Itoh T, Takamata A.** Thermoregulation and body fluid in hot environment. *Prog Brain Res* 115: 499–508, 1998.
28. **Oian P, Tollan A, Fadnes HO, Noddeland H, Maltau JM.** Transcapillary fluid dynamics during the menstrual cycle. *Am J Obstet Gynecol* 156: 952–955, 1987.
29. **Pivarnik JM, Marichal CJ, Spillman T, Morrow JR.** Menstrual cycle phase affects temperature regulation during endurance exercise. *J Appl Physiol* 72: 543–548, 1992.
30. **Redman LM, Weatherby RP.** Measuring performance during the menstrual cycle: a model using oral contraceptives. *Med Sci Sports Exerc* 36: 130–136, 2004.
31. **Stachenfeld NS, Keefe DL.** Estrogen effects on osmotic regulation of AVP and fluid balance. *Am J Physiol Endocrinol Metab* 283: E711–E721, 2002.
32. **Stachenfeld NS, Keefe DL, Palter SF.** Estrogen and progesterone effects on transcapillary fluid dynamics. *Am J Physiol Regul Integr Comp Physiol* 281: R1319–R1329, 2001.
33. **Stachenfeld NS, Silva C, Keefe DL.** Estrogen modifies the temperature effects of progesterone. *J Appl Physiol* 88: 1643–1649, 2000.
34. **Stachenfeld NS, Silva C, Keefe DL, Kokoszka CA, Nadel ER.** Effects of oral contraceptives on body fluid regulation. *J Appl Physiol* 87: 1016–1025, 1999.
35. **Stachenfeld NS, Taylor HS.** Effects of estrogen and progesterone administration on extracellular fluid. *J Appl Physiol* 96: 1011–1018, 2004.
36. **Stachenfeld NS, Taylor HS.** Progesterone increases plasma volume independent of estradiol. *J Appl Physiol* 98: 1991–1997, 2005.
37. **Stephens DP, Bennett LA, Aoki K, Kosiba WA, Charkoudian N, Johnson JM.** Sympathetic nonnoradrenergic cutaneous vasoconstriction in women is associated with reproductive hormone status. *Am J Physiol Heart Circ Physiol* 282: H264–H272, 2002.
38. **Stephenson LA, Kolka MA.** Esophageal temperature threshold for sweating decreases before ovulation in premenopausal women. *J Appl Physiol* 86: 22–28, 1999.
39. **Stephenson LA, Kolka MA.** Plasma volume during heat stress and exercise in women. *Eur J Appl Physiol* 57: 373–381, 1988.
40. **Sims ST, van Vilet L, Cotter JD, Rehrer NJ.** Sodium loading aids fluid balance and reduces physiological strain of trained men exercising in the heat. *Med Sci Sports Exerc* 39: 123–130, 2007.
41. **Ramanathan LN.** A new weighing system for mean surface temperature of the human body. *J Appl Physiol* 19: 531, 1964.