Heat acclimation and physical training adaptations of young women using different contraceptive hormones

Lawrence E. Armstrong,1,2 Carl M. Maresh,1,2 NiCole R. Keith,1 Tabatha A. Elliott,1 Jaci L. VanHeest,1 Timothy P. Scheet,1 James Stoppani,1 Daniel A. Judelson,1 and Mary Jane De Souza1,3

1Human Performance Laboratory, Department of Kinesiology and 2Department of Physiology and Neurobiology, University of Connecticut, Storrs, Connecticut; and 3Women’s Exercise and Bone Health Laboratory, Department of Exercise Science, University of Toronto, Toronto, Ontario, Canada

Submitted 10 September 2004; accepted in final form 9 December 2004

Armstrong, Lawrence E., Carl M. Maresh, NiCole R. Keith, Tabatha A. Elliott, Jaci L. VanHeest, Timothy P. Scheet, James Stoppani, Daniel A. Judelson, and Mary Jane De Souza. Heat acclimation and physical training adaptations of young women using different contraceptive hormones. Am J Physiol Endocrinol Metab 288: E868–E875, 2005. First published December 14, 2004; doi:10.1152/ajpendo.00434.2004.—Although endogenous and exogenous steroid hormones affect numerous physiological processes, the interactions of reproductive hormones, chronic exercise training, and heat acclimation are unknown. This investigation evaluated the responses and adaptations of 36 inactive females [age 21 ± 3 (SD) years] as they undertook a 7- to 8-wk program [heat acclimation and physical training (HAPT)] of indoor heat acclimation (90 min/day; 3 days/wk) and outdoor physical training (3 days/wk) while using either an oral estradiol-progestin contraceptive (ORAL, n = 15), a contraceptive injection of depot medroxyprogesterone acetate (DEPO, n = 7), or no contraceptive (EU-OV, n = 14; control). Standardized physical fitness and exercise-heat tolerance tests (36.5°C, 37% relative humidity) were performed before and after HAPT, demonstrating that the three subject groups successfully (P < 0.05) acclimated to heat (i.e., rectal temperature, heart rate) and improved muscular endurance (i.e., sit-ups, push-ups, 4.6-km run time) and body composition characteristics. Examination of anterior hypothalamic (AH) neuron activity and body composition variables. A significant difference post-HAPT in the onset temperature of local sweating, ORAL (37.2 ± 0.4°C vs. DEPO (37.7 ± 0.2°C), suggested that steroid hormones influenced this adaptation. In summary, virtually all adaptations of healthy young women as they participated in a 7- to 8-wk program (i.e., spanning two menstrual cycles) of heat acclimation and physical training (HAPT). Each woman was voluntarily using either oral contraceptive pills, an injectable steroid contraceptive, or no contraceptive (control).

This research is important and relevant to the unique physiological adaptations and lifestyles of women in two ways. First, ~11.2 million women in the United States, aged 18–44 yr, utilize a pharmacological contraceptive preparation (13), 28% of all women engage in regular leisure-time physical activities (5), and outdoor exercise is more common during summer months when ambient temperatures are high. The effects of programs like HAPT on reproductive physiology are unknown. Because the present investigation involved moderate- to high-intensity exercise, we hypothesized that HAPT would not alter menstrual cycle length/phase characteristics, ovulation, or plasma hormone concentrations in the eumenorheic-ovulatory (control) subjects. Second, because the steroid environment of synthetic steroid preparations and the ovarian steroid hormones of the menstrual cycle are markedly differ-

estrogen; progesterone; estradiol; progestin; rectal temperature

DURING THE PAST TWO decades, animal and human investigations have clarified the neuroendocrine interactions between the thermoregulatory and reproductive systems. Specifically in mature rats, estradiol (E2) applied to in vitro tissue slices increased the activity and firing rate of warm-sensitive neurons but not cold-sensitive neurons harvested from the preoptic area of the anterior hypothalamus (32), whereas progesterone (P4) decreased these responses in warm-sensitive neurons and increased them in cold-sensitive neurons (28). In humans, it is generally recognized that E2 decreases body temperature by enhancing effector responses (i.e., sudomotor and vasomotor) and heat loss (31). Conversely, systemic P4 increases core temperature (6), suggesting an upward shift in the thermoregulatory set point (24). These observations agree with the known changes of E2, P4, and body temperature during the human menstrual cycle. During the early follicular phase, both serum E2 and P4 concentrations are low; during the late follicular phase, E2 is elevated and P4 is low; and both E2 and P4 are high during the midluteal phase (12, 23). Because of these hormone fluctuations, body temperature during the luteal phase is ~0.3–0.5°C higher than during the late follicular phase (17, 23, 34).

Several human studies have used oral exogenous hormones to evaluate the effects of ethinyl estradiol (EE) and progestins on exercise performance (9, 26), thermoregulation in mild (24–28°C; see Refs. 14, 21, and 31) and hot (30–36°C; see Refs. 34–36) environments, exercise responses while wearing impermeable suits at 40°C (37), and resting vasomotor responses while wearing a water-perfused suit heated to 38.5°C (15). However, little is known about the interactions of exogenous hormones, chronic exercise training, and heat acclimation. Therefore, the purpose of the present investigation was to measure the thermal, metabolic, cardiorespiratory, performance, body composition, and perceptual responses and adaptations of healthy young women as they participated in a 7- to 8-wk program (i.e., spanning two menstrual cycles) of heat acclimation and physical training (HAPT). Each woman was voluntarily using either oral contraceptive pills, an injectable steroid contraceptive, or no contraceptive (control).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
ent, it is possible that oral estrogen-progestin formulations and injectable depot medroxyprogesterone acetate (MPA) influence HAPT adaptations differently. However, after considering that 3 days of estrogen supplementation had no effect (i.e., in adult women using no contraception) on heat transfer to the skin or evaporative cooling during exercise (14), we hypothesized that physiological responses and adaptations would be similar in the three experimental groups throughout HAPT.

METHODS

Subjects. Thirty-six healthy females [mean ± SD; age, 21 ± 3 yr; maximal aerobic power (V\textsubscript{O2 max}), 37.1 ± 4.1 ml kg\textsuperscript{-1} min\textsuperscript{-1}; height, 162.5 ± 7.6 cm; body mass, 65.80 ± 11.10 kg] were selected as subjects from a sample of 72 potential participants. Fourteen women were eumenorrheic, had documented normal ovulatory patterns, were not taking exogenous contraceptive preparations, and served as the control group (EU-OV). Fifteen other participants used oral EE-progestin contraceptives (ORAL), and seven received contraceptive injections of depot MPA (DEPO; Depo-Provera; Pfizer, New York, NY). Subjects did not change their prestudy contraceptive status (i.e., previous 3 mo) during this investigation. The University of Connecticut Institutional Review Board for Human Studies approved all procedures. During the month of October, potential subjects attended an informational briefing, completed medical history questionnaires, and provided their signed informed consent to participate.

Preliminary screening. During the month of November, exercise history and eating disorder questionnaires were completed and evaluated. Likely candidates for each treatment group then provided an initial venous blood sample. This sample was analyzed to verify self-reported nonpregnancy and normal thyroid function [i.e., prolactin (PRL), thyroid-stimulating hormone, free thyroxin]. Each participant provided documentation of a recent medical examination, permission of her personal physician to participate in this research, and a normal Papanicolaou smear. Other exclusionary criteria included: change in menstrual pattern or contraceptive use within the previous 3 mo, use of oral contraceptive preparations containing mestranol, recent heat exposure or heat acclimatization, previous history of exertional heat illness, major depression, acute or chronic illness (i.e., of a respiratory, cardiovascular, convulsive, hypertensive, or metabolic nature), self-reported frequent physical training, tobacco use, and drug or alcohol dependence.

Secondary screening. Normal ovulatory menstrual physiology was evaluated for EU-OV subjects via venous fasting blood sample analyses [E\textsubscript{2}, P\textsubscript{4}, luteinizing hormone (LH), follicle-stimulating hormone, and sex hormone binding globulin (SHBG)] during November and December in three phases. First, subjects reported to the laboratory for venipuncture within 2–5 days of the first day of menses to verify the early follicular phase via E\textsubscript{2} and P\textsubscript{4} serum analyses. Samples were then collected every third day until approximately days 9–11 (dependent on previous menstrual cycle length) and then daily to document the occurrence of an LH surge. In these samples, E\textsubscript{2}, P\textsubscript{4}, and LH were assayed. After the documentation of an LH surge, blood samples were collected every third day until the end of the cycle to document luteal phase characteristics. Subjects who failed to demonstrate an LH surge or who had a luteal phase length <10 days and a midluteal P\textsubscript{4} <15.9 nmol/l were excluded from the study. These analyses were continued for the duration of the study. Analyses of circulating reproductive hormones were performed at the Department of Fertility and Reproductive Endocrinology, New Britain General Hospital (New Britain, CT).

All subjects voluntarily ingesting oral contraceptives (ORAL group) reported the exact preparation, as shown in Table 1, and duration of use. All oral preparations contained EE and a second- or third-generation progestin. These subjects also provided investigators with empty pill packs at the end of each month to document therapy details. Compliance to therapy was evaluated by measuring the concentration of plasma EE before training and at the end of training. All subjects voluntarily receiving long-acting injectable depot MPA contraceptive therapy (DEPO group) reported the exact dose, duration of use, and compliance to therapy. The concentration of plasma MPA was measured in serum before training and at the end of training to verify compliance.

Table 1. Synthetic steroid content of the oral contraceptives that were voluntarily taken by women in the ORAL group

<table>
<thead>
<tr>
<th>Estrogen (mg)</th>
<th>Progestin</th>
<th>Oral Contraceptive Trade Name</th>
<th>No. of Subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td>EE (0.035)</td>
<td>Norethindrone (1.00 mg)</td>
<td>Ortho-Novum 10/11</td>
<td>3</td>
</tr>
<tr>
<td>EE (0.035)</td>
<td>Norethindrone (0.50, 0.75, and 1.00 mg)</td>
<td>Ortho-Novum 7/7/7</td>
<td>3</td>
</tr>
<tr>
<td>EE (0.035)</td>
<td>Norgestimate (0.25 mg)</td>
<td>Ortho-Cyclen</td>
<td>2</td>
</tr>
<tr>
<td>EE (0.035)</td>
<td>Norgestimate (0.18, 0.22, and 0.25 mg)</td>
<td>Ortho-TriCyclen</td>
<td>3</td>
</tr>
<tr>
<td>EE (0.030)</td>
<td>Desogestrel (0.02 mg)</td>
<td>Marvelon 28</td>
<td>2</td>
</tr>
<tr>
<td>EE (0.030)</td>
<td>Gestodene (0.75 mg)</td>
<td>Femodene</td>
<td>2</td>
</tr>
</tbody>
</table>

Note: the values in columns 1 and 2 indicate the amount of ethinyl estradiol (EE) or progestin consumed for 21 days, followed by 7 days of placebo pills or no pills. ORAL, oral estradiol-progestin contraceptive.
Body Composition and $\dot{V}O_2_{\text{max}}$. Each subject was weighed under water while suspended from a computer-monitored strain gauge and on land to determine body density, as described elsewhere (1). From body density values, the percent body fat and the fat-free mass were calculated according to published normative equations (33). On the same day, each subject completed an incremental run on a motorized treadmill to determine $\dot{V}O_2_{\text{max}}$. This test began at a brisk pace (i.e., based on each individual’s previous running experience or estimated time to complete a 5-km run; ~75% $\dot{V}O_2_{\text{max}}$) for 4 min at 0% grade. The treadmill grade was then increased to 4% for 2 min. At 2-min intervals thereafter, the grade was further increased 2% until the subject reached volitional exhaustion. Oxygen uptake, minute ventilation and respiratory exchange ratio were measured every 30 s via indirect calorimetry (metabolic cart, model CPX/D; Medical Graphics, St. Paul, MN). Gas concentrations and flow volume were calibrated to manufacturer specifications before each $\dot{V}O_2_{\text{max}}$ test using certified $O_2$–$CO_2$ gas mixtures and a 3-liter calibration syringe (model RS550; Vacumed, Ventura, CA).

Two of the following criteria verified the attainment of $\dot{V}O_2_{\text{max}}$: an increase of oxygen consumption >150 ml/min despite an increase in work load, heart rate (HR; Vantage XL heart rate monitor; Polar Electro, Woodbury, NY) >90% of predicted maximum (220 beats/min – chronological age), or respiratory exchange ratio ($\dot{V}CO_2/\dot{V}O_2$) >1.10. Both the body composition and the $\dot{V}O_2_{\text{max}}$ procedures were repeated at the end of training.

HAPT program. After successfully completing screening procedures, subjects participated in 7–8 wk (i.e., two menstrual cycles) of supervised HAPT. HAPT occurred 6 days/wk as three outdoor sessions and three indoor sessions during the months of January and February. Outdoor physical training and indoor heat acclimation activities in both cold and hot environments were combined to simulate the variety of stressors encountered by athletes and military personnel.

Outdoor physical training involved strenuous running for 4.6 km at an individualized pace (i.e., exercise intensity estimated at 60–85% $\dot{V}O_2_{\text{max}}$, push-ups, and sit-ups; exercise intensity increased weekly as training progressed). During morning running sessions, the mean ± SD outdoor air dry bulb and dew point temperatures were -3.9 ± 0.8 and -5.9 ± 0.9°C, respectively. Adaptations to the 7- to 8-wk physical training program were evaluated as change in body composition and fitness test scores (before vs. after HAPT). The physical fitness tests measured the maximum number of standardized sit-ups and push-ups that could be completed in 60 s as well as a timed 4.6-km competitive footrace.

Indoor 90-min heat acclimation sessions took place in climatically controlled conditions of 36.0–37.0°C and 33–37% relative humidity (%rh); chilled drinking water was consumed ad libitum. Partial results concerning water consumption and thirst have been reported elsewhere (29). Exercise heat exposure employed a circuit of bench stepping, stair climbing, cycle ergometry, and walking on a motorized treadmill at ~50–70% $\dot{V}O_2_{\text{max}}$ for each mode. Subjects were encouraged to exercise continuously for 90 min, and, after the initial 1–3 days, subjects accomplished 90 min/day with very few sporadic exceptions. If a subject voluntarily stopped exercising because of fatigue, she rested in the heat for the entire 90-min period. Subjects inserted a rectal thermometer (YSI 401 rectal probe; Yellow Springs, Yellow Springs, OH) 10 cm beyond the external anal sphincter and wore a heart rate monitor to allow rectal temperature ($T_{re}$) and HR, respectively, to be monitored. If a subject announced that she was unable to continue exercising, she was encouraged to rest in the environmental chamber for the entire 90-min session. However, subjects were removed from the environmental chamber if one of the following predetermined criteria was reached: $T_{re}$ ≥39.5°C, HR >180 beats/min for five consecutive minutes, or signs of exertional heat illness. A trained investigator monitored all training sessions.

**EHT testing.** Subjects performed a standardized EHT before and after HAPT; both EHT began at the same point relative to the reproductive hormone cycle and at the same time within each day. Water deprivation (24 h) plus mixed aerobic activity (2–3 h) were used to induce dehydration before each EHT. This dehydration was incorporated to increase the total stress experienced by test subjects. Body mass (±50 g; floor scale, model DS44L; Ohaus, Pine Brook, NJ) was measured immediately before and after water restriction plus activity to ensure the desired 3.0% body mass loss; also, if urine specific gravity (refractometer) did not exceed 1.020, the test was not conducted. Body mass again was measured upon entering, and immediately before leaving, the environmental chamber. These hydration procedures were controlled during both EHT before and after HAPT.

Once inside the chamber (36.5 ± 1.4°C, 37 ± 1%rh, 2.3 m/s air flow), subjects stood quietly for 20 min to allow body fluids, plasma volume (PV), and skin temperatures ($T_{sk}$) to stabilize. After this equilibration, subjects walked on a motorized treadmill at 93.6 m/min and 5% grade. Treadmill belt speed was verified during each test with a hand-held tachometer (model 8240–20; Cole Parmer Instruments, Chicago, IL). No water was consumed during both EHT. Physiological criteria for test termination were identical to those used during indoor heat acclimation sessions (above). In addition, EHT were stopped if subjects verbally announced volitional exhaustion or completed the entire 90 min of exercise.

Throughout the EHT, physiological and perceptual measures were obtained, as depicted in Fig. 1. Changes in body mass were used to calculate whole body sweat rate ($SR_{wb}$), corrected for urine production and sweat absorbed by clothing. Blood pressure was measured with a sphygmomanometer and stethoscope immediately before exercise and at 30-min intervals thereafter. HR (Vantage XL heart rate monitor; Polar Electro), $T_{re}$ (YSI 401 Rectal Probe; Yellow Springs, Pushups, OH), and ratings of perceived exertion (6- to 20-point scale;
see Ref. 8) were determined at rest and every 15 min during and at the conclusion of the EHT. Measures of oxygen uptake were obtained at 20 and 40 min of exercise using the metabolic cart described above. Total exercise time was recorded to the nearest second.

Tsk were measured at three sites at the end of the EHT. During the standing equilibration, ~6 cm² of skin over the left lateral midcalf, the left dorsal midforearm, and the left pectoralis muscle (at the midpoint between the subject’s nipple and midclavicle) were shaved and cleaned with alcohol. Tsk were measured at these sites with an infrared temperature scanner (Otogram model HTTS-3000; Exergen, Watertown, MA). Mean weighted Tsk was calculated using the following formula (11):

\[ T_{sk} = 0.50 \text{ (chest)} + 0.14 \text{ (forearm)} + 0.36 \text{ (calf)} \]

Sweat sensitivity. Within 24 h of both EHT, a separate experiment was administered to determine sweat sensitivity. Upon arrival at the laboratory, euhydration was verified by a urine specific gravity measurement <1.020. Subjects inserted a rectal thermistor (YSI 401 Rectal Probe; Yellow Springs) 10 cm beyond the external anal sphincter. Exercise involved ~20 min of cycle ergometry at 45% \( \dot{V}O_2 \text{max} \) while in a euhydrated state in a mild environment (22.5°C).

This test was designed to eliminate dehydration and high ambient temperature as confounding factors. Local sweat rate was measured continuously with a dew point sensor (model BI-2; Bi-Tronics, Guilford, CT) attached with surgical cement to the skin above the medial border of the left scapula. Tsk was observed with a digital thermometer (±0.1°C) concurrent with each local sweat rate measurement. The threshold temperature for the onset of sweating was identified by observing the inflection point (i.e., from resting baseline sweat rate) during continuous measurements. Sweat sensitivity was defined as the slope of the relationship between Tsk plotted against local sweat rate (i.e., statistical regression of the linear segment).

Blood analyses. Immediately before and at the conclusion of the EHT, blood samples were obtained via an indwelling cannula (JELCO4054; Johnson and Johnson Medical, Arlington, TX). Posture was controlled (see above). Blood (10 ml) was withdrawn in a plain glass tube (Becton-Dickinson, Franklin Lakes, NJ) for the measurement of hematocrit and Hb. Hematocrit was determined in triplicate via the microcapillary technique. Hb was measured in triplicate via the cyanmethemoglobin method (kit 525; Sigma Chemical). Percent change in PV was calculated using hematocrit and Hb values (19). An additional 10 ml blood were withdrawn in a chilled glass tube (Becton-Dickinson) pretreated with EDTA for the measurement of plasma glucose and lactate. These samples were centrifuged at 3,000 rpm and 4.0°C for 15 min. Plasma lactate and glucose values were determined in duplicate using a glucose and l-lactate analyzer (model 2300; Yellow Springs Instruments).

Data analyses. The mean and SD were calculated for all variables. Variables were evaluated with repeated-measures univariate or multivariate ANOVA to calculate differences between groups, across time, and/or their interaction where appropriate. Significant F-ratios were further evaluated with Tukey’s post hoc test. Significance was established at P < 0.05.

### RESULTS

**Reproductive hormones.** The concentrations of E2, P4, and PRL in all groups (P > 0.05) did not change during the course of HAPT (not acclimated vs. heat acclimated values; Table 2). Significantly higher levels of SHBG (P < 0.05; before vs. after HAPT) were measured in ORAL (vs. EU-OV and DEPO).

Although HAPT enhanced physical fitness and induced heat acclimation in all groups (see below), no main effects (between groups or across time; P > 0.05) were observed for E2 and P4 concentrations. However, PRL significantly increased during HAPT (P < 0.05) in EU-OV (+33.3%) but not ORAL (+3.4%) or DEPO (+24.3%).

All menstrual cycles of EU-OV, before and after the HAPT intervention, were ovulatory. In EU-OV, menstrual cycle length (before HAPT, 27.1 ± 0.6 days; after HAPT, 28.1 ± 1.0 days), day of ovulation (before HAPT, 15.2 ± 0.4 days; after HAPT, 15.2 ± 0.6 days), follicular phase length (before HAPT, 15.2 ± 0.4 days; after HAPT, 15.2 ± 0.6 days), and luteal phase length (before HAPT, 11.9 ± 0.3 days; after HAPT, 12.8 ± 0.4 days) were not altered by HAPT.

The menstrual cycle characteristics, as described by hormone concentrations, are presented in Table 2. Concentrations of E2 measured midcycle (before HAPT, 852.0 ± 81.5 pmol/l; after HAPT, 915.9 ± 91.4 pmol/l) and during the midluteal phase (before HAPT, 483.1 ± 57.6 pmol/l; after HAPT, 487.9 ± 40.4 pmol/l) were unaffected by HAPT. Similarly, midluteal measurements of P4 (before HAPT, 12.9 ± 1.4 ng/ml; after HAPT, 12.5 ± 0.9 ng/ml) and peak LH concentration (before HAPT, 44.4 ± 4.1 IU/l; after HAPT, 44.1 ± 4.9 IU/l) were unchanged by HAPT. The entire HAPT program was completed with high compliance (95% participation) for daily outdoor and indoor sessions.

**Physical training.** Table 3 presents the measurements that indicated improved physical fitness in all groups. The following variables significantly (P < 0.05) improved in all groups, indicating improved physical fitness after 7–8 wk of HAPT intervention. Push-ups per 60 s (+134.2%), sit-ups per 60 s (+46.3%), 4.6-km run time (−24.7%), \( \dot{V}O_2 \text{max} (+9.4\%) \), body fat percentage (−6.7%), and fat-free mass (+3.0%) were all significantly improved across the groups. Because of an increase in fat-free mass and a decrease in fat mass, body mass did not change significantly during HAPT (+0.3%, P > 0.05). No between-group differences existed for any variable. All body composition variables were similar for ORAL, EU-OV.

### Table 2. Endogenous blood variables related to reproductive function measured at rest before (NA) and after (ACCL) the HAPT program

<table>
<thead>
<tr>
<th>Treatment Group</th>
<th>E2, pmol/l</th>
<th>P4, ng/ml</th>
<th>PRL, µg/l</th>
<th>SHBG, nmol/l</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
</tr>
<tr>
<td>ORAL (15)</td>
<td>120.8±18.0</td>
<td>106.8±11.7</td>
<td>0.9±0.2</td>
<td>0.9±0.1</td>
</tr>
<tr>
<td>EU-OV (14)</td>
<td>112.7±11.4</td>
<td>96.9±9.5</td>
<td>0.8±0.1</td>
<td>0.9±0.1</td>
</tr>
<tr>
<td>DEPO (7)</td>
<td>87.7±8.4</td>
<td>82.2±5.9</td>
<td>0.8±0.2</td>
<td>0.8±0.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>14.7±2.1</td>
<td>15.2±2.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>157.1±14.5</td>
<td>180.0±20.6*</td>
</tr>
</tbody>
</table>

Values are means ± SD; n = 10 subjects in parentheses. HAPT, heat acclimation and physical training program; NA, not heat acclimated, before HAPT; ACCL, heat acclimated, after HAPT; ORAL, oral contraceptive group; EU-OV, eumenorrheic-ovulatory group; DEPO. Depo Provera group; E2, estradiol; P4, progesterone; PRL, prolactin; SHBG, sex hormone-binding globulin. Blood samples were drawn as follows: EU-OV, during the early follicular phase (days 2–5); ORAL, during days 2–5 of the placebo phase of the pill pack; DEPO, during days 2–5 of an arbitrary 28-day period, as assigned by investigators. P < 0.05, significant between-group difference (ORAL vs. EU-OV and ORAL vs. DEPO; †) and significant difference across time (NA vs. ACCL, *).
and DEPO throughout HAPT (not acclimated vs. acclimated values).

**Heat acclimation.** Preexercise dehydration level, before EHT, was statistically similar across time and between groups (initial EHT, −2.8 ± 0.6%; final EHT, −2.9 ± 0.6% body mass; all groups combined). E2 and P4 were similar in all groups on the morning of HAPT tests (Table 2).

The influence of HAPT on resting Tre and Tsk was evaluated. A significant (P < 0.03, n = 34) main effect of HAPT was detected, and the mean ± SD baseline Tre values of all groups combined, measured immediately before entering the environmental chamber, were 37.2 ± 0.3°C (before HAPT) and 37.1 ± 0.3°C (after 7–8 wk of HAPT). The baseline Tsk values, measured at the same time as Tre, were not influenced by HAPT (before, 32.2 ± 0.8°C; after, 32.2 ± 0.9°C).

The following variables indicated that all groups achieved heat acclimation. Total exercise time increased after HAPT by 70.9% (P < 0.05), but no mean differences were detected between treatment groups. Final SRwb was not affected by HAPT (Table 4) or contraceptive type. When considered at identical time points in both EHT (i.e., the time at which the initial EHT ended), 7–8 wk of HAPT significantly decreased final HR, final Tre, final Tsk, and final rating of perceived exertion, regardless of group. Oxygen consumption during the EHT decreased by 4.4% (−0.9 ml·kg⁻¹·min⁻¹; initial EHT vs. final EHT) when measured at identical treadmill speeds, despite similar body masses. As noted above, V0₂max significantly increased during this investigation. Thus the mean exercise intensities were 56 ± 5% VO₂max during the initial EHT and 49 ± 5% VO₂max during the final EHT. These results from EHT (36.5°C) are shown in Table 4.

The change of rectal temperature (ΔTre) and change of skin temperature (ΔTsk) during the EHT were altered by 7–8 wk of HAPT, but there were no differences between groups in the change of ΔTre or ΔTsk. During the initial EHT, mean ΔTre (+1.2 ± 0.4°C) and ΔTsk (+0.8 ± 0.8°C) were significantly different (P < 0.005 and P < 0.0001, respectively; n = 34) from the ΔTre (+1.0 ± 0.3°C) and ΔTsk (−0.3 ± 0.8°C) measured during the second EHT.

Table 5 contains plasma measurements from EHT blood samples. The main effect (between group) was not significant for any variable. However, as a result of HAPT, plasma lactate (at post-EHT) significantly decreased (P < 0.05).

Diastolic blood pressure decreased during the initial EHT (P < 0.0005) and during the final EHT (P < 0.002) in all groups. The grand means for diastolic values were as follows: initial EHT (pre, 73 ± 10 mmHg; post, 63 ± 10 mmHg), second EHT (pre, 72 ± 8 mmHg; post, 65 ± 8 mmHg). Systolic blood pressure was similar during the initial EHT (baseline vs. end of exercise, P > 0.05) but rose during the second EHT (P < 0.0005) in all groups. The grand means ± SD for systolic values were as follows: initial EHT (pre, 103 ± 12 mmHg; post, 108 ± 17 mmHg), second EHT (pre, 101 ± 7 mmHg; post, 115 ± 10 mmHg). No differences resulting from treatment group or training were detected.

**Sweat sensitivity.** Local sweat rate measurements (22.5°C environment) are shown in Fig. 2. During HAPT, the sweat onset temperature decreased significantly in ORAL (before HAPT, 37.5 ± 0.2°C; after HAPT, 37.2 ± 0.4°C; P < 0.05) but not EU-OV (before HAPT, 37.5 ± 0.2°C; after HAPT, 37.4 ± 0.2°C) or DEPO (before HAPT, 37.7 ± 0.1°C; after HAPT, 37.7 ± 0.2°C) groups. This resulted in a statistically lower threshold of sweat onset temperature in the ORAL group compared with the DEPO group (post-HAPT; P < 0.05). Sweat sensitivity (i.e., the slope of the linear regression of Tre plotted vs. local sweat rate; linear portion of continuous measurements; Fig. 2) was statistically similar between treatment groups and was not altered by HAPT.

**DISCUSSION**

This investigation was designed to evaluate the differential effects of oral and injectable contraceptive therapies on HAPT

<table>
<thead>
<tr>
<th>Group</th>
<th>Push-ups,* 60 s⁻¹</th>
<th>Sit-ups,* 60 s⁻¹</th>
<th>4.6-km Run* Time, min</th>
<th>Body Mass, kg</th>
<th>Body Fat, %*</th>
<th>Fat-Free Mass, kg</th>
<th>VO₂max,* l·min⁻¹</th>
<th>VO₂max,* m³kg⁻¹·min⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
</tr>
<tr>
<td>ORAL</td>
<td>19 ± 6</td>
<td>45 ± 9</td>
<td>53 ± 10</td>
<td>76 ± 20</td>
<td>40 ± 5.6</td>
<td>65 ± 12.4</td>
<td>26 ± 5.1</td>
<td>108 ± 25</td>
</tr>
<tr>
<td>EU-OV</td>
<td>17 ± 10</td>
<td>44 ± 6.2</td>
<td>71 ± 21</td>
<td>39.4 ± 7.9</td>
<td>28.6 ± 3.1</td>
<td>62.8 ± 9.4</td>
<td>24 ± 5.9</td>
<td>122 ± 26</td>
</tr>
<tr>
<td>DEPO</td>
<td>20 ± 19</td>
<td>37 ± 17</td>
<td>48 ± 10</td>
<td>75 ± 13</td>
<td>39.2 ± 9.1</td>
<td>71.6 ± 10.5</td>
<td>46 ± 4.3</td>
<td>131 ± 21</td>
</tr>
</tbody>
</table>

Values are means ± SD. VO₂max, maximal aerobic power. All variables, except the 4.6-km outdoor run, were measured in a 22.5°C environment. *Significant main effect (across time, P < 0.05) during HAPT. No between-group differences existed for any variable.

### Table 3. Physical training and body composition variables measured before (NA) and after (ACCL) HAPT

<table>
<thead>
<tr>
<th>Group</th>
<th>Push-ups,* 60 s⁻¹</th>
<th>Sit-ups,* 60 s⁻¹</th>
<th>4.6-km Run* Time, min</th>
<th>Body Mass, kg</th>
<th>Body Fat, %*</th>
<th>Fat-Free Mass, kg</th>
<th>VO₂max,* l·min⁻¹</th>
<th>VO₂max,* m³kg⁻¹·min⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
<td>NA ACCL</td>
</tr>
<tr>
<td>ORAL</td>
<td>19 ± 6</td>
<td>45 ± 9</td>
<td>53 ± 10</td>
<td>76 ± 20</td>
<td>40 ± 5.6</td>
<td>65 ± 12.4</td>
<td>26 ± 5.1</td>
<td>108 ± 25</td>
</tr>
<tr>
<td>EU-OV</td>
<td>17 ± 10</td>
<td>44 ± 6.2</td>
<td>71 ± 21</td>
<td>39.4 ± 7.9</td>
<td>28.6 ± 3.1</td>
<td>62.8 ± 9.4</td>
<td>24 ± 5.9</td>
<td>122 ± 26</td>
</tr>
<tr>
<td>DEPO</td>
<td>20 ± 19</td>
<td>37 ± 17</td>
<td>48 ± 10</td>
<td>75 ± 13</td>
<td>39.2 ± 9.1</td>
<td>71.6 ± 10.5</td>
<td>46 ± 4.3</td>
<td>131 ± 21</td>
</tr>
</tbody>
</table>

Values are means ± SD. VO₂max, maximal aerobic power. All variables, except the 4.6-km outdoor run, were measured in a 22.5°C environment. *Significant main effect (across time, P < 0.05) during HAPT. No between-group differences existed for any variable.
adaptations during 7–8 wk of supervised exercise sessions. Consistent with initial hypotheses, 1) the stress of HAPT did not disrupt the menstrual cycle length/phase characteristics, ovulation, or plasma hormone concentrations of EU-OV, and 2) no between-group differences \( (P > 0.05) \) existed for \( T_e \), \( T_a \), metabolic, cardiorespiratory (baseline, final, delta; during EHT), muscular endurance, or body composition variables. In addition, controlled measurements of fitness, body com...
74.0 ± 16.1 min; all groups combined), at the same absolute treadmill speed, plasma lactate concentration decreased. This finding likely arose from either the lower relative exercise intensity (i.e., because of an increased \( V_{\text{O2 max}} \) and improved exercise efficiency subsequent to HAPT) or enhanced lactate clearance (not measured) during the final EHT (7).

**Thermal responses.** The ORAL group experienced a decrease of the threshold \( T_{\text{re}} \) for the onset of sweating (i.e., within-group, not acclimated vs. acclimated) as a result of HAPT. This adaptation also was observed in women who completed 60 days of physical training with no contraception therapy (2). However, similar oral contraceptive preparations (i.e., combined estrogen and progestin) have been shown to increase the sweating threshold of young women in a manner similar to the endogenous hormones of the luteal phase (15, 21). Thus future studies are required to clarify why a significant mean difference of 0.6°C occurred between the threshold temperatures of ORAL and DEPO. Such studies should include women who undertake physical training or heat acclimation programs.

Sweat sensitivity (Fig. 2), measured locally over the scapula, was not different between treatment groups before or after HAPT. This finding is similar to observations published by Charkoudian and Johnson (15) and Grucza et al. (21) that involved the chronic use of female reproductive hormones and changes in the threshold temperatures for effector responses with no change in sweat sensitivity. Furthermore, when estrogen supplementation was administered for only 3 days, Chang et al. (14) observed no adaptation of either the threshold temperature or the sweat sensitivity. At present, the evidence suggests that sweat sensitivity is not influenced by female reproductive hormones.

Sweat production is controlled by the anterior hypothalamus, which integrates afferent \( T_{\text{re}} \) and \( T_{\text{sk}} \) impulses and regulates effector responses, including eccrine sweat gland secretion and skin blood flow (27). Final \( S_{\text{Rwb}} \) did not change as a result of either HAPT or contraceptive type (i.e., both main effects, \( P > 0.05 \); Table 4). This phenomenon was recognized by Henane (22) as one result of the reduced body temperature that occurs with heat acclimation. Because final \( T_{\text{re}}, \Delta T_{\text{re}}, \) and \( \Delta T_{\text{sk}} \) decreased during HAPT, the neural drive for sweat production decreased.

In summary, numerous long-term adaptations occurred during HAPT (i.e., significant main effect of time) in response to HAPT sessions performed by 36 young women. However, the differences of adaptations in ORAL, EU-OV, and DEPO were few, small in magnitude, and did not impart superior physical fitness or heat acclimation to any treatment group. Perhaps the most interesting between-group differences involved the threshold temperatures for local sweating. This threshold decreased in ORAL as a result of HAPT (\( P < 0.05 \)), suggesting that an EE-progestin formulation may lower the hypothalamic set point; also, the use of depot MPA did not induce a significant change of this threshold during HAPT. Although some pharmacological agents may alter sweat rate or reduce cardiac contractility during exercise in a hot environment (25), the use of contraceptives in the present investigation (ORAL and DEPO vs. EU-OV) did not impair or improve the ability of women to complete 7−8 wk of strenuous HAPT.

**ACKNOWLEDGMENTS.** We gladly acknowledge the technical contributions of the following individuals: Dean Arresco, Lynn Bairros, Timothy Bilodeau, David Blair, Dr. Douglas R. Bolster, Valerie A. Collins, Stephen L. Gaffin, Jorge Herrera, Jennifer Holub, Gregory Kane, Dr. Brian E. Miller, Jennifer Ormerod, and Dr. Melissa Roti. Dr. Jeffrey Anderson served as the medical monitor for this investigation.

**GRANTS.**

This research was funded by a grant from the U.S. Army Medical Research and Materiel Command, Fort Detrick, MD, under the auspices of the Defense Women’s Health Research Program.

**REFERENCES.**


