Reproductive Hormone Influences on Thermoregulation in Women

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ABSTRACT

The present discussion reviews current knowledge regarding influences of the primary reproductive hormones on mechanisms of thermoregulatory control in women. The human body is remarkably capable of maintaining body temperature within a few tenths of a degree of normal (37°C) over a wide range of activity and environmental exposures; this regulation is accomplished via integration of central and peripheral thermal information at the preoptic area of the anterior hypothalamus (PO/AH). We describe both central and peripheral mechanisms involved in controlling thermoregulation in humans, and how these mechanisms are affected by sex and hormone exposure. Estrogens generally promote vasodilation, heat dissipation, and lower body temperature and progesterone or progestins generally have the opposite effect. Estrogens and progesterone/progestins can also interact with androgens; this is an important point because androgens in the body can increase in both older and younger women. The study of reproductive hormone (estrogens, progesterone, luteinizing, and follicle stimulating hormones) effects on body systems is challenging because of the complex and multifaceted influences of these hormones, both individually and in combination. Thus, a number of methods to alter hormone exposure are explained in this article. We conclude that men and women do not exhibit major quantitative differences in physiological thermoregulatory responses to exercise and/or body heating when factors such as fitness and body size are taken into account. However, female and male reproductive hormones have important influences that can significantly alter individual thermoregulatory responses at various points throughout the lifespan. Published 2014. *Compr Physiol* 4:793-804, 2014.

Introduction

Reproductive hormones have numerous influences on nonreproductive organs and systems, including important influences on integrative mechanisms regulating body temperature. Our goal in the present discussion is to review current knowledge regarding influences of the primary ovarian and testicular hormones (estrogen, progesterone, and testosterone) on thermoregulatory mechanisms of control in humans. A secondary goal is to discuss differences between the sexes in thermoregulatory responses and adaptations. Our focus will be primarily on evidence from human studies.

The overarching themes of the present discussion are as follows. First, despite historical assumptions that women are less able to respond or adapt to thermal challenges, more recent research suggests minimal differences between the sexes in terms of overall ability to thermoregulate and acclimate to the heat. Second, female reproductive hormones have important influences on thermoregulatory responses with estrogen generally promoting vasodilation, heat dissipation, and lower body temperature and progestins generally having the opposite effect. Many of these influences cross over multiple physiological systems, including cardiovascular and neurological, in addition to strictly thermoregulatory. Testosterone has important modulatory effects as well, but these remain less well understood. Third, there are a variety of approaches to evaluate the integrative physiological influences of reproductive hormones; strengths and weaknesses of each will be discussed. Finally, the influences of reproductive hormones on thermoregulation have important clinical implications across the lifespan, particularly in an era in which exercise and physical activity are so strongly encouraged for all people. In young women, these include influences of menstrual cycle and oral contraceptive use; in older women, changes in endogenous hormones and exogenous hormone therapy both have important effects. Moreover, as more younger and older women and men take androgens, important future directions should include the study of androgen effects on thermoregulation.

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Historical Perspective

Most early studies on thermoregulation were conducted on male subjects, in part due to a general assumption (in the early part of the 20th century) that women were not likely to be as physically active as men and that physiological responses in women could and would vary from those of men. Excluding women from research led to a general failure to recognize the important effects of ovarian hormones on thermoregulation. Despite this neglect in many laboratories, a few studies examined thermoregulation in women (30, 31). Hardy and Milhorat (31) demonstrated lower skin temperature (T_{Sk}) in women ($n = 3$) in the cold, but higher T_{Sk} compared to men in a warm environment, and that the threshold for sweating was delayed in the women. These investigators speculated that the sex differences were a function of an insulating layer of superficial tissue in the women because women had lower heat loss per unit of surface area and lower tissue transduction relative to men ($n = 2$) from previous experiments (31). Hardy and DuBois summarized their examination of sex differences in thermoregulation stating that women ($n = 4$) had a "physiological advantage" in both cold and warm zones (at rest) (30). In the cold women had greater nonshivering thermogenesis response and had reduced heat loss by radiation and convection, while in a warm environment they had a lower metabolic rate (43) and a delayed sweating threshold and lower sweat rate (30). While these studies appeared ground breaking at the time, there were significant and important limitations. For example, the studies did not control for menstrual cycle (although cycle phase was recorded in some studies) so the important influences of sex hormones on thermoregulation were uncontrolled and unnoted. Moreover, the sample sizes were small and were frequently compared to data on men from earlier experiments. Finally, these investigators did not consider acclimation or exposure to the heat while active in either male or female subjects; factors we now know can contribute to thermoregulation. Nonetheless, these investigations set the groundwork for future studies in women.

In 1993, the U.S. Congress passed the NIH Revitalization Act (1) directing the National Institutes of Health to ensure the inclusion of women in clinical research unless "a clear and compelling rationale and justification establishes to the satisfaction of the relevant Institute/Center Director that inclusion is inappropriate with respect to the health of the subjects or the purpose of the research." This Act of Congress also ensured that women of childbearing age could not be systematically excluded from scientific research unless the research did not and would not concern women's health (e.g., prostate cancer). This Act had an enormous influence on physiological studies, and required scientists to rethink many assumptions regarding study design and approach.

Specific to thermoregulation, historical study design and interpretation of data were often based on the assumption that women were less active, and therefore less fit, than men. For example, in 1969 Fox and colleagues (21) studied sweating in men and women and reported that women had a higher sweat onset threshold and lower sweat "capacity" compared to men [similar to the findings in Hardy's laboratory (30, 31)]. This interpretation is similar to what would be seen in a comparison between fit and unfit individuals (47, 54); the investigators proposed that the sex differences may represent fundamental physiological differences, but may also be the result of differences in behavior. They note in their discussion "For social reasons, women probably prefer to avoid sweating more than men. Women usually wear lighter clothing and, in general, take less part in athletic activities demanding a sustained high level of energy expenditure." The women in their study were primarily nurses, with an average VO_2max of 35 mL/kg/min, compared to the average male VO_2max of 46 mL/kg/min. Thus, differences in physical fitness likely contributed to any group differences that were observed in this study.

Another impetus for greater interest in the potential for physiological differences in thermoregulation between men and women was a drastic increase in women's sports participation in the 1960s and 1970s. Although "Title IX of the Educational Amendments," passed in 1972, related to sex discrimination in all education activities, the public impact of this law has been largely notable in sports participation. Specifically, the law stated that "No person in the United States shall, on the basis of sex, be excluded from participation in, be denied the benefits of, or be subjected to discrimination under any education program or activity receiving Federal financial assistance." This law meant that any educational institution providing financial support for school activities was required to distribute the dollars similarly between men's and women's programs. The practical outcome of this law was that financial support for girls' sports in public schools were adjusted to match that of boys and therefore skyrocketed. Title IX was coincident with (or the cause of) a more general recognition of the importance of regular physical activity for health in all people. Title IX resulted in greater participation of girls and women in athletics on a more regular basis and under varied environmental conditions. For example, before the 1980s there were no women's distance races in the Olympics; the 1500 meter was the longest race, having been instituted in 1972 (42). Prior to 1972 women had not been allowed to participate in marathons, as they were considered "physically unable to complete a marathon" (42).

Heat Acclimatization

Until the early 1960s, there was some debate about whether or not women had the physiological capacity to acclimate to the heat. Subsequently, investigations demonstrated that women had augmented physiological heat dissipation mechanisms with repeated heat exposure (that is, women have the physiological capacity to acclimatize to heat) (33, 34, 71, 74). Hertig and colleagues (33, 34) were the first to demonstrate progressive decreases in rectal temperature and heart rate during a 10-day heat acclimation period involving treadmill walking in the heat in women indicating their ability to acclimatize. In

their discussion, however, these investigators stated ". . . at the risk of extrapolating from too few data, we suggest that the available evidence indicates that women may be inherently less able to deal with heat stress than men." This conclusion erroneously suggested that even given adequate training and acclimation, women would be unable to tolerate exercise in the heat to the extent of their male counterparts. Our current understanding of thermoregulation indicates that sex differences are negligible when factors such as fitness and body size are taken into account.

One of the challenges in interpreting these earlier studies is vague presentation and quantification of data relative to current standards. In particular, it was difficult to challenge the assumption that heat tolerance was lower, and cardiovascular strain was higher, in women compared to men. For example, a report from Brouha et al. (5) was often cited as evidence that women have less than adequate "circulatory adjustments" to exercise-heat stress. The authors describe heart rate and blood pressure responses to work and recovery under "hot wet", "hot dry," and "room temperature" conditions (5). The article showed figures of the heart rate and blood pressure responses of the men, and noting only in the text that heart rate ". . . reactions of the women were comparable but at a higher level" and blood pressure "reactions for the women were similar but at a lower level" (5); importantly, the cardiovascular and thermoregulatory data from women *were not presented* in this paper. The authors (5) did, however, provide sufficient data to convert oxygen consumption (VO_{2max}) data to $\%VO_{2max}$ in the women using maximal work capacity. They demonstrated a strong linear correlation between HR and VO_2 (% max) in both men and women, and that women had similar cardiovascular responses at similar $\%VO_{2max}$ to that of the men.

Cardiovascular strain and plasma volume

The idea that women may experience greater cardiovascular strain (in particular, higher heart rates) than men during exercise in the heat was noted in several early investigations. Wyndham and colleagues (74) noted that women ($n = 4$) reached higher heart rates and higher rectal temperatures more rapidly than men during the first few days of an 11-day heat acclimatization protocol. However, as with the studies described above, these studies are also difficult to interpret due to the small sample size and lack of statistical analysis. Contrary to the authors' interpretation, the women ultimately exhibited physiological heat acclimatization similar to the men such that their final heart rate and rectal temperature responses were similar to that of the men in two different exercise/heat stress conditions.

Fortney and Senay (20) tested the hypothesis that the greater cardiovascular strain reported in women was specifically related to plasma volume differences between men and women. They examined this question by looking at potential differences in blood and plasma volumes in men and women, and how these differences might relate to cardiovascular responses to exercise and heat stress. They also tested whether exercise training and heat acclimation, both of which have the capacity to expand plasma volume, reversed or mitigated the greater cardiovascular strain previously reported in female subjects. They demonstrated that heat acclimation resulted in significant decreases in steady state heart rate at 40 min of exercise in the heat in both men and women, concomitant with significant increases in plasma volume following the 2-week program of heat acclimation. Although the authors did not measure stroke volume, they concluded that the expansion of plasma volume likely contributed to the decreased cardiovascular strain (i.e., decreased heart rate) via an increase in stroke volume.

In summary, the second half of the 20th century showed a dramatic increase in women's participation in sports and organized physical activity. When women first began focusing on sports, exercise and physical activity, scientists assumed that women were somehow less able to tolerate the thermal and cardiovascular strains associated with exercise in the heat compared to men. More recent studies using modern methods of assessing thermal strain (22-26), concomitant with cultural shifts in thinking about women in sports, have led us to a point in the early 21st century where we recognize that women's responses to thermoregulatory strain are more similar than different to those in men.

Overview of Thermoregulation

The human body is remarkably capable of maintaining body temperature within a few tenths of a degree of normal (37°C) over a wide range of activity and environmental exposures. Thermoregulation is accomplished via integration of central and peripheral thermal information at the preoptic area of the anterior hypothalamus (PO/AH), which is the primary central nervous system site for control and coordination of efferent thermoregulatory responses. During increases in body temperature (hyperthermia), human heat dissipation is dependent on cutaneous vasodilation and sweating, whereas during decreases in body temperature (hypothermia), both heat conservation (via cutaneous vasoconstriction) and heat generation (via shivering) contribute to maintaining core temperature. The following sections will provide an overview of these thermoregulatory mechanisms in humans as a basis for subsequent discussion of the influences of sex hormones on these responses.

Central neural control of thermoregulation

The PO/AH region contains both temperature-sensitive and temperature-insensitive neurons, which interact to regulate body temperature and coordinate thermoregulatory efferent responses (2). Direct sensing of core temperature includes "warm-sensitive" neurons in the PO/AH, which increase their firing in response to increases in neuronal temperature. This leads to increased efferent heat dissipation responses of cutaneous vasodilation and sweating. Conversely, decreases

in body temperature decrease heat dissipation responses, and increase heat conservation/heat generation responses. In humans, heat generation is primarily via shivering, which involves involuntary, rhythmic muscular contractions, existing specifically for the purpose of generating metabolic heat (rather than for locomotion or other purposes).

With regard to PO/AH control of body temperature, both directly sensed temperature at the level of temperature-sensitive neurons, as well as afferent information from cutaneous, spinal, and visceral thermal receptors are important for optimizing the thermoregulatory response to a given internal and external environment (46). Thus, afferent information about surface (skin) temperature will augment heat dissipation responses in a warm environment and will diminish heat dissipation and/or increase heat generation responses in a cooler environment.

Skin blood flow

Skin blood flow in humans is controlled by both reflex (neurogenic) and local thermal mechanisms. Reflex control includes sympathetic noradrenergic vasoconstriction, and a nonadrenergic active vasodilator system. Local thermal control does not require intact reflex innervation, and includes locally mediated vasodilation in response to increases in local temperature, as well as locally mediated vasoconstriction during local cooling.

Sympathetic noradrenergic vasoconstrictor nerves are tonically active in so-called "neutral zone" environments—those in which body temperature is maintained without sweating or shivering (58). These nerves also cause reflex vasoconstriction in response to cold exposure (8, 37). During mild cooling, this vasoconstriction represents the first line of defense against a lowering of body core temperature by decreasing convective transfer of heat from the core to the periphery, and thus heat dissipation from the core to the cold environment. Cutaneous sympathetic vasoconstrictor nerves release norepinephrine and cotransmitters (such as neuropeptide Y), and cause vasoconstriction (8).

The sympathetic vasoconstrictor nerves are tonically active in normothermic environments, so decreases in their activity during exposure to slightly warmer environments cause a passive vasodilation. In contrast to vasoconstrictor nerves, the sympathetic active vasodilator system does not exhibit tonic activity. The sympathetic vasodilator system is unique to human skin (37). Vasodilator nerves are only turned on during increases in body temperature. Once activated, however, this system is responsible for as much as 80% to 90% of the very large vasodilation that occurs in human skin during hyperthermia. Given that skin blood flow can reach as high as 6 to 8 L/min or 60% of cardiac output with severe heat stress, under such conditions reflex control of skin blood flow has important hemodynamic implications in addition to those for thermoregulation (57).

With regard to both sweating and skin blood flow responses to heat stress, a common approach to analysis of these responses is to graph the effector response (sweating or SkBF) as a function of core temperature. In this way, a threshold core temperature can be identified at which the onset of the effector response occurs, and the slope of the postthreshold relationship can be analyzed to give an index of the sensitivity/gain or responsiveness of the effector to further increments in core temperature. An example of such a graph is shown in Figure 1. Factors (including reproductive hormones, heat acclimation, and hydration) that alter the threshold and/or sensitivity of these responses have major influences on the amount of heat dissipation in a variety of conditions. In doing such analyses, it is important to recognize that skin surface temperature can also influence the responsiveness to increases in core temperature such that high skin temperatures augment responsiveness and low skin temperatures have the opposite effect (51).

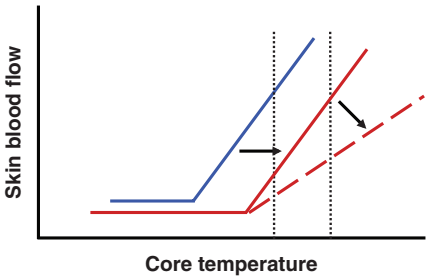

Figure 1 Schematic example of graphs used in analysis of thermoregulatory effector mechanisms (primarily sweating and cutaneous vasodilation), showing the relevant effector as a function of core body temperature. As core temperature increases, a point is reached (the threshold) at which the heat dissipation mechanism begins to increase. The slope of the relationship after this threshold is referred to as the sensitivity of the response. A "rightward" shift in threshold and/or a decrease in sensitivity will decrease the amount of heat dissipation for a given core temperature, resulting in less efficient heat loss. Vertical lines show the change in the amount of a given effector response (at a given core temperature) caused by a shift in threshold or sensitivity.

Sweating

Sweating works with skin blood flow to increase dissipation of heat from the body. Where cutaneous vasodilation increases convective transfer of heat from the core to the periphery, the evaporation of sweat cools the skin such that heat dissipation can occur more effectively at the skin surface. Efferent sympathetic cholinergic nerves innervate eccrine sweat glands to induce sweating. Increases in core body temperature activate these nerves, which release acetylcholine. The acetylcholine binds cholinergic muscarinic receptors at the sweat gland. Important for humans, sweating is only as effective as the ability of the sweat to evaporate. Thus, a humid environment tends to be less comfortable in part because sweat evaporation is less efficient when ambient water vapor pressure is high. Sweating is much less effective in actual body cooling when most of that sweat is dripping off the body surface rather than evaporating (8).

Like skin blood flow, reflex control of sweating can be studied by evaluating the response as a function of body temperature. Moreover, factors that influence threshold and

sensitivity of the sweating response provide insight into altered central and peripheral control mechanisms. For example, in dehydrated individuals, both the threshold and sensitivity of the sweating response are increased, meaning that sweating is not initiated until higher core temperatures are reached, and that sweating responds less to further increases in core temperature. Both of these alterations reduce the effectiveness of heat dissipation and increase the risk of hyperthermia.

Peripheral mechanisms of the sweating response also provide important insight into integrative control mechanisms. Direct (peripheral) stimulation of sweat glands can be performed using chemicals such as acetylcholine or pilocarpine (a nonselective muscarinic receptor agonist). These substances directly interact with cholinergic receptors and activate sweat glands, and are, therefore, used to evaluate potential dysfunction or altered function of pathways downstream of cholinergic receptor binding.

Studying Estrogen and Progesterone Effects on Physiological Systems in Women

The study of reproductive hormone (estrogens, progesterone, luteinizing, and follicle stimulating hormones) effects on body systems is challenging because of the complex and multifaceted influences of these hormones, both individually and in combination. The impact of estrogens and progesterone on temperature regulation has been studied in humans using a variety of approaches to alter exposure to estradiol and progesterone, most notably during different phases of the menstrual cycle with oral contraceptive administration, with ovarian hormone suppression combined with hormone administration in young women (see Fig. 2), and with hormone therapy in postmenopausal women.

Any discussion of the impact of hormone exposures on any physiological system must take into account that

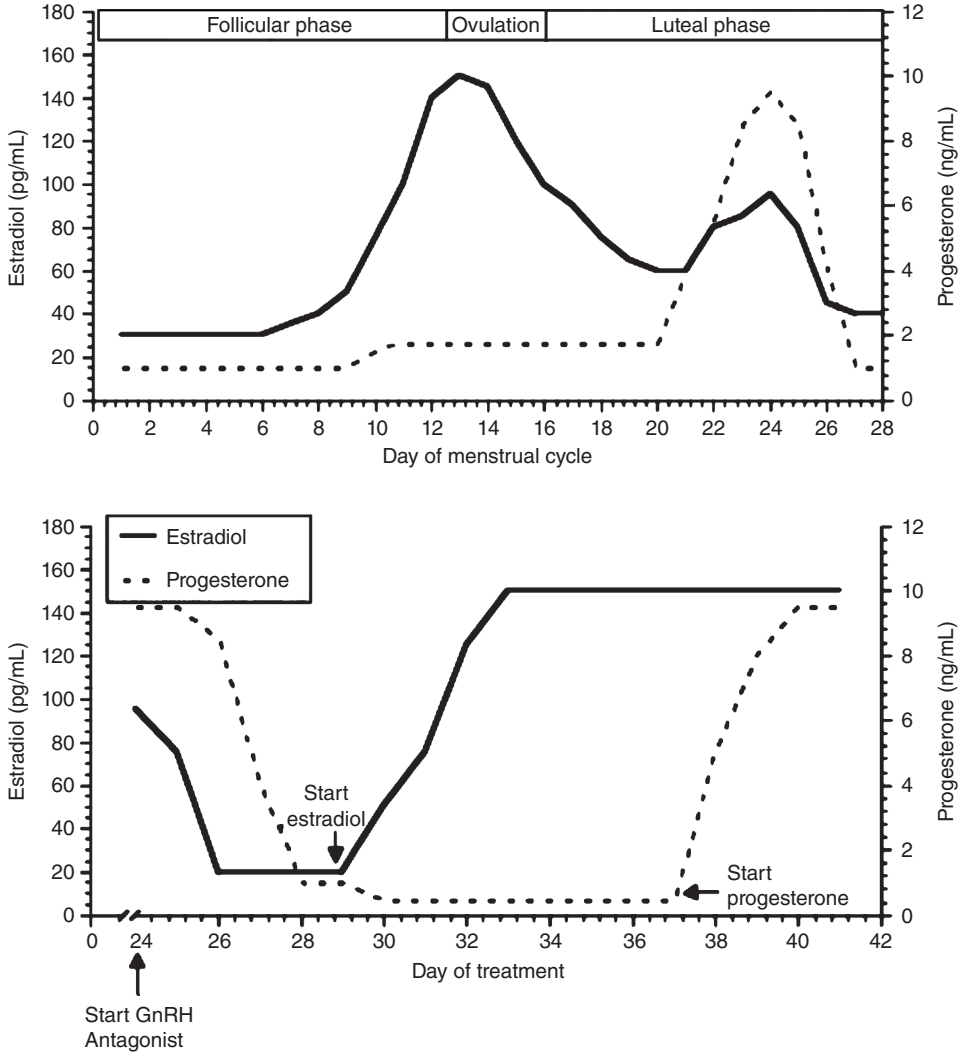

Figure 2 Typical concentrations of plasma estradiol and progesterone over a normal menstrual cycle (top) and during treatment with gonadotropin-releasing hormone (GnRH) antagonist, followed by estradiol and progesterone (bottom). (65) *With permission.*

reproductive hormones fluctuate across the menstrual cycle in young women, and continue to change as women age. Reproductive hormonal changes are under both central nervous system (brain) and peripheral (ovarian, testicular, and adrenal) control. The primary ovarian hormones (estrogens and progesterone) influence body temperature and thermoregulation, both at rest and during exercise. This hormonal input is an integrative part of reproduction, with the primary purpose of creating an environment most hospitable for conception and a developing fetus. These hormone effects on thermoregulation also influence many other physiological systems, including body temperature itself and any physiological system dependent on a controlled temperature environment.

Experimental considerations: Designing studies to control for hormonal effects

There are two main reasons why investigators try to control for the influences of reproductive hormones in studies of thermoregulation: first (and more common), the goal is often to minimize any such influences which may "confound" results regarding scientific questions unrelated to reproductive hormones *per se*. Second, investigators control for reproductive hormone levels when they want to specifically investigate the thermoregulatory influences of one or more of these hormones.

There have been a number of experimental designs employed to control for/minimize the influences of reproductive hormones on thermoregulatory variables in women. The most commonly used method in human studies to control for ovarian hormone fluctuation is to test within the same phase of the menstrual cycle across subjects or study days. Investigators usually use the early follicular phase (the first 5-7 days of the menstrual cycle) because it is in this period of the cycle that both estrogens and progesterone are at their lowest (Fig. 2). While this method is reasonable, a weakness is that women are only examined in one phase of the cycle. Thus, while the goal of controlling for and/or minimizing the influence of reproductive hormones may be addressed, the broader clinical relevance may be limited, as women are only in this part of their cycle for ~25% or less of their reproductive lives.

For those studies in which the goal is to evaluate specific influences of reproductive hormones on thermoregulatory variables, various approaches have been used. Hormonal contraceptives (usually combinations of various types of estrogens and progestins) are often used to control hormone exposures. Using hormonal contraceptives increases hormone exposures above endogenous levels of estrogens and progesterone, and combined hormonal contraception provides a steady state environment with which to compare to women not taking hormones, or during the standard "placebo" week, when the women cycle off the hormones. A strength of this design is that many women now take hormonal contraceptives, which improves the clinical applicability of findings. Depending on the specific research question(s) to be addressed, important considerations regarding the contraception method include the fact that progestins found in hormonal contraceptives differ in some of their basic hormonal actions when compared to endogenous progesterone. For example, progestins in hormonal contraceptives can have mild androgenic properties relative to endogenous progesterone (62), and progesterone, progestins, and androgens can alter peripheral circulation (16,17,61,72), blood pressure (53,55), and temperature regulation (64). In this context, investigators often use the placebo week as a basis for comparison; an important limitation is that they cannot control or measure tissue exposure to hormones and their metabolites during this time.

A more controlled method to isolate individual effects of estradiol or progesterone on temperature regulation is temporary suppression of the menstrual cycle with a gonadotropin releasing hormone agonist (leuprolide acetate, *Lupron*) or antagonist (ganirelix acetate, *Antagon*). Leuprolide, the agonist, has greater GnRH receptor binding and decreased degradation compared to endogenous GnRH, so is a potent inhibitor of gonadotropin secretion. Continuous leuprolide administration downregulates the hypothalamic pituitary ovarian axis causing internalization and uncoupling of the GnRH receptors in the pituitary. Thus leuprolide interrupts the normal pulsatile stimulation of GnRH receptors so that they become desensitized. Thus, administration of leuprolide causes an initial FSH stimulation and related steroidogenesis, followed by low or undetectable estrogen and progesterone concentrations within 14 days (63). Ganirelix acetate is a synthetic decapeptide, which competes with naturally occurring GnRH for receptor binding so functions as a competitive receptor antagonist, inducing a rapid, reversible suppression of gonadotropin secretion (49,50). In eumenorrheic women, administration of ganirelix acetate leads to suppression of estrogens and progesterone to postmenopausal levels after 36 to 48 h of administration (Fig. 2) (49,50,66). While this is the method most able to permit causal inferences about hormonal effects on the system targeted for study, its primary weakness is that it is the most invasive and the most difficult for subjects because of the GnRH agonist/antagonist injections. Also, the GnRH agonist/antagonist + hormone protocol may not mimic any specific natural hormone exposure period. Moreover, studies with the agonist can take up to 6 weeks, so may induce vasomotor symptoms, especially in the warm weather. These limitations can be partially addressed by the methods and doses of the hormones used during administration.

Influences of Reproductive Hormones on Thermoregulation

Estrogen and progesterone

Studies examining temperature regulation across the menstrual cycle have demonstrated an increase in the thermoregulatory set point when progesterone exposure is high, but a

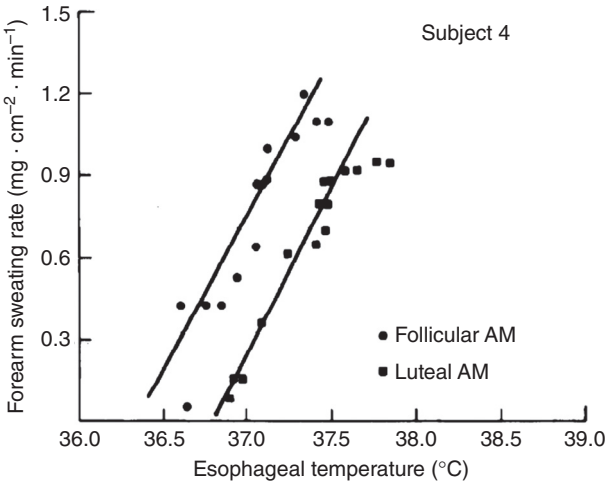

Figure 3 A classic example showing sweating rate as a function of esophageal temperature during the early follicular and mid luteal phases of the menstrual cycle. Note the shift to higher body core temperature of the sweating response in the luteal phase, resulting in lower sweating (and, presumably, heat dissipation) for a given core temperature in this phase (68). *With permission.*

lower thermoregulatory set point when estradiol is elevated unopposed by progesterone (40, 67). The progesterone peak during the mid-luteal phase is associated with higher resting core temperature (T_c) and a rightward shift in the T_c threshold for thermoregulatory peripheral effector responses (Fig. 3) (68). This is consistent with a shift in the onset of reflex cutaneous vasodilator responses to higher T_c during combined (estradiol + progestin) contraception administration during passive heating (10) and exercise (56). This shift appears to be due primarily to a shift in the onset of the active vasodilator system to higher body core temperature (10, 11).

Conversely, during the preovulatory phase of the menstrual cycle, the cycle phase characterized by rising estradiol levels, resting T_c and the thresholds for cutaneous vasodilation and sweating are shifted to a lower T_c during exercise (67). This shift reflects of an overall shift to a lower set-point T_c around which body temperature is regulated. This is consistent with central neuronal data from Silva and Boulant, who showed that estradiol increases the firing rate of warm-sensitive neurons in the rat PO/AH, suggesting the promotion of heat dissipation via central mechanisms (60). Figure 4 shows an example of this phenomenon.

This effect of estradiol may decrease the risk of overheating by shifting overall regulation of body temperature to lower levels. In terms of absolute body temperature levels, this may decrease risk for heat illness/heat stroke, since temperature is regulated at lower levels for any given exposure. Conversely, the hormone-associated delay in these thresholds to a later T_c results in overall higher body temperatures, potentially bringing women closer to temperatures that would be associated with dangerous hyperthermia. However, to our knowledge, it is unknown whether an increased risk of hyperthermia has in fact been documented in luteal compared to follicular phases. Taken together, existing data suggest that estradiol and progesterone may have opposite effects on temperature regulation, but the progesterone effects appear to predominate when the two are concomitantly increased (3,9-11,35,38,39,68,69).

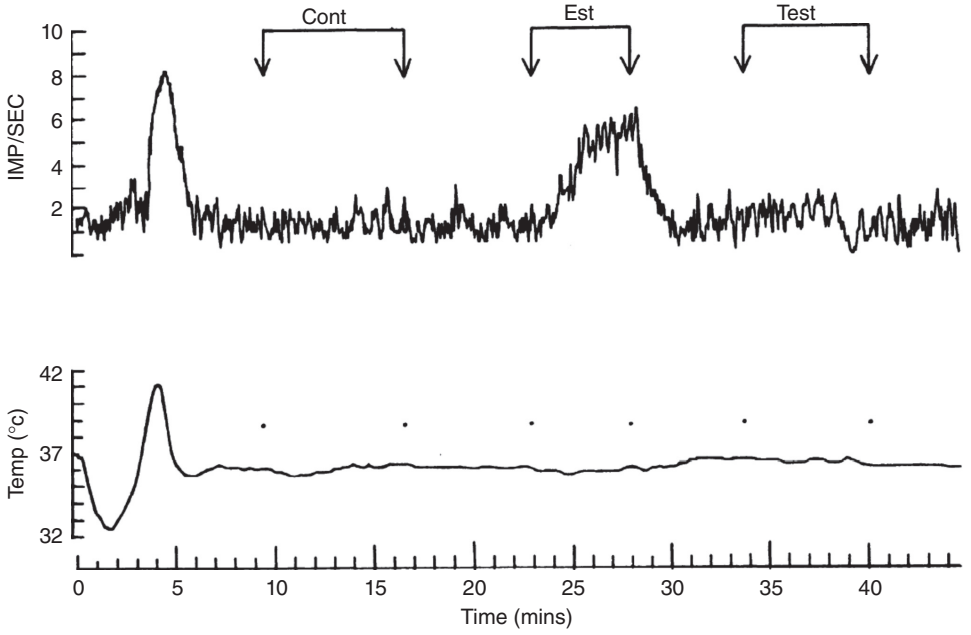

Figure 4 Data from Silva and Boulant (60) showing the response of a warm sensitive neuron in the rat PO/AH region to estradiol and testosterone perfusion. This particular neuron increased its activity in response to estradiol but not to testosterone. Overall, 26% of warm sensitive neurons studied increased activity in response to estradiol, and 32% increased activity in response to testosterone. *With permission.*

However, the interpretation that progesterone effects on temperature regulation predominate over those of estradiol may be a simplified one. For example, estrogen exposure can modulate progesterone-associated temperature increases (64), an influence which may be due to modulatory effects of cold-sensitive neurons, temperature insensitive neurons of the PO/AH (60) or through peripheral alteration of effector responses (e.g., a direct vascular effect of estrogen to promote vasodilation). In some physiological systems, progesterone receptors require upregulation by estradiol in order for progesterone to induce physiological responses (15, 70). Thus the shifts in thermoregulation seen during the mid-luteal phase may be due to estradiol-mediated upregulation of progesterone receptors involved in the control of temperature, rather than a predomination of progesterone over estrogen on thermoregulatory effector responses. Moreover, changes in the sex hormones and changes in temperature regulation may be an indication of a common point of control, rather than being causally related.

Compared to the early follicular phase of the menstrual cycle, progestin-only contraception administration increases resting T_c and delays the T_c threshold for the onset of sweating during exercise. These changes in T_c were attenuated with a combined (64) ethinyl estradiol-progestin contraception treatment (64). In these same subjects, the sweating threshold was delayed during the mid-luteal phase relative to the early follicular phase but was also delayed compared to the combined estradiol/progestin contraceptive administration. Because in the mid luteal phase both estrogens and progesterone are elevated (see Fig. 2) as they are with combined estradiol/progestin contraception, these findings suggest that contraception administration does not mimic the hormonal milieu as it changes over the course of the menstrual cycle. Likely the most important difference is that the progestins in oral contraceptives are structurally different than endogenous progesterone, so receptors in the thermoregulatory system in both the brain and periphery respond differently to progestin versus progesterone exposure.

Hormone therapy

Although there has been controversy regarding the clinical use of hormone therapy, many women in the perimenopausal and menopausal years use exogenous estrogens, progesterone or a combination of these hormones. It is in these transitional periods that hormones are successful interventions to treat symptoms of menopause, including vasomotor symptoms ("hot flushes") (14,36,52). Combined estradiol and progesterone treatments are generally considered safe with regard to cardiovascular disease during this perimenopausal and early menopausal periods, while some studies have demonstrated that beginning these hormones later in menopause can increase the risk of heart disease and stroke (28). With regard to thermoregulation, estrogen exposure generally reduces or completely alleviates vasomotor symptoms (14, 36, 52). Moreover, progesterone-only therapy may reduce vasomotor symptom "rebound" after therapy is discontinued (52). Short term (3-day) (3) and chronic (4) 17 β-estradiol administration decreases body temperature and reduces the T_c threshold for activation of cutaneous vasodilation during passive heating in postmenopausal women. Combined estradiol with progesterone did not elicit this response.

Testosterone effects in women

Testosterone declines with age in both men and women, although it is unclear if menopause itself has profound effects on serum androgens. Some evidence suggests that there is a rapid decline in testosterone after menopause, but that levels begin to increase with advancing age (after 70 years) (41), whereas other investigators have reported no change (6) or an increase in testosterone during menopause (44). In perimenopause, low blood testosterone concentration is associated with low quality of life (27), and testosterone administration is increasingly prescribed to peri- and postmenopausal women to treat a variety of conditions ranging from bone loss to suppressed libido. Some perimenopausal women taking estrogen therapy have lower testosterone levels; conversely, high testosterone levels are associated with greater frequency of vasomotor symptoms (27). While there is little understanding of the mechanism(s) involved, it is likely that important interactions exist among estrogens, progesterone, and testosterone and these interactions can impact thermoregulation.

Synthetic testosterone is sometimes used by young women to improve body composition. Anabolic steroid use is increasing in adolescent girls (18, 75). In fact, the Centers for Disease Control reported in 2003 that 5% of girls between ages 12 to 18 had used anabolic steroids, a level exceeding that of boys of similar age (18). With regard to thermoregulatory mechanisms, when testosterone and estradiol are concomitantly increased, the presence of testosterone may complicate interpretation of estradiol effects in the PO/AH. However, these interactions are poorly understood. Both estradiol and testosterone can stimulate the activity of warm-sensitive neurons in the rat brain (48,60). This would suggest that concomitant administration of estrogens and testosterone in women should induce more effective heat loss mechanisms, lower body temperature and improve thermoregulatory responses to heat stress. However, contrary to these expectations, testosterone exposure *delays* sweating onset, compromises maximum sweating and is associated with increased T_c during exercise in young women (Fig. 5) (66).

Mechanisms for Central Sex Steroid Effects

Changes in thermoregulation associated with sex hormones are generally attributed to direct effects on central neurons controlling peripheral responses to changes in temperature. Sex steroids appear unlikely to act via secondary mediators, such as cytokines (56) or heat shock proteins (7) to alter

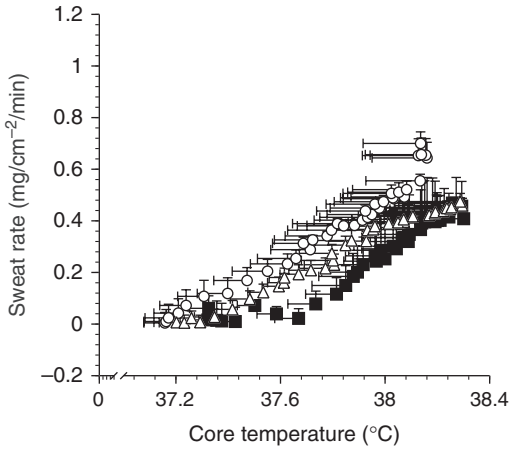

Figure 5 Sweating rate as a function of core temperature in women under three different hormone conditions. Women were studied while taking GnRH antagonist with estradiol (E_2) (open circles), and while taking GnRH (open triangles) antagonist with estradiol + testosterone ($E_2 + T$) (solid squares). Data are expressed as mean ± SEM. (66) *With Permission*.

thermoregulation. Neither IL-1β nor IL-6 is elevated during combined oral contraceptive administration despite large increases in T_c (56). Temperature increases with oral contraceptive administration occurred despite prostaglandin inhibition with ibuprofen (9) and estradiol administration had no effect on heat shock proteins in young women (7). Although the effects of sex hormones on temperature regulation have been recognized for some time, the individual effects of these hormones, and the mechanisms for these effects are still under investigation.

A recent study from Mittelman-Smith and colleagues (45) provides new evidence regarding the potential role of a specific sub-population of central neurons in the influence of estrogens on thermoregulatory responses. These authors evaluated a group of neurons in the arcuate nucleus known to be involved in the regulation of GnRH secretion, a specific subpopulation expressing the ER-alpha receptor. These receptors are known as KNDy neurons because they also express kisspeptin, neurokinin B, neurokinin-3 receptor, and dynorphin. Withdrawal of estrogen caused morphological changes in these neurons, and their proximity to, and interaction with, nuclei involved in thermoregulation (such as the median preoptic nucleus), led the investigators to hypothesize that these KNDy neurons may have a role in thermoregulatory changes seen at menopause (particularly vasomotor symptoms).

They, therefore, conducted specific ablations of KNDy neurons in rats with and without ovariectomy to remove endogenous estrogen during injections of a toxin selective for neurokinin-3 expressing neurons into the arcuate nucleus. They found KNDy neuron ablation not only lowered tail-skin temperature, but also prevented the estradiol-mediated reduction in the tail-skin temperature during exposure to heat (in the light phase). These data indicated an inherent role in thermoregulation for these KNDy neurons and suggested that KNDy neurons are important in the pathways by which estrogen alters thermoregulation. To the extent that neuronal studies in rats can be extended to humans, this suggests that these KNDy arcuate neurons may have a role in estrogen effects on thermoregulation and in the changes in thermoregulation associated with perimenopause and menopause.

Ovarian Hormone Effects on Local Thermal Control of Skin Blood Flow

Local mechanisms in skin blood flow control are those mechanisms which are localized in or around the blood vessels and do not require reflex (i.e., central neuronal) pathways to elicit their effects. In human thermoregulation, any given environment results in some combination of local and reflex thermal influences on the skin circulation. Estradiol and progesterone influence such mechanisms in the local thermal control of skin blood flow (i.e., local heating and cooling) (9, 10) although the mechanisms for these effects remains poorly understood.

Local warming-induced vasodilation of the skin is augmented during the high hormone phase of oral contraceptive hormones compared to the placebo phase; there is no effect on local cooling-induced vasoconstriction (12). The effect on local thermal vasodilation was thought to be due primarily to the influence of estradiol to promote vasodilation via nitric oxide, which is the primary mediator of its vasodilator influence (32) However, complicating interpretation of data such as these are the opposing effects that estradiol and progestogens can have on blood flow regulation. In some physiological systems, progesterone has a vasoconstrictor effect and/or attenuates estradiol-mediated vasodilator effects (19, 35). To date, there have been no studies examining the separate effects of estradiol, progestogens, or testosterone on skin blood flow responses during local heating.

Sex Differences in Thermoregulation

Differences between the sexes in thermoregulatory function have been of interest to physiologists for decades. When used in the context of global health affairs, "sex" refers to the biological and physiological characteristics that define males and females, while "gender" refers to the socially constructed roles, behaviors, activities, and attributes that a given society considers appropriate for men and women (73). "Gender" generally refers to how a person thinks of him or herself regardless of his/her biological sex. This is an important distinction because sex describes biological characteristics inherent in humans or animals, while gender describes characteristics of humans as a function of the society in which they live. Sexual characteristics (genitalia, menstruation, and childbirth) are similar across societies but gender characteristics (income disparities, medical access and treatment, and social customs) vary greatly. Both sex and gender have

important influences on human health. For example, estrogens may protect young women from cardiovascular disease, while gender-related inadequate medical care to young women can have the opposite effect on cardiovascular health.

In the context of the present discussion, historical data suggesting increased cardiovascular or thermal strain in women was for the most part to be explained by group differences in body size, fitness, and/or acclimation status between the men and women studied (13) and environmental conditions, such as relative humidity (13, 59).

However, a few recent, well-controlled studies have carefully documented differences in sweating responsiveness between men and women when exercising at very high thermal and metabolic loads (22, 25, 26). Gagnon and Kenny (25) quantitatively evaluated sex differences in thermoregulatory responses to exercise-heat stress while attempting to control for factors such as body size and fitness which may have confounded previous studies. They compared sweating and skin blood flow responses of men and women exercising at the same overall requirement for heat loss (25). At the highest exercise intensity, sweating responses to the identical exercise-heat stress were lower in women compared to men, both when measured as overall evaporative heat loss (measured in a whole-body direct calorimeter) and specifically due to differences in sweat gland output. The sex differences were not consistently shown at the lower exercise intensities. Taken together, this series of studies suggests that there may be differences in sweat output between men and women, but that these differences are only evident at very high exercise intensities combined with very high requirements for heat loss (high ambient temperatures). Finally, there were no differences in skin blood flow responses between the sexes.

The same group then evaluated whether sex differences exist in peripheral mechanisms contributing to sweating in humans (22). They used increasing concentrations of intradermal microdialysis infusions of acetylcholine and methacholine and evaluated sweating responses in men and women. Although the EC50 was similar between the sexes, women had lower maximal sweating responses to the pharmacological stimulation, indicating that peripheral mechanisms contribute to lower "maximal" sweating that was observed in women in the prior study. In particular, cholinergic receptor responsiveness and/or intracellular signaling appear to contribute to the lower maximal sweating in women. These observations during microdialysis were consistent with those seen during whole body heating, where a decreased sensitivity (slope, responsiveness) of sweating as a function of core temperature was seen in women compared to men (25) In contrast, cutaneous vasodilator responses were not different between men and women, both during pharmacological (microdialysis) trials and during whole body passive heat stress. Gagnon and Kenny have recently conducted an excellent review of mechanisms potentially contributing to differences in thermoeffector responses between men and women during exercise in the heat (24).

Overall Summary and Conclusions

The current state of the literature indicates that, overall, men and women do not exhibit major differences in physiological thermoregulatory responses to exercise and/or body heating. In spite of these similarities, it is clear that reproductive hormones have important influences on mechanisms of thermoregulation. Estrogen promotes heat dissipation via peripheral vascular effects favoring vasodilation, as well as central neural thermoregulatory effects promoting more efficient cutaneous vasodilator and sweating responses. Progestogens may have the opposite effect: the combination of progesterone and estrogen favors heat conservation and/or increased body temperature (such as that seen in the mid-luteal phase of the menstrual cycle). Like estrogen, the influences of progestogens may also include both CNS and peripheral influences. Testosterone has influences on central neural control of thermoregulation, and may interact with estrogen in overall thermoregulation; however, both central and peripheral thermoregulatory influences of this hormone await further clarification. From a practical perspective, it is relevant that the reproductive hormone influences discussed here are unlikely to result in substantial sex differences in core temperature during many commonly performed activities (mild to moderate intensity exercise) in healthy people. Nonetheless, the significant, quantitative effects of reproductive hormones on thermoregulatory responses are important for a comprehensive understanding of the physiology, pathophysiology, and clinical treatment of women across the lifespan.

Because of the ubiquitous nature of reproductive hormones (and the common pharmacological use of agonists and antagonists of their receptors), it is of high clinical and translational importance to continue to investigate the mechanisms by which these hormones affect thermoregulation. In an era where we (as a biomedical community) are encouraging all people to exercise and be as physically active as possible, it is important that we understand how each person's physiological status affects his/her ability to regulate body temperature and avoid excessive hyperthermia and risk of heat illness.

The views, opinions, and/or findings contained in this article are those of the authors and should not be construed as an official Department of the Army position, or decision, unless so designated by other official documentation. Approved for public release; distribution unlimited.

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